

# **Temporal Dynamics of the Defense Cascade**

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### **ABSTRACT**

Understanding physiological responses to threat can inform therapeutic interventions for phobias, anxieties, and PTSD. The defense cascade is reviewed as a theoretical model that predicts behavioral and physiological responses to threats. Nineteen undergraduates (five male), average age 19.4 experienced a novel virtual reality (VR) threat scenario while their physiology was measured. The Subjective Units of Distress Scale (SUDS) was used as a self-report indicator of distress in the research setting. Averaged SUDS reports suggested that the VR stimulus was experienced as threatening for most participants, but their autonomic response patterns did not fit those predicted by the defense cascade. Participants who had scored high on adaptive response questionnaires tended to show uncoupled ANS activation during baseline, but varied across the stimulus condition. Nearly all participants showed either coactivation or reciprocal activation during the stimulus period except those reporting the most dissociative trauma experiences, who mostly showed uncoupled ANS activation.

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### GENERAL AUDIENCE ABSTRACT

The more we understand about how people's bodies and their energies act when they feel threatened, the better we can find help for folks who struggle with anxiety, trauma or other challenging conditions. This research uses a theoretical model called the *defense cascade* to explore how people respond mentally and physically to threatening situations. Nineteen undergraduates went through a virtual reality (VR) experience that was designed to feel threatening while their body and its energy systems were measured. A scale was introduced called the Subjective Units of Distress Scale (SUDS) and was used to help the researchers understand how distressed people felt while they were in the VR experience. Averaged SUDS reports suggested that the VR stimulus was experienced as threatening for most participants, but their body response patterns did not fit those predicted by the defense cascade. Participants whose questionnaire responses suggested they were not anxiety-prone or traumatized, tended to show bodily activation that uncoupled their two autonomic bodily systems during a baseline period before the threatening stimulus. However, their autonomic responses during the stimulus period varied. Nearly all participants showed either both autonomic systems acting together or only one system acting in a mutually exclusive way to the other system during the stimulus period. This was the case for most participants except those reporting the most trauma involving dissociative experiences. This latter group mostly showed uncoupled autonomic bodily patterns.

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## Temporal Dynamics of the Defense Cascade

Occasionally the monotony of daily stress may be interrupted by an immediate danger that awakens the senses and energizes the body for defense. In these moments, one hopes that the toll of those daily stresses hasn't sapped the body of the survival energies needed to respond to the danger. Where do these survival energies draw from, and how do they play out when dangers escalate? How are they impacted by a lifetime of routine stress? Is there any hope for survival if one has exhausted one's energy reserves?

Research on survival behavior explores such questions, guided by a theoretical model called the *defense cascade* (Lang, Bradley, & Cuthbert, 1997). This model describes the series of "strategies" marshalled to face dangers from escalating threat. These strategies are actually instincts, inherited from ancestors escaping life threats over the millennia. This evolutionary inheritance serves to elevate the survival instincts while turning off the thinking brain during times of even mild stress (Arnsten & Goldman-Rakic, 1998). In situations of real danger, survival instincts confer the benefit of near-instant reaction time, providing advantages over a carefully thought-out plan. Understanding these responses can help improve therapeutic interventions for everything from "routine" stress and anxiety to phobias and posttraumatic stress disorder (PTSD). Beyond clinical settings, people who understand the survival playbook embedded in their physiology can gain greater self-awareness and self-acceptance after navigating a trauma or anxiety-provoking situation.

This paper reports the results of a study conducted to examine physiological patterns of response to a threatening stimulus, how those patterns map to the defense cascade, and how they are influenced by certain pathologies, such as anxiety and posttraumatic stress disorder (PTSD). Virtual reality (VR) was utilized to develop a stimulus that simulated increasing threat while

physiological and self-report variables were tracked. This research was originally conceptualized as an empirical study with a large enough sample size to allow inferential statistics. However, in the wake of the COVID-19 pandemic and subsequent research shut-down, the research was halted after running only 19 participants. As a result, the research has been repurposed as a proof of concept study.

### **Stages of Defense**

Defensive strategies start with *freezing* to the threat, followed by a *flight* attempt, and then direct engagement through *fight* (Lang et al., 1997; Roelofs, Hageraars & Stins, 2010, Cannon, 1932). If all of these strategies fail, more extreme behaviors will involve immobilizing to the threat in a *fright* response or even *flagging* and *fainting* (Bracha, 2005; Schauer & Elbert, 2010). These extreme immobile responses are sometimes understood as a state of shock, or akin to an opossum playing dead. As in the opossum example, it becomes clear that even inactivity can sometimes be the most effective survival strategy.

This dynamic range of options equips someone under threat to address a wide array of environmental challenges, from mild stress to dire life threats. When a person draws on these instincts to address a threat, they are demonstrating human adaptability. However, repeated exposure to such challenges can cause those pre-potent responses to become habit-forming. Previous threats that result in a particular defensive strategy can set the conditions for moving quickly to this same stage in future threats (Adenauer, Catani, Keil, Aichinger, & Neuner, 2009).

### **Defense and Psychopathology**

Disorders can result from patterns of defense that become habitual: phobias invoke flight responses that are disproportionate to the actual threat, but repeated flight/avoidance nevertheless sets the conditions for habitual response patterns (Lovibond, Mitchell, Minard, Brady, &



Menzies; 2009). Anxiety and worry marshal survival energies for situations that do not actually allow these energies to be expressed (McTeague & Lang; 2012). When survival instincts are utilized, they have the capacity to either sensitize or recalibrate the system to future environmental challenges. Experiencing trauma can be especially powerful experience in this regard. For some, experiencing trauma can sensitize them to threat, amplifying their perception of danger, resulting in PTSD symptoms of avoidance (flight) and arousal/reactivity (flight or fight) via a process described as emotional undermodulation (Lanius, Brand, Vermetten, Frewen, & Spiegel, 2012). For others, experiencing trauma can recalibrate their danger “radar”, leaving them numb to the fluctuations of daily existence. These people can show dissociative symptoms through a mechanism called emotional overmodulation (Lanius et al., 2012).

The common thread among these states is a disconnection between the actual environment and the expression of the energies it requires. A person may be moving through the flight or fight portion of the defense cascade in response to a situation that does not require such defensive behavior; e.g., exhibiting phobias, anxiety disorders, or PTSD with avoidance or arousal and reactivity. Or, one might not sense a nuanced danger if their previous trauma recalibrated their danger radar to be sensitive to only extreme dangers – this person may not be in the defense cascade when the situation actually requires them to be. Finally, one may move through the first stages of the defense cascade and respond with the extreme danger options of immobilization or numbness, when the situation actually calls for escape energies. PTSD with dissociative features can lead to either of the two latter situations (Lanius et al., 2012).

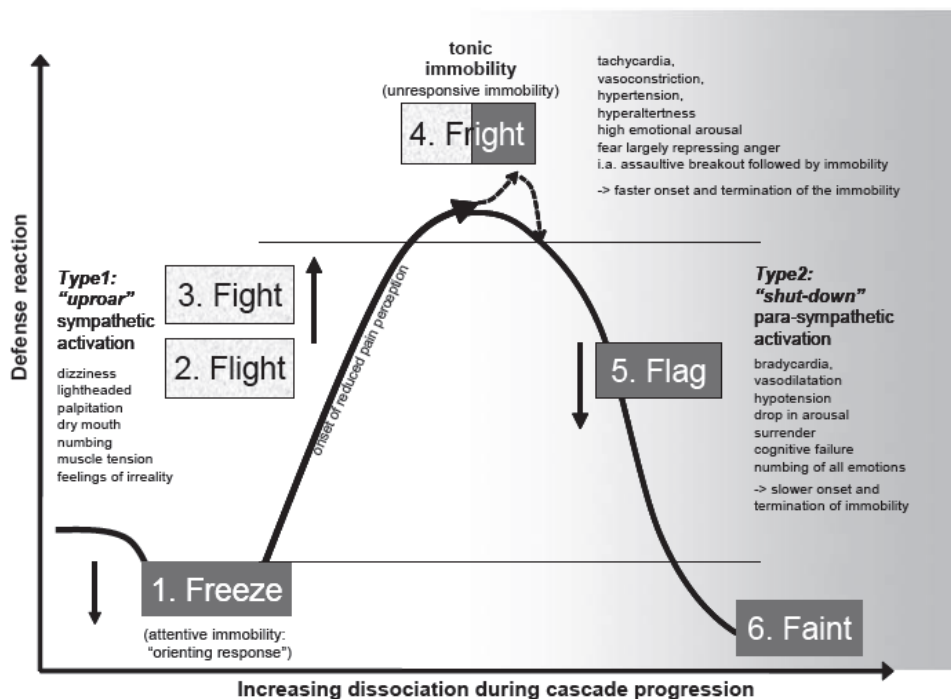
Thus, an adaptive response to threat requires an accurate match between the environmental demands of the actual situation and the energies used to address this situation. Preexisting phobias, anxiety disorders and PTSD can cause survival responses to differ than in

individuals without such conditions (Hamm; 2020). I propose that a well-designed study into the physiology of survival behavior should ensure that these conditions are made explicit.

## Defense and the Autonomic Nervous System

For people with or without disorders, threat can subject the mind and body to a roller coaster of responses. In fact, the six stages of the defense cascade are typically portrayed in a hypothetical figure reminiscent of a roller coaster (Schauer & Elbert, 2010; see Figure 1).

*Figure 1: The six stages of the defense cascade*



*Reprinted from "Dissociation following traumatic stress," by M. Schauer and T. Elbert, 2015, Journal of Psychology, 218(2), p. 111. Reprinted with permission.*

Along with its helpful visual, this figure also establishes a framework for understanding the sequence of physiological changes required to navigate the defense cascade. To understand these physiological changes, it is helpful to start with the traditional view of the autonomic nervous system (ANS). This view describes the parasympathetic nervous system (PNS) as being

predominantly active during times of safety, as it regulates restorative and reproductive activities of the body. Porges (2007) describes this response of the vagus nerve as equipping the Social Engagement System. This situation is not shown in Figure 1, since the figure illustrates a situation of danger.

ANS dynamics in defensive situations starts with the sympathetic nervous system (SNS) is characterized as the “fight or flight” system (Cannon, 1932), regulating energy expenditures for both defense and offense behavior such as hunting and predation. The defensive SNS state is illustrated in the uphill portion of the “roller coaster” in Figure 1.

Extending beyond the introductory-level understanding of the ANS, the PNS is also predominantly active during times of extreme danger, and is responsible for immobilization when flight or fight options have been exhausted (Bracha, 2005; Kleinknecht, 1987; Öst, 1984; Porges, 2007; Schauer & Elbert, 2010). This can be seen primarily in the downward slope of the “roller coaster” in Figure 1.

Figure 1 therefore lays out the physiology behind the uphill SNS climb and the downhill PNS slide. These two scenarios reflect reciprocal activation in the ANS – the situation where activation of one branch dominates the other branch. This reciprocal mode allows the ANS to maximize the range of physiological response (Berntson, 2019), with PNS dominance strongly decreasing heart rate or SNS dominance strongly increasing heart rate to meet environmental challenges. Reciprocal ANS activation is important in the extreme survival situations characterized by the uphill and downhill portions of the defense cascade, as they illustrate times when rapid changes in heart rate are needed to support defensive response.

While defensive physiology often shows extreme responses, it is important to recognize that fine-grained autonomic activation patterns are far more common in a typical day. Jänig &

Häbler (2000) detailed the ANS nuances needed for more homeostatic functions. Their research is a reminder that it is rare for the SNS and PNS to function in the all-or-none manner portrayed in Figure 1. In addition, the ANS regulates more than just the heart, with different organs being targeted for different homeostatic functions. Understanding these maintenance functions of the ANS brings a broader perspective to an investigation into defensive physiology, since research participants presumably will not be in defensive mode throughout the experiment.

Even in defensive situations, ANS patterns can take on more than just the extreme reciprocal activation shown in the uphill and downhill portions of Figure 1. Connecting those portions of the graph are the apparent slope changes, where PNS dominance transitions to SNS dominance and vice versa. Lang, Bradley, & Cuthbert (1997) suggested that the inflection point from PNS to SNS dominance in the freeze state involves “increasing stimulation from both sides of the ANS” (p. 124), a finding corroborated by subsequent research (Roelofs, Hagenaaers & Stins, 2010). This transition point occurs during the first stage in the defense cascade, in which the person freezes upon detecting initial possible danger. Just prior to detecting the threat, the person would likely be in a PNS-dominated state of safety (Porges, 2007). Engagement of the SNS would be necessary to prime the system and prepare it for active defense if needed. But simultaneously, it is necessary to quiet the body so that attention can be directed to the potential threat for further assessment – the PNS immobilizes the body for this response.. Freezing is the preferred initial response because it preserves survival energies if the detected potential threat is assessed not to be a threat. Further, even if there is a real threat, freezing can often avoid predator detection – the most efficient defense of all.

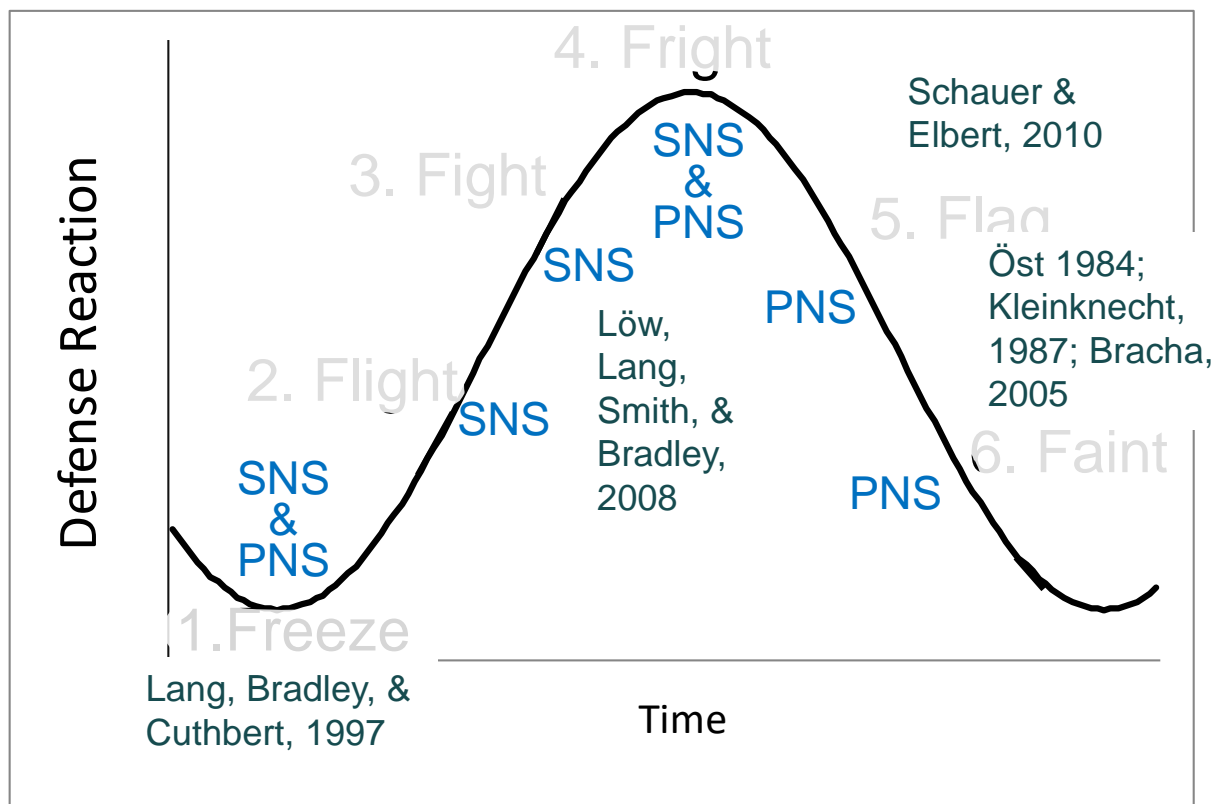
Löw, Lang, Smith, & Bradley, 2008 investigated the inflection point later in the defense cascade, in which flight and fight options have been exhausted and the organism begins to switch

from SNS dominance to a PNS shut-down state. This stage “was characterized by coactivation of the sympathetic and parasympathetic systems...” (p. 872). Immobilization in this stage is invoked when the mobile defenses have failed. Survival energies have either been exhausted or rendered ineffective. Immobilization conserves whatever energy is left, minimizes blood loss from potential injuries by dropping heart rate and blood pressure, while also deflating the aggressor’s killer instincts: “an inert body inspires little interest,” (Stouffer, 1993).

### Mapping Defense with Physiology

Each of the aforementioned findings can be connected to form a holistic picture of the physiological dynamics proposed to support the defense cascade, shown in Figure 2. The uphill SNS dominance and the downhill PNS dominance are joined by SNS-PNS coactivation at the inflection points.

Figure 2: The six stages of the defense cascade with ANS correlates suggested by indicated studies.



This possibility of ANS coactivation illustrates the limits of the stereotypical gas pedal/brake pedal analogies often used to characterize the SNS/PNS relationship. Such an analogy suggests that the reciprocal mode of ANS activation is the only mode available. In fact, ANS coactivation is not only possible; literature suggests that it is critical in times where maximum autonomic flexibility is required. (Berntson et al, 1993; Berntson, 2019). Defense cascade inflection points provide a perfect example of when maximum flexibility is needed. The organism must be ready to choose either extreme: instantaneous acceleration or deceleration.

A third possible mode of ANS activation involves uncoupled activation, in which activation of one branch happens irrespective of activation of the other branch. Berntson (2019) suggests that this mode is simply a middle ground between reciprocal and coactive ANS modes. However, I propose in this study that the ANS uncoupled mode is invoked in times when neither extreme flexibility nor an extreme range of responses is needed. Rather, I propose that uncoupled activation is seen when fine-tuned ANS coupling or reciprocity is not required.

Building on the concepts of ANS coupling and reciprocity, the term *cardiac autonomic balance (CAB)* has been introduced to characterize the degree to which the PNS and the SNS are working reciprocally (Berntson, Norman, Hawkley, & Cacioppo, 2008). In contrast, *cardiac autonomic regulation (CAR)* describes the degree to which the two systems are coactive. Both measures are used in combination to chart ANS variability within the construct of *autonomic space* (Berntson, Cacioppo, & Quigley, 1991, Berntson et al., 1994). In Berntson et al. 2008, four-minute resting averages of measures indexing SNS and PNS revealed CAR to be the better predictor of physical health conditions, with more CAR revealing better health.

## Methodology

Most studies of the defense cascade used one of two methodologies for estimating ANS activation. One method was to target a specific stage with a stimulus and then average ANS measures over the duration of the study (Roelofs et al., 2010). The other method examined dynamic changes in ANS measures, but did so using simplified measures such as heart rate (Lang et al., 1997; Löw et al., 2008). The heart is dually-innervated by both the SNS and the PNS, so a “simple” decrease in heart rate is not a guarantee that PNS has increased. Returning to the car analogy, when one is preparing to exit the freeway, two options are available: pressing the brake pedal is one option; removing the foot from the gas pedal is a second option. Typically, the driver will start by removing the foot from the gas pedal, and then later fine tune the stop using the brake. Similarly, the PNS and SNS can each individually reduce the speed of the heart or they can do so working together, depending on the needs of the moment.

Heart rate accelerations are somewhat harder to explain with the gas/brake analogy, because unlike with cars, the ANS always has a tonic level of SNS and PNS activation in the system, even when at rest, although PNS influence prevails during such quiet states (Porges, 2007). This is important, because the SNS adrenergic neurotransmitter pathway affects heart rate more slowly than the vagal cholinergic pathway (Saul, 1990). As a result, a rapid and large heart rate acceleration is usually accomplished by removing the PNS brake and allowing the tonic SNS to prevail. Such action allows quicker heart rate acceleration than does applying the slower-acting SNS gas pedal.

Because the heart is dually innervated by both branches of the ANS, a more sophisticated methodology than heart rate alone is needed to index them. Estimating PNS control of the heart begins with an electrocardiograph (ECG) which provides a time series of the electrical signal

generated by the heart, measured in millivolts. The peak voltage for each heartbeat (the “R-wave”) provides a marker from which to measure the time between each heartbeat, called the inter-beat interval (IBI). The variability in these IBIs can reveal a wealth of information about the physiological state. Research has shown that heart rate variability in the high frequency spectrum (HF-HRV; high frequency defined as 0.15 to 0.40 Hz) can index vagal control of the heart, which is under PNS control (Thayer et al., 2012).

Measures of heart rate variability fall into one of two categories: frequency-domain values and time domain values (Shaffer & Ginsberg, 2017). Frequency-domain measures focus on the amount of power in certain frequency bands. The most common methods for calculating this power are the Fast Fourier Transformation (FFT) and autoregressive (AR) modeling. Time domain values calculate the amount of variability in the time between each heart beat known as the inter-beat interval (IBI). One popular time domain measure of heart rate variability calculates the root mean square of the successive differences between heart beats (RMSSD).

Respiratory sinus arrhythmia (RSA) refers to the variations in heart rate that are tied to the breathing cycle. Heart rate speeds up during inhalation and slows down during exhalation. This respiration-related fluctuation in heart rate rhythm reflects vagal control of the heart. Therefore, quantifying the degree of RSA is another method for indexing PNS (Katona & Jih; 1975). There are several competing methods for quantifying RSA, but one of the most common is the peak-to-trough method, which takes the longest (IBI) during exhalation and subtracts the shortest IBI during the inhalation (Grossman, Van Beek, & Wientjes; 1990).

Estimating SNS control of the heart also begins with the ECG but adds a second cardiac measure called impedance cardiography (ICG), which is a noninvasive measure of heart contractility. Taking the derivative of the raw impedance signal provides distinctive waveforms



that correspond to cardiac events. One such form, the B point corresponds with the opening of the aortic valve. Returning to the ECG signal, taking the Q-wave from the onset of the QRS complex and subtracting the B point from the ICG signal provides a calculation of cardiac pre-ejection period (PEP; Sherwood et al., 1990). PEP is the period of time when the blood is in the left ventricle of the heart before it is ejected into the aorta. It begins with electrical stimulation of the heart and ends with the ejection from the left ventricle, and is thus a measure of electromechanical activation of the heart. PEP is an inverse index of SNS in that a decrease in PEP reflects an increase in SNS. Electrodermal activity (EDA) indexes SNS control of the sweat glands and therefore can be useful in indexing SNS influence beyond the heart.

Even the most sophisticated physiological measures cannot alone ensure that a study of the defense cascade is actually inducing research participants into a defensive response. A manipulation check is essential in determining whether the physiology recorded is reflecting a physiology under threat. A self-report scale from clinical psychology can be brought to bear, called the Subjective Units of Distress Scale (SUDS; Wolpe & Lazarus; 1966). One version of this scale uses a 0 to 10 scale, where 0 is no distress at all and 10 is the most extreme distress one has ever had (Shapiro, 1995). Beyond its utility as a manipulation check, I propose that SUDS can also provide clues about where in the defense cascade a person is. Because it is a strictly theoretical model, the defense cascade poses challenges to the researcher hoping to index one stage to the exclusion of the others. Although it is used in therapeutic settings, SUDS is not typically seen as a therapeutic intervention in and of itself. However, I hypothesized that repeated use of this tool can confer therapeutic benefits of increased emotional and/or interoceptive awareness.

The current study was conducted to develop a more precise mapping of physiological changes under threat, using non-invasive psychophysiological measures of ANS activity and SUDS as for self-reported subjective experience of distress. Participants were exposed to a threat scenario in virtual reality (VR) while physiological variables were acquired. Simultaneously, participants gave SUDS ratings throughout the threat experience. An additional manipulation check involved video recording participants throughout the experience, a tool used for both behavioral analysis and to cross reference apparently aberrant physiological data.

Employing a Baseline-Stimulus-Recovery framework, this study was designed to allow traditional epoch averaging and comparisons of physiological variables within epochs, while adding a more in-depth qualitative analysis of the time series data. Further, SUDS was used as a psychological variable against which to analyze physiological variables. Behavioral measures were added, including mobility measures, researcher behavioral scoring, and video recordings of participant behavior throughout the study.

I hypothesized that participants during baseline would show physiological indicators of parasympathetic influence on the heart, reflecting a relatively less-threatened state from the stimulus period. I further hypothesized that participants entering the threat scenario would show PNS-SNS coactivation, and that measures of increasing sympathetic cardiac influence would be detected as participants moved into and through stages 1, 2 and 3 of the defense cascade, coupled with decreasing PNS cardiac influence. If participants moved as far as stage 4, fright, I hypothesized that the PNS influence on the heart would show an increase, while SNS influence would continue to have strong coactive influence as well. I further hypothesized that SUDS ratings would be positively correlated with the stage of the defense cascade up to fright. Because

stages 4, 5, and 6 require intense levels of perceived danger, the current study was designed to avoid invoking such stages in respect for ethical considerations.

## Method

### Participants

Undergraduate psychology students from Virginia Polytechnic Institute and State University (Virginia Tech) were recruited using the Sona Experiment Management System (Sona). Two hundred fifty-two participants completed an online screening to ensure they met inclusion criteria, which included being over 18 years old, no history of heart conditions, high blood pressure, neurological disorders or seizures, no medications affecting the cardiovascular system and people who were not heavy smokers as defined by using tobacco products less than 6 days a week. One hundred sixty-six participants met these inclusion criteria, of which 86 were invited to the lab for the study. Nineteen completed the study.<sup>1</sup> Approval for the study was obtained from the Virginia Tech Institutional Review Board, and all participants gave informed consent for their participation.

The sample consisted of 5 male and 14 female participants, with a mean of 19.4 years (range: 18 - 22 years). Seventy-four percent were White, 11% Asian, 5% Hispanic/Latino, 5% African-American, and 5% multi-racial.

### Stimuli and Procedure

A novel virtual reality (VR) stimulus was developed for this study. This VR development involved an interdisciplinary collaboration as follows: a Virginia Tech cinema student with specialty in horror designed the screenplay, the stimulus video, inspired by the classic inkblot projective test (Rorschach, 1921/1942), and the cinematic sound score. Three

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<sup>1</sup> A sample size of 85 was targeted to obtain a target correlation of 0.3, with  $\alpha=0.05$  and  $\beta=0.20$  based on a power analysis from the G\*Power applet (Faul et al., 2007) However, this number was not attained due to research restrictions imposed in response to the COVID-19 pandemic.

Virginia Tech visual arts students jointly developed and animated an inkblot-inspired “monster” who acted as the principal threat in the VR experience. This team also created the entire VR setting and all other artistic assets. Two computer science students at Virginia Tech and two students with dual specialties in visual arts and computer science assembled these aforementioned components into a complete VR experience using Unity software, adding in elements such as the two baseline videos, onscreen SUDS requests, and additional sound effects. This stimulus was therefore developed to the explicit specifications to ensure the experience invoked the desired sequence of responses indexing the different stages of the defense cascade.

Participants were recruited via the Sona system and through advertisement flyers posted in the psychology department. Interested participants completed an online pre-screen along with a battery of questionnaires. The pre-screen consisted of the Mind-Body Lab Health History screening questionnaire (HHQ; See Appendix A) to assess for participant alignment with target inclusion criteria as described above. These participants also completed questionnaires assessing their psychological metrics as follows: the Penn State Worry Questionnaire (PSWQ; Meyer, Miller, Metzger, & Borkovec, 1990), the Trauma History Screen (THS; Carlson et al., 2011), the Patient Health Questionnaire – Panic Disorder subscale (PHQ-PD; Spitzer, et al., 1994), the Anxiety Sensitivity Index-3 (ASI-3; Taylor, 2007), the Anxiety Control Questionnaire (ACQ; Rapee, Craske, Brown, & Barlow, 1996), the Five Facet Mindfulness Questionnaire (FFMQ; Baer, Smith, Hopkins, Krietemeyer, & Toney, 2006).<sup>2</sup>

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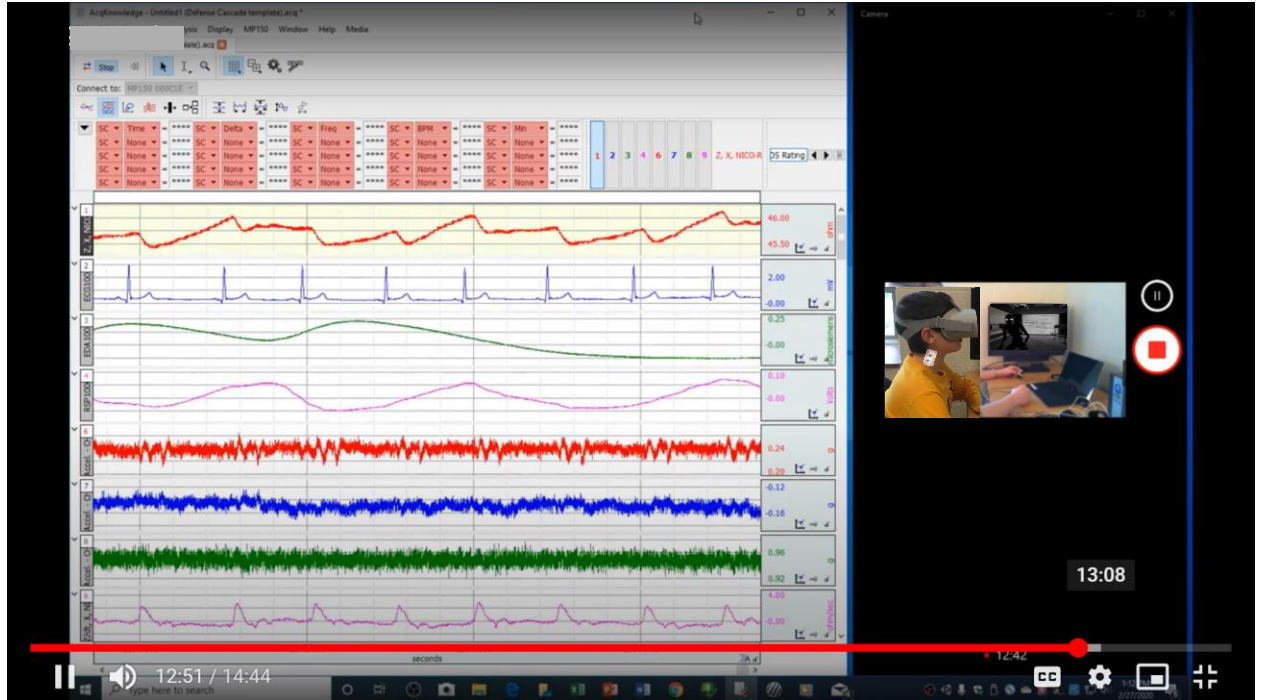
<sup>2</sup> The following two additional questionnaires were completed but not used for this study: the UCLA Loneliness scale, Version 3 (Russell, 1996), and the Big Five Aspects Scale (BFAS; DeYoung, Quilty, & Peterson, 2007).

Participants who passed the HHQ pre-screen were invited to participate in the lab study and instructed to abstain from the following: alcohol for 12 hours prior to the study, caffeine and other non-prescription drugs for 6 hours, and eating and exercise for 2 hours. When participants arrived for the study, they reviewed the explanation of study and signed informed consent. Participants then completed a short survey assessing their compliance with the substance abstinence. All participants reported compliance. Participants were then connected to the physiological equipment as described below.

Once connected to the physiological equipment, participants completed an additional series of questionnaires including the Multidimensional Assessment of Interoceptive Awareness (MAIA; Mehling et al., 2012), the PTSD checklist (PCL; Weathers, Litz, Herman, Huska, & Keane, 1993), the State-Trait Anxiety Inventory (STAI; Spielberger, 2010), and the Fear Survey Schedule (FSS III; Wolpe, Lang, & Ther, 1964). Participants were then briefly instructed on the use of the Subjective Units of Distress Scale (SUDS).

The virtual reality (VR) headset was then placed on the participant and adjusted to ensure optimal viewing. A webcam began video recording the participant from a side angle while in the background, a monitor continuously screencasted the VR imagery seen by the participant. Concurrently, real-time physiological data scrolled on the primary research computer. All details were captured in a live screen recording (see Figure 3 for an illustration).

*Figure 3: Illustration of Real-Time Data Capture of Physiology, Behavior, and Stimulus*



Physiological recording continued throughout the subsequent participant experience. The virtual reality experience was as follows:

Participants appear to themselves to be seated in a hard-backed wooden chair in a basement-like room reminiscent of a physiological lab from the 1960s. Old fashioned physiological equipment sits on countertops on both sides of the participant, and a medical cart sits at a one o'clock angle. Overhead is a movie projector and straight in front is a large movie screen, partially obscured by the medical cart. (See Figure 4 for illustration.)

*Figure 4: Virtual Reality Setting*



When participants indicated that they were ready to begin, a researcher started the scene in motion, which included a series of videos projected on the VR movie screen and culminating in a three dimensional VR experience. The first video was a 3-minute instructional video in the use of the SUDS rating system. At the conclusion of this video, the video screen went black for 10 seconds while participants were given an on-screen prompt to state aloud their first SUDS rating. Subsequent on-screen SUDS prompts were given at approximate one minute intervals throughout the remainder of the experiment.

After the ten second black-out pause for the first SUDS rating, the video then projected a roughly four-minute ‘vanilla’ baseline video depicting marine life:

<https://www.youtube.com/watch?v=O9AaqmMv4HU>. This video was designed to capture participants at rest without confounds of boredom from a complete absence of stimuli (Piferi, Kline, Younger, & Lawler, 2000).

As the baseline video was fading to black, the lights in the VR space flickered and went out, accompanied by a sudden loud electrical buzzing sound effect. This marked the end of the



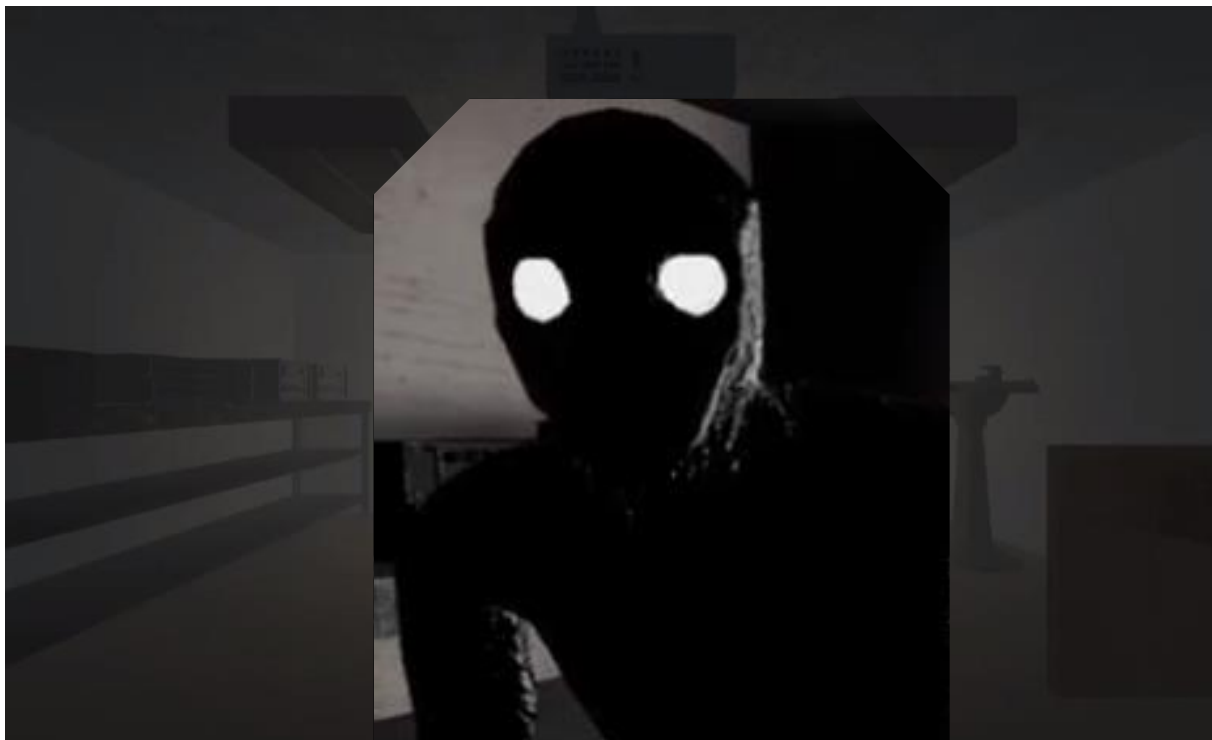
baseline period and the simultaneous beginning of the stimulus period. Following this stimulus, no new stimuli were presented in the VR space for 90 seconds. The VR space during this time remained darkened to simulate the lights out in a room with only faint ambient lighting to keep the participants present within the VR experience. Figure 5 illustrates this condition.

*Figure 5: Darkened Virtual Reality Room Displayed for First 90 Seconds of Stimulus*



After the 90 second pause, a five minute threat video was shown. The video, developed by a Virginia Tech cinema student with a specialty in the horror genre, was reminiscent of the Rorschach Inkblot Test (Rorschach, 1921/1942). Inkblots were presented to the backdrop of a cinematic sound score that included the occasional vague echoing voices, and a musical score that increased in tempo throughout the experience. Just over two minutes into the video, the image of an inkblot-inspired monster with glowing eyes appeared suddenly, as if in face of the participant. Figure 6 shows is an illustration of this stimulus.

*Figure 6: Illustration of Ink Blot-Inspired Monster (Flashed 2 minutes into Fear Video)*

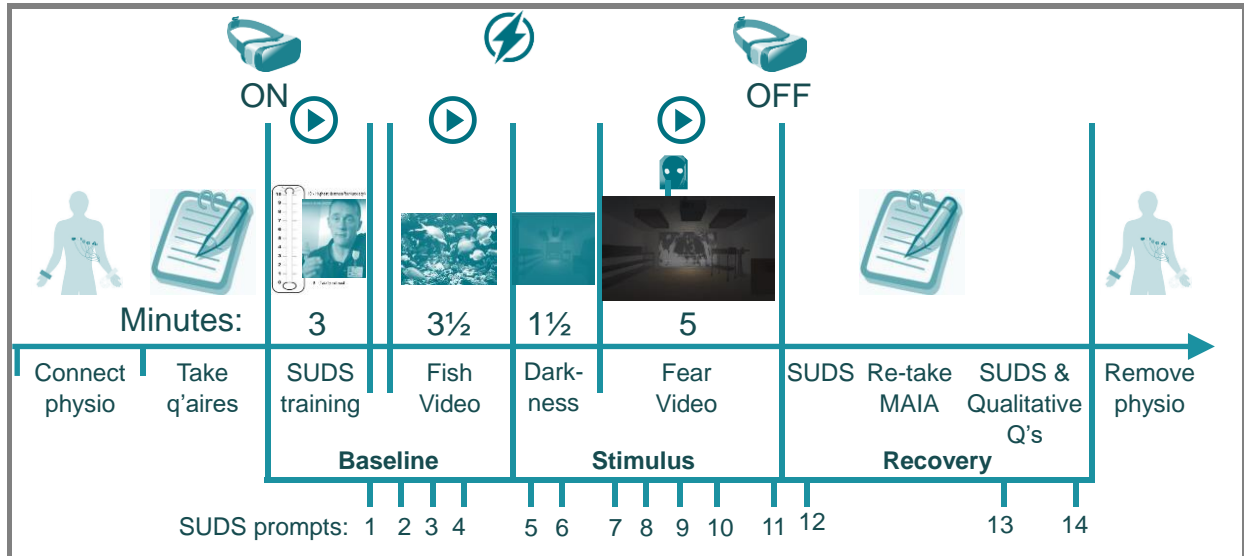


This flash of the monster lasted only one second, and then it disappeared. The video culminated in the emergence of the monster, who appeared to climb out of the VR movie screen and approach the viewer, while grunting and breathing heavily. After lunging for the viewer's neck, the monster continued to stand in immediate proximity, appearing to stare down the viewer with glowing eyes. Twenty seconds later, the participant was prompted for the final SUDS rating of the stimulus period.

After the participant stated this SUDS rating, the researchers announced that it was the end of the VR stimulus and helped the participant remove the VR headset. This transition marked the end of the stimulus and the beginning of the recovery period. Once the headset was removed, the participant was asked for another SUDS rating. Participants then completed the MAIA questionnaire again. This lasted between 2 and 6 minutes. Once complete, participants were asked for another SUDS rating, and then asked questions about their subjective experience.

Then they gave their final SUDS rating. This marked the end of the recovery period. The physiological equipment was then removed and participants were given a debriefing that included information about psychological services available following the study. Figure 7 provides a schematic illustration of the procedure.

Figure 7: Study Procedure



### Data recording and reduction

Behavioral measures were extracted from each video recording. One researcher recorded behavioral observations throughout the baseline and stimulus periods. The video depicting participants from the side was used to assess movements toward the stimulus (approach/fight behaviors) and movements away from the stimulus (avoid/flight behaviors).

Physiological measures were extracted using specific transducers in connection with the Biopac MP150 Data Acquisition System (Biopac Systems, Inc., Goleta, CA). Electrocardiogram (ECG) was acquired at 2000 samples per second using the Biopac ECG amplifier (ECG100C). An ECG Lead II signal was acquired using a 3 electrode system with leads placed on the chest

wall equidistant from the heart. Although this system typically requires connection of a ground lead, in this case the ground was eliminated because a ground was already employed for the EDA as noted below.

Impedance cardiography (ICG) was gathered at 2000 samples per second using the BioNomadix wireless Noninvasive Cardiac Output System (BN-NICO). This involved connecting 8 total electrodes (4 paired electrodes) in a configuration with 2 output leads on the left and right side of the neck, 2 input leads direction below the output leads, 2 input leads on the left and right side of the torso, and 2 output leads directly below the torso input leads.

Electrodermal Activity (EDA) was obtained using the Biopac Electrodermal Activity Amplifier (EDA100C). One EDA electrode was placed on the index finger of the participant's non-dominant hand and the second electrode was placed on the middle finger of the same hand.

Respiration was gathered using the Biopac respiration amplifier (RSP100C) and the BIOPAC transducer TSD-201 respiration belt. The belt was placed on each participant's sternum tightly enough to capture both inspiration and expiration but not so tightly that it impeded participants' breathing. Researchers explicitly explained these parameters to the participants to ensure proper fitting of the respiration belt. Acceleration was gathered using the Biopac Accelerometer (TSD109C3).

In order to prepare data for analysis, a bandpass filter was applied to the raw ECG signal to filter out frequencies below 0.5 Hz and above 35 Hz, which simultaneously flattened the isoelectric line and filtered out high frequency noise between cardiac cycles. The resulting waveform was visually inspected for aberrant cardiac cycles caused by movement artifact – where such signal noise existed, the waveform was manually adjusted to eliminate artificial outliers.

From this cleaned ECG signal, several calculations were possible. First, the Biopac Acqknowledge 4.4.3 software (Goleta, CA) was used to calculate a time series of the interbeat interval (IBI). This IBI signal was then imported to Kubios HRV v2.0 (Tarvainen et al., 2014) where a battery of metrics were calculated, including HF-HRV and RMSSD. For HF-HRV, the power of heart rate variability in the high-frequency spectrum (0.15 to 0.40 Hz) was divided by total heart rate variability power to achieve a percent power value in the high-frequency range. Two different methods were used to extract high-frequency heart rate variability: the Fast Fourier Transform (HF-HRV FFT) and the Auto Regressive technique (HF-HRV AR). Results from both methods are reported in the Results section.

The AcqKnowledge software was then used to calculate a time series of RSA from the respiration data and the IBI data, using the peak-to-trough method as outlined above. In order to calculate PEP, the first derivative of the ICG signal was classified using AcqKnowledge 4.4.3. PEP was then calculated from the classified  $dZ/dt$  signal as described in the Methodology section, and then cleaned ECG signal (Sherwood, 1990) using the R-to-C polynomial model outlined in Lozano et al. (2007). The PEP and the classified  $dZ/dt$  signal were then visually inspected for outliers caused by noise in the sensitive  $dZ/dt$  signal. Outlier PEP values were addressed by eliminating  $dZ/dt$  labels that were erroneously placed on the  $dZ/dt$  signal that was too noisy to be labeled accurately. After thorough  $dZ/dt$  label cleaning, PEP was recalculated and re-inspected.

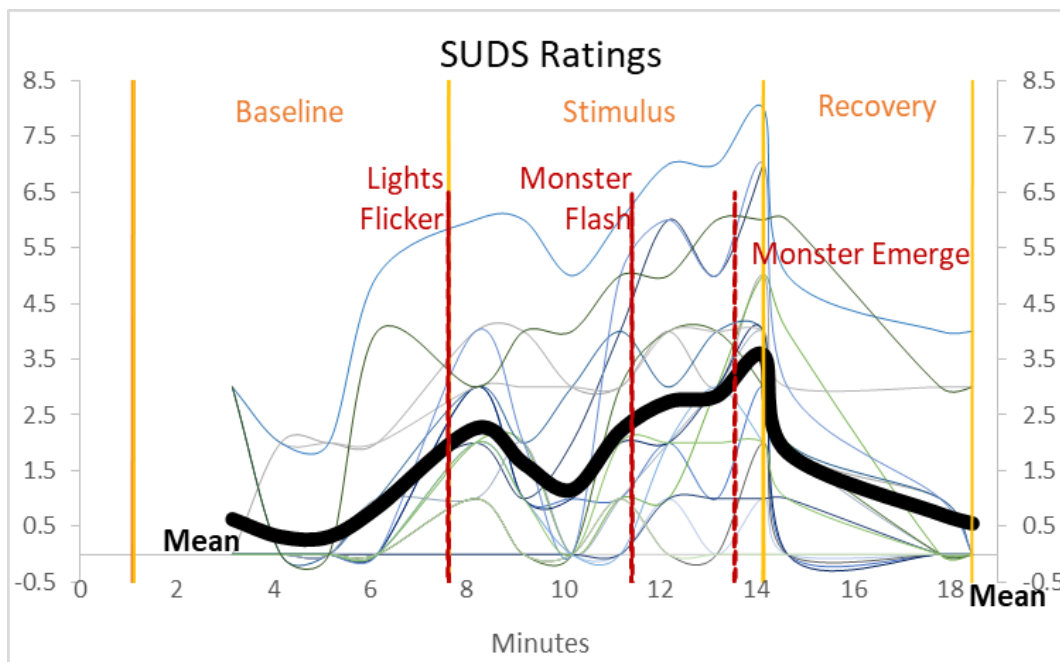
## Results

Analysis began with a manipulation check to determine the extent to which the stimulus had the desired effect. Then results were analyzed in the traditional aggregated approach using epoch averaging. Next a behavioral analysis was conducted to add qualitative color to the quantitative data. A qualitative analysis of the time series data followed and then a correlational analysis was conducted. Each method of analysis is summarized in a separate section below.

### Potency of the Stimulus

Participant ratings on the Subjective Units of Distress Scale (SUDS) indicate that the VR stimulus was effective at inducing a sense of threat for most participants. Averaged across all participants, mean SUDS rating for the baseline period was 0.5 out of 10, ( $SD = 0.92$ , range = 0 to 5), mean SUDS rating for the stimulus was 2.3 ( $SD = 1.72$ , range 0 to 8), and mean SUDS rating for recovery was 1.0 ( $SD = 1.38$ , range 0 to 6). Figure 8 shows a time series of SUDS ratings for all participants and for the mean SUDS rating.

*Figure 8: SUDS Ratings of All Participants (thin lines) and Mean SUDS (bold line)*



Participants were also asked, “Overall, how fearful did this video make you on a scale of 0 to 10?” Mean rating was 3.1 (SD = 1.76, range = 0 to 7).

Participant answers to qualitative questions add subjective color to these data. Participants were asked “How difficult was it to stay engaged with this video?”, “If you had difficulty, can you explain why?” Eight out of the 19 participants indicated that they had difficulty staying engaged with the baseline SUDS and/or the marine video, but none of the participants reported difficulty staying engaged with the stimulus. Several other participants indicated feeling worried that the fish video would produce a fearful stimulus. The advertised title of the study was, “Virtual Reality: Physiology and Subjective Experience under Stress or Threat” and featured two images of frightened-looking people with VR headsets on.

When participants were finally asked, “Do you have any other comments or suggestions about the experience you’d like to add?”, several informative comments were offered. An excerpt of these comments includes:

“Whoever made that should be checked on!”

“It was more disturbing than it was stressful.”

“Do the eyes go away? I don’t like the eyes.”

When the lights suddenly flickered, one participant simply exclaimed, “Oh s\*\*\*\*!”

### **Aggregation Analysis**

Physiological and self-report data were acquired for the 6.5 minute baseline period that included the SUDS and marine life videos and for the 6.5 minute stimulus period that began with the lights flickering in the VR experience and ended each the participant’s 11<sup>th</sup> SUDS report, after the emergence of the monster. The recovery period began immediately after the 11<sup>th</sup> SUDS report, when the researcher removed the VR headset. The recovery period was not consistent

across participants, as it was dependent on how quickly they completed the final MAIA survey, and how quickly they answered the subjective questions. This recovery period varied from just over 3.5 minutes to 8 minutes with four observations being at least as long as the baseline and stimulus epochs, and 12 observations lasting at least 5 minutes.

Aggregation analyses were performed on 4 measures of PNS, 1 measure of SNS, and SUDS. Average change from baseline to stimulus (B-to-S) and from stimulus to recovery (S-to-R) were calculated for each of the above 6 measures. The results are summarized in Table 1 and described in detail below.

*Table 1: Percent Change in Conditions across All Participants*

Average Percent Change across All Participants (n = 19)						
Variable	Baseline-to- Stimulus			Stimulus-to-Recovery		
	M	SD	p	M	SD	p
PEP	-1.09%	2.87%	0.062	1.58%	5.31%	0.129
FFT HF % Pwr	18.20%	43.97%	0.069	-24.46%	29.64%	0.002
AR HF % Pwr	3.02%	30.11%	0.481	-23.44%	29.04%	0.000
RMSSD	3.83%	29.76%	0.370	586.23%	407.68%	0.000
RSA	-1.44%	31.42%	0.141	38.11%	63.83%	0.005

To assess sympathetic activation during the different conditions, analyses were performed on pre-ejection period (PEP).<sup>3</sup> Change in average PEP was calculated from baseline to stimulus, and from stimulus to recovery, for each participant, and was then averaged across participants. PEP is inversely related to SNS activation; i.e., PEP decreases when SNS increases. Average changes in PEP were in the expected direction, showing a drop in PEP from Baseline to Stimulus and an increase in PEP from Stimulus to Recovery. The percentage change in PEP from baseline to stimulus (B-to-S) approached significance ( $M = -1.09\%$ ,  $SD = 2.87\%$ ,  $p =$

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<sup>3</sup> Electrodermal activity (EDA) analysis was also planned but not possible due to an erroneous filter that was applied when the data were collected.



0.062). The percentage change in PEP from stimulus to recovery (S-to-R) was in the expected direction but was not significant ( $M = 1.58\%$ ,  $SD = 5.31\%$ ,  $p = 0.129$ ).

Similar analysis was performed on several indices of parasympathetic activation, including RMSSD, RSA, and HF-HRV. Collectively, the PNS indices showed similar overall patterns: all showed small changes from B-to-S, but none achieved significance for this condition. HF-HRV FFT approached significance ( $mean\ change = 18.2\%$ ,  $SD = 44.0\%$ ,  $p = 0.069$ ). RMSSD and RSA showed large increases from S-to-R and were significant: RMSSD ( $mean\ change = 582.6\%$ ,  $SD = 407.7\%$ ,  $p < 0.001$ ), RSA ( $mean\ change = 38.1\%$ ,  $SD = 63.8\%$ ,  $p = 0.005$ ). Both methods of HF-HRV also achieved significance, but showed changes in the opposite direction, showing an unexpected *decrease* from stimulus to baseline: HF-HRV FFT ( $M = -24.5\%$ ,  $SD = 29.6\%$ ,  $p = 0.002$ ), HF-HRV AR ( $M = -23.4\%$ ,  $SD = 29\%$ ,  $p < 0.001$ ).

Participant-reported scores on the Subjective Units of Distress Scale (SUDS) were analyzed in a similar fashion to the physiological measures. Because SUDS values can and often did include the value 0, a percent change calculation was not possible. Instead, raw difference values were used to measure changes. Overall average change in SUDS values were significant and in the expected direction for both conditions: B-to-S ( $M = 1.8$ ,  $SD = 1.2$ ,  $p < 0.001$ ) and S-to-R ( $M = -1.4$ ,  $SD = 1.1$ ,  $p < 0.001$ ). SUDS changes are shown in Table 2.

Table 2: Percent Change in Conditions across All Participants

SUDS Change across All Participants (n = 19)						
Variable	Baseline-to- Stimulus			Stimulus-to-Recovery		
	M	SD	p	M	SD	p
SUDS	1.8	1.2	0.000	-1.4	1.1	0.000

These preceding aggregation analyses are also reported for a subset of participants based on their scores on the battery of questionnaires. Those scoring in the highest and lowest quartile on each questionnaire were categorized as Hi and Lo groups, and the above 6 measures were

calculated by group. With the small sample size, most of these comparisons were not significant. However, because such an analysis was planned, the results of this Hi-Low quartile analyses are presented in Appendix B.

Scores on interoceptive awareness mostly improved, comparing questionnaires taken immediately before the stimulus to those taken immediately after the stimulus. Average scores on the MAIA showed significant increases for the “Noticing” subscale (mean increase = 26%,  $p = 0.004$ ) and for the “Attention Regulation” subscale (mean increase = 10%,  $p = 0.04$ ). Average score increases approached significance for a third subscale, “Self-Regulation” (mean increase = 39%,  $p = 0.055$ .) Scores are shown in Table 3.

*Table 3: Scores on the MAIA Before and After the Intervention*

Multi-dimensional Assessment of Interoceptive Awareness scale (MAIA) Version 1																
	Noticing		Not-Distracting		Not-Worrying		Attention Regulation		Emotional Awareness		Self-Regulation		Body Listening		Trusting	
	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post
Mean	2.6	3.3	2.1	2.2	2.6	2.7	2.8	3.1	3.4	3.5	2.9	3.2	2.7	2.8	3.6	3.6
Dif in Means	0.67		0.09		0.07		0.27		0.07		0.33		0.12		0.02	
Percent Increase	26%		4%		3%		10%		2%		39%		5%		0%	
Sig. One-Tailed $p =$	0.004		0.318		0.369		0.035		0.305		0.055		0.290		0.455	

A further exploration of the merits of the different PNS indices is warranted. An exhaustive analysis is beyond the scope of this research, but as a starting point, a correlation matrix was calculated for the PNS indices in Table 4.

*Table 4: Correlation Matrix of Parasympathetic Nervous System Indices*

<b>RMSSD</b>			0.48
<b>AR HF % Pwr</b>		-0.08	0.58
<b>FFT HF % Pwr</b>	0.94	-0.05	0.53
	<b>AR HF % Pwr</b>	<b>RMSSD</b>	<b>RSA</b>

*Correlation matrix of the four PNS indices used:*

*RMSSD: Root Mean Square of the Successive Differences of each heartbeat*

*AR HF % Pwr: High-Frequency Heart Rate Variability % Power using the Auto Regressive method*

*FFT HF % Pwr: High-Frequency Heart Rate Variability % Power using the Fast Fourier Transform method*

*RSA: Respiratory Sinus Arrhythmia using the peak-to-trough method*

Both HF-HRV indices show a large positive correlation with each other. RMSSD shows no correlation with any of the three other indices. RSA shows a moderate  $\sim 0.5$  correlation with all 3 indices.

### **Behavioral Analysis**

Behavioral analysis was conducted using the accelerometry data and by reviewing the video recordings of the participants. Patterns of accelerometer mobility most closely correlated with behavioral patterns of head movement – the accelerometer was positioned on top of the VR headset in hopes that it would measure postural changes. However, it was noted that most head movements corresponded with visual exploration of the VR environment. Therefore, moments of greatest accelerometer readings tended to correspond more with environmental exploration than with a sensitivity to perceived threat. Future studies using a VR stimulus are recommended to position accelerometer transducers on the sternum.

Analysis via behavioral scoring was planned by looking at the degree to which participants leaned into the stimulus (approach behavior suggestive of fight) or leaned away from the stimulus (avoidance behavior suggestive of flight). However, participants showed such a limited range of movement that such a behavioral coding scheme was rendered useless for this analysis. Behaviors most discernable amounted to finger fidgeting, facial expressions, and verbalizations during the stimulus. However, because no behavioral coding scheme was planned for these behaviors, no further behavioral analysis was conducted. Qualitative color from the verbal comments was used to better understand the potency of the stimulus as noted above.

### **Qualitative Analysis of Time Series Data**

Analysis of the temporal dynamics examined key physiological variables against one another and against the SUDS time series. Most indices of PNS activity, including HF-HRV and

RMSSD, are recommended to be averaged over at least five minutes (Malik, 1996). However, one measure, RSA can be calculated as a time series, so long as respiration data is gathered along with the ECG signal (Grossman et al., 1990). Therefore, a time series of RSA was used to index PNS activity and a time series of PEP was used to index SNS activity.

In the time series analysis that follows, participant-level time series are presented and analyzed for participants scoring in the highest and lowest quartiles on most questionnaires. Participants scoring mostly in the middle two quartiles were not examined in this analysis. In each graph, the questionnaires on which each participant scored in the highest or lowest quartile are listed at the top of each graph.

Within each time series graph, several key time markers are noted: beginning and ending of baseline, stimulus, and recovery periods are marked with orange lines. Three key events during the stimulus period are highlighted with red lines. First, “Lts Flkr” indicates when the lights flickered and went out in the VR experience, accompanied by a surprisingly loud and sudden electrical buzzing noise. This marked the end of baseline and the simultaneous beginning of the stimulus period. The second red line labeled “H Flash” marks the brief, in-your-face appearance of the inkblot monster (affectionately nicknamed Hermie by the researchers, in honor of Hermann Rorschach, whose inkblots were the inspiration for the fear video.) “H Emg” indicates the final emergence of Hermie, when he appears to climb out of the movie screen and approach the participant’s neck.

Where possible, the first page of graphs for each Figure reflects participants scoring in the highest quartile on questionnaires indexing adaptive traits, including the Five Factor Mindfulness Questionnaire (FFMQ), the Multidimensional Assessment of Interoceptive Awareness (MAIA), and the Anxiety Control Questionnaire (ACQ).

The second page of graphs in each Figure includes mostly participants that scored in the highest quartile on disorder-related questionnaires, including the Trauma History Screen (THS), PTSD Checklist (PCL), State-Trait Anxiety Index (STAI), the Patient Health Questionnaire – Panic Disorder subscale (PHQ-PD), the Anxiety Sensitivity Index (ASI), the Penn State Worry Questionnaire (PSWQ), and the Fear Survey Schedule – Blood Injury subscale (FSS-BI).

The third page of graphs includes specifically the four participants that scored in the highest quartile for number of previous dissociative experiences on the THS (abbreviated to DISS on the graphs).

Exceptions to the above categorizations exist. For example, the PHQ-PD is designed to determine whether a person has clinical or sub-clinical level panic or not. Sixteen of the 19 participants scored the lowest possible score on this questionnaire: 0. Therefore three-quarters of the participants are listed as being in the in the lowest “quartile” for PHQ-PD.

Also, several participants scoring in the highest quartile on the FSS-BI also scored in the top quartile for adaptive questionnaires.

Finally, two participants scored in the highest quartile for both the THS and several adaptive questionnaires including collectively the FFMQ, MAIA-Pre, MAIA-Post, and the ACQ. These two participants are also analyzed in further detail in the next section.

### ***PEP vs. SUDS Time Series***

PEP is calculated from two different aspects of the cardiac cycle, which means that each new value in a PEP time series is on a time scale that is unique to that participant. One participant may have shorter cardiac cycles than the next participant, making their individual PEP times series mathematically problematic to align and compare directly with other

participants. Therefore, the most straightforward approach to view PEP time series is to view them at the participant level.

Figure 9 shows time series for PEP and SUDS by participant, for the selected participants mentioned above. As a raw signal, PEP is extremely variable, leaving a graph of raw PEP difficult to read with respect to trends. Further, raw PEP signals are extremely sensitive to noise, causing significant numbers of “0” values when a value could not be calculated. In order to eliminate these inaccurate 0 values, and to smooth the raw PEP signal to identify trends, a 37-cycle centered moving average was calculated. For completeness, both the raw signal and the 37-cycle moving average are plotted in Figure 9.

Figure 9: PEP and SUDS Time Series by Participant

Figure 9a: Participants in the Highest Quartile for Questionnaires Indexing Adaptive Functioning

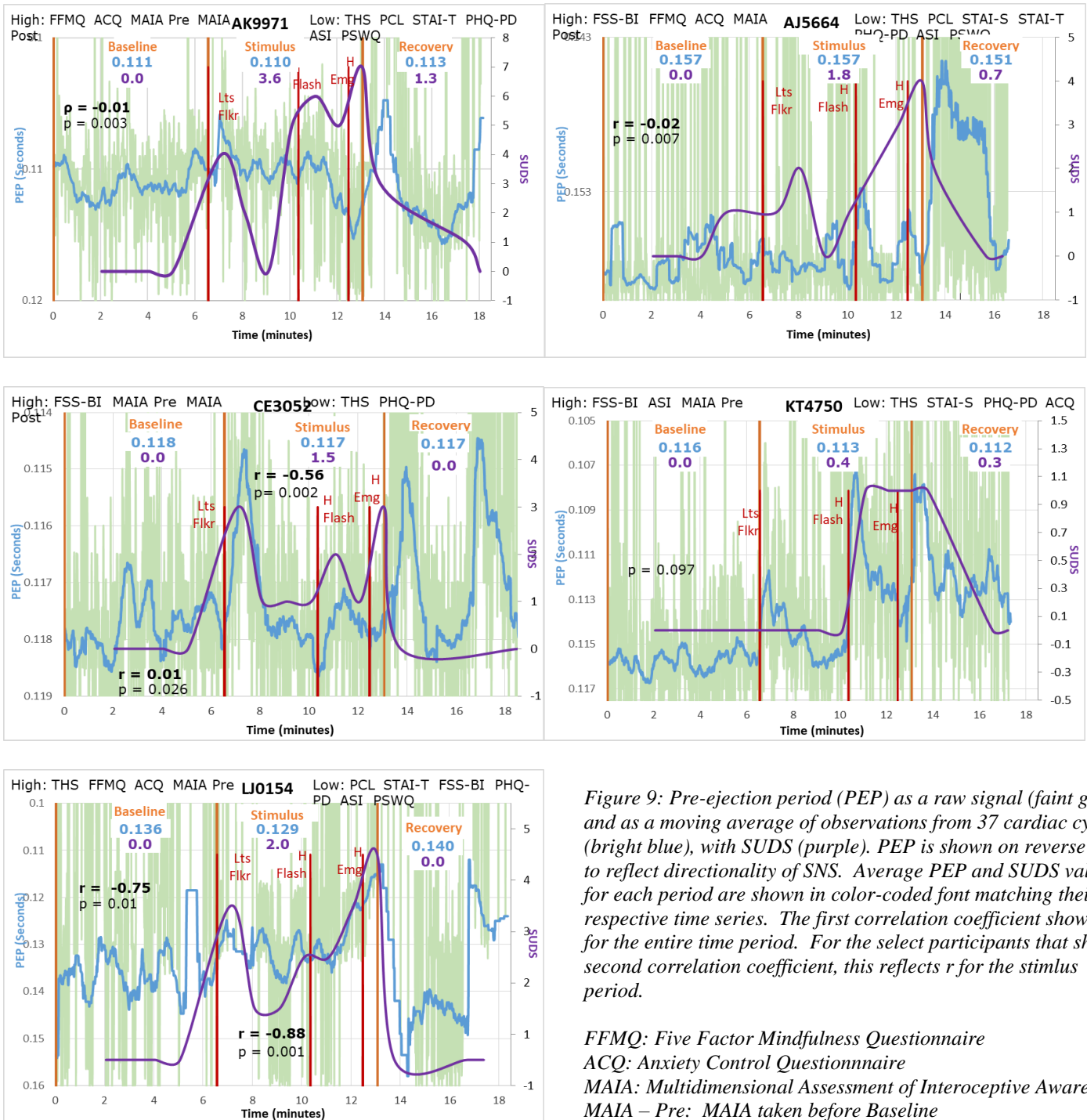


Figure 9: Pre-ejection period (PEP) as a raw signal (faint green) and as a moving average of observations from 37 cardiac cycles (bright blue), with SUDS (purple). PEP is shown on reverse scale to reflect directionality of SNS. Average PEP and SUDS value for each period are shown in color-coded font matching their respective time series. The first correlation coefficient shown is for the entire time period. For the select participants that show a second correlation coefficient, this reflects  $r$  for the stimulus period.

FFMQ: Five Factor Mindfulness Questionnaire  
 ACQ: Anxiety Control Questionnaire  
 MAIA: Multidimensional Assessment of Interoceptive Awareness  
 MAIA – Pre: MAIA taken before Baseline  
 MAIA – Post: MAIA taken after Stimulus  
 FSS-BI: Fear Survey Schedule – Blood Injury subscale  
 THS: Trauma History Screen  
 PCL: PTSD Checklist  
 STAI: State Trait Anxiety Index  
 STAI-T: Trait subscale of the STAI  
 STAI-S: State subscale of the STAI  
 PHQ-PD: Patient Health Questionnaire – Panic Disorder  
 ASI: Anxiety Sensitivity Index  
 PSWQ: Penn State Worry Questionnaire  
 DISS: Dissociative experiences from the THS

Figure 9b: Participants in the Highest Quartile for Disorder-Related Questionnaires

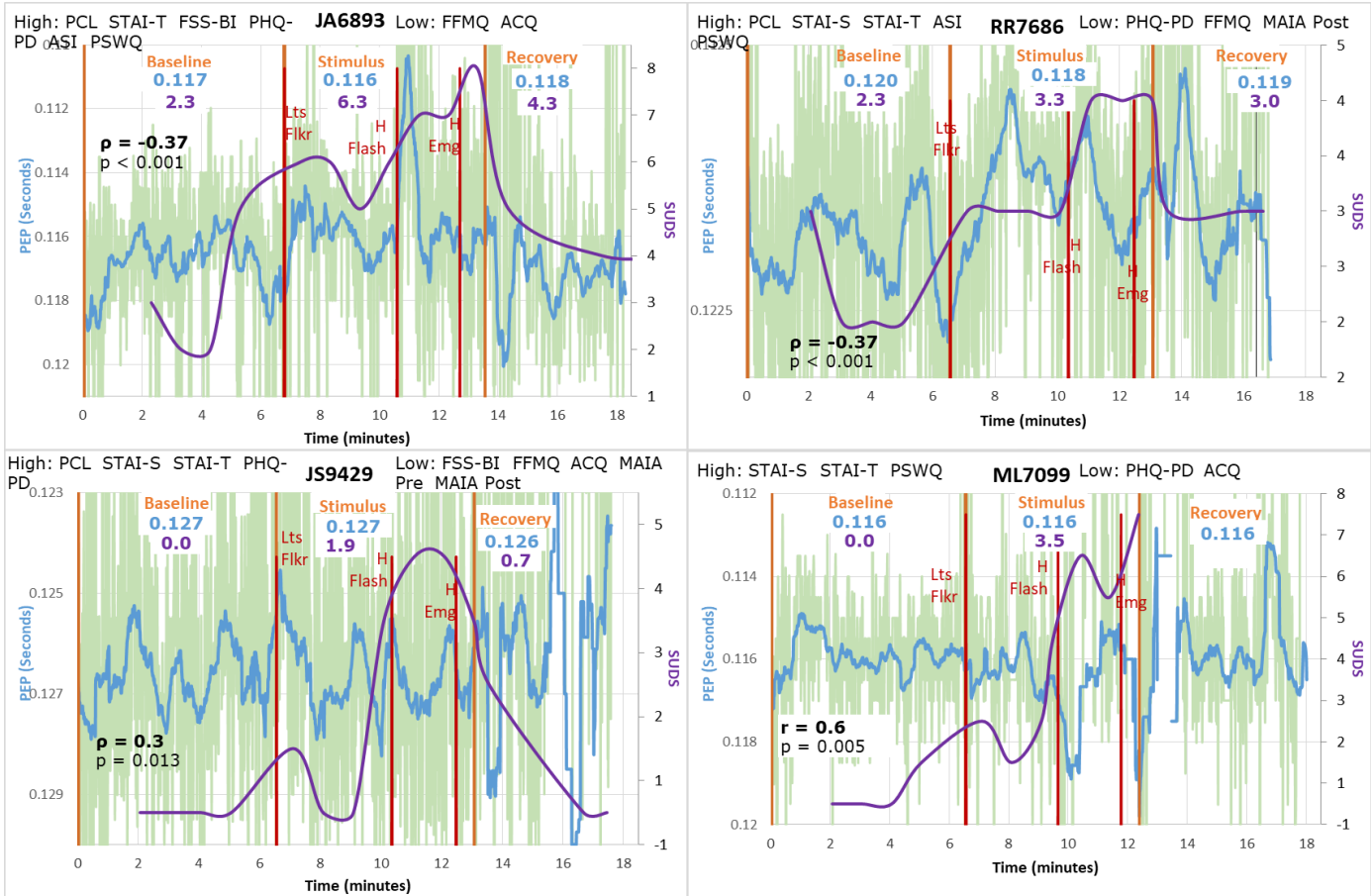
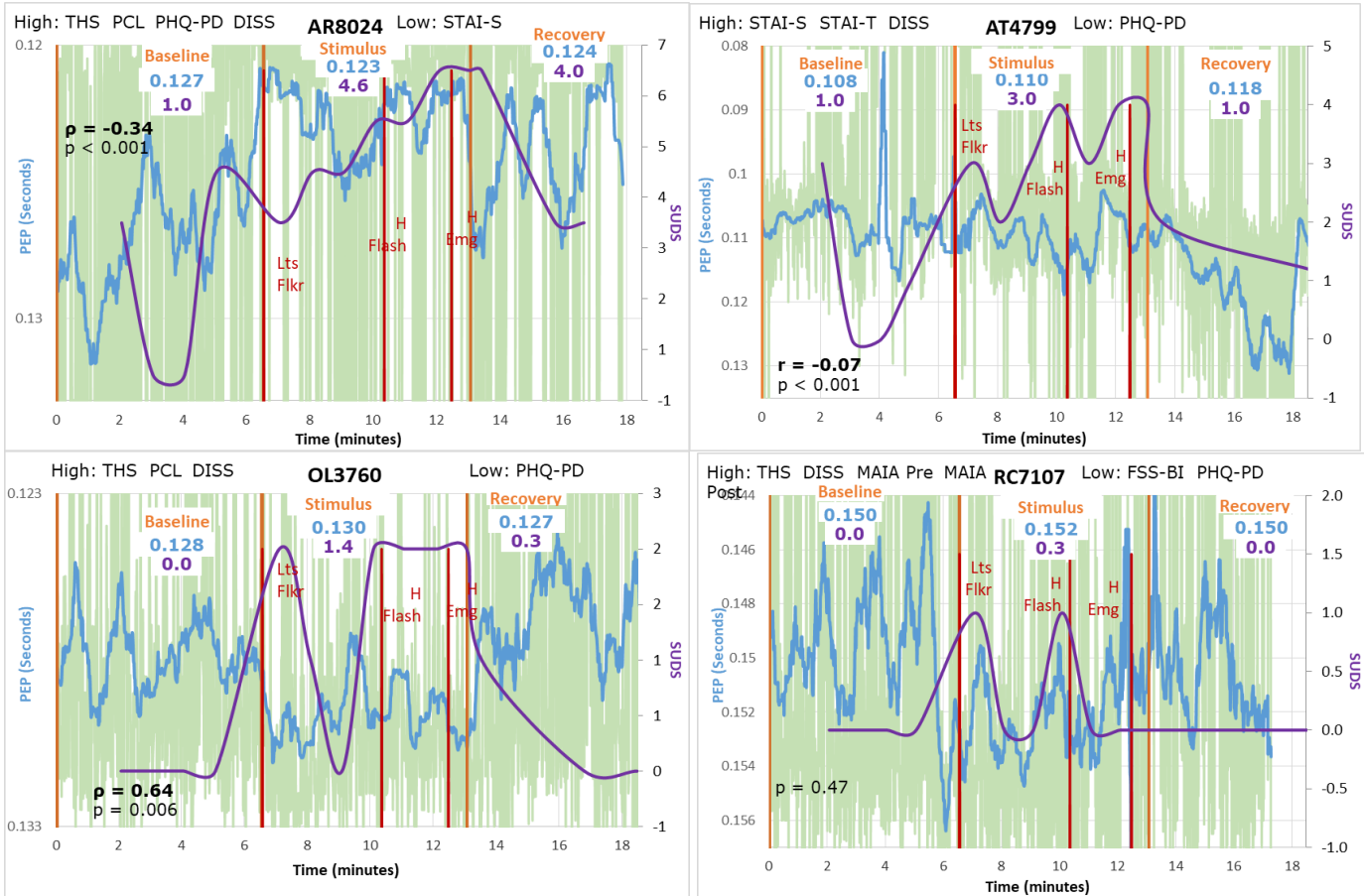




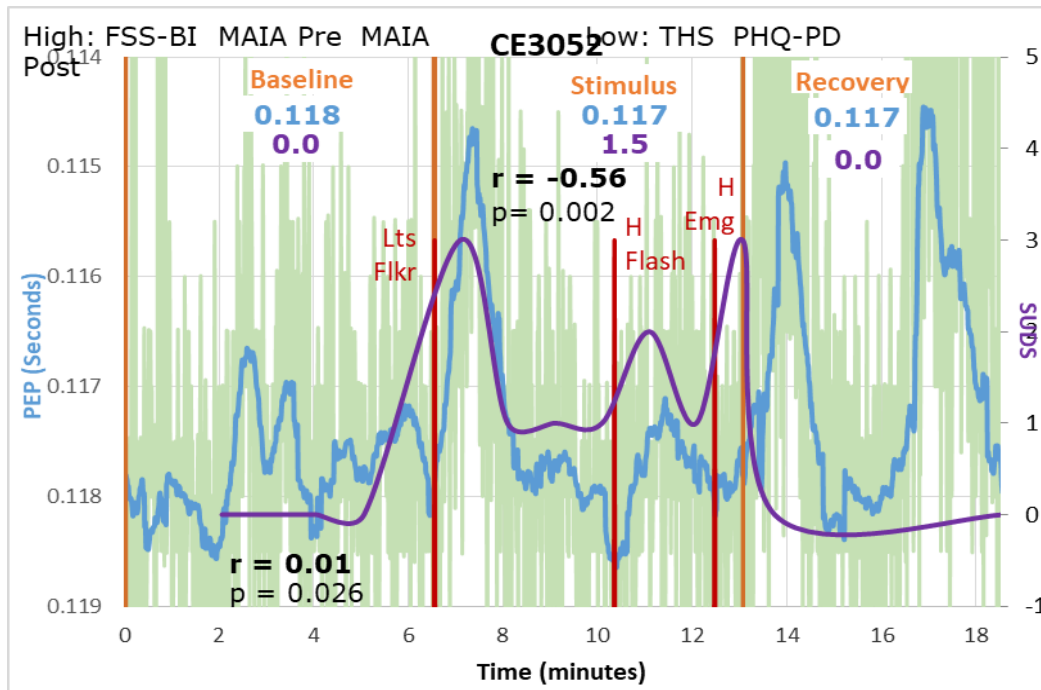
Figure 9c: Participants in the Highest Quartile for Dissociative Scores



For the sake of direct comparison between averaged values and time series values, the mean PEP value and the mean SANS value are each displayed in the graph for Baseline, Stimulus, and Recovery conditions. Because PEP is a negative index of SNS (lower PEP values reflect higher SNS activation), the PEP scale is shown in reverse order, where lower values are at the top of the graph and higher values are at the bottom. Therefore, when the PEP graph increases, it is accurately visualized as SNS increases. However, correlation coefficients do not reflect this directional change. Therefore, an apparently high positive concordance between PEP and SANS time series will show up as a negative correlation coefficient. All but two participants' correlations were statistically significant. For the two that were not significant, no correlation coefficient is shown; only the insignificant p value is shown.

In order to better examine the information conveyed in the graphs of Figure 9, it is helpful to examine a participant in detail. Participant “CE3052” has been excerpted and is reproduced in Figure 9d for further analysis.

Figure 9d: PEP as raw value (faint green) and 37-cycle moving average with SUDS for participant CE305



The mean PEP score for each period is reported in light blue bolded font, shown during the respective period for which it is averaged. PEP values as shown are as follows: Baseline = 0.118, Stimulus = 0.117, and Recovery = 0.117. These numbers may be significant, but appear quite unimpressive.

The time series however, reveals a quite different, more dramatic picture. First, the onset of the stimulus when the lights flickered and buzzed in the VR experience (described in the Method section) had a dramatic response in PEP. A quick cross-reference to the participant’s self-reported SUDS ratings during the same time period supports the conclusion that this stimulus onset had the intended, distressing effect. Once the intensity of this stimulus was

processed, this participant's PEP value returned quite quickly to its pre-stimulus point, and remained so until the next jarring image of the monster unexpectedly staring them in the face. PEP bounced again from this encounter and SUDS tracked right with it. Then PEP restored to its original pre-stimulus level for nearly the duration of the stimulus period. Averaged all together, those PEP dips and spikes presented a bland 0.117 mean PEP value for the stimulus period, just barely different from the baseline level of 0.118.

Similarly, averaged all together, the SUDS mean value for the stimulus period was 1.5. But in between those averages, SUDS shot up from 0 to 3 from the lights flickering, dropped back to 1, bumped up to 2 after the monster flash, dropped again to 1, and then shot back up to 3 when the monster emerged. Condition averages repeatedly appear to miss the essence of each participant's actual experience.

The theme of averaging can be repeated when observing the reported correlations in Figure 9d. The first correlation coefficient reported for CE3052 is  $r = 0.01$ , with a significant  $p = .026$ . If one were to take this correlation at face value, one would assume that PEP and SUDS did not move very much in tandem for this participant during the study. However, a quick visual scan of the actual time series in the graph suggests otherwise. The stimulus period especially seems to show PEP and SUDS moving in lock-step. Indeed, the PEP-SUDS correlation for the stimulus period was a whopping  $r = -0.56$ . The graph reports this correlation as negative only because again, PEP is a negative index of SNS. The  $r = -0.56$  is best interpreted as  $r = 0.56$  between SNS and SUDS. Thus, it is important to note that averaging over too much data erases all utility the data may have offered. Only a time series view of CE3052's experience could reveal what more should be examined beyond the paltry  $r = 0.01$  for the overall experiment.

Relatedly, it is important to note that the SUDS values for many participants were zero, for many reports over the course of the experiment. In fact, CE3052's SUDS value was zero for all 4 baseline values and for all 3 recovery values. A correlation coefficient cannot be calculated for two series when one of its series is unchanging throughout the epoch of interest. Therefore, it was impossible to further unpack CE3052's experiment-wide  $r = 0.01$  to determine a baseline and recovery correlation coefficient.

Directly below the graph for CE3052, participant LJ0154 reveals an even stronger SNS-SUDS correlation of 0.88 for the stimulus period (-0.88 for PEP-SUDS). However, not all participants showed such strongly positive SNS-SUDS correlations (negative PEP-SUDS correlations), and several even the opposite relationship. A simple rule of thumb for quickly visualizing correlations from time series is that if they appear to be mirror images of each other, they are likely negatively correlated. ML7099 from Figure 9b and OL3760 in Figure 9c are excellent examples. (Of course their  $r$  values read as positive because of the reverse PEP relationship with SNS.) A third example is JS9429, shown directly before ML7099 in Figure 9b. Notably, all three of these seeming exceptions scored in the highest quartile for several disorder questionnaires.

Returning to participant LJ0154, an interesting dichotomy is revealed: this participant reported the second highest number of personal traumas in their history across all participants, as indexed by the THS. And yet simultaneously, LJ0154 scored in the highest quartile on mindfulness (FFMQ), anxiety control (ACQ), and interoceptive awareness (MAIA-Pre) and scored in the lowest quartile for the PCL. THS indexes personal history of traumas, whereas PCL indexes PTSD symptoms. Relatedly, RC7107 also scored in the highest quartile for THS, but also scored in the highest quartile for both versions of the MAIA revealing a strong

mindfulness to balance the history of trauma. These participants provide two poignant examples of how trauma history does not necessarily sentence a person to a lifetime of PTSD.

### *RSA vs. SUDS Time Series*

Respiratory Sinus Arrhythmia (RSA) is calculated from two different channels of data: the ECG channel and the respiration channel. It can be obtained by taking the longest inter-beat interval (IBI) during exhalation and subtracting the shortest IBI during the inhalation. This calculation, called the peak-to-trough method, leverages the natural increase in heart rate that occurs during inhalation and the decrease during exhalation. This respiration-related fluctuation in heart rate rhythm reflects vagal control of the heart, so measuring RSA provides an index of PNS. As such, RSA values are anchored to the respiration cycle – a new value is registered at the beginning of each new respiration. In this way, respiration cycles, like cardiac cycles, are unique to the participant. One participant may have shorter cycles than the next participant. Therefore, much like with PEP, individual RSA time series are mathematically problematic to align and compare directly with other participants. Once again, the most straightforward approach to view RSA time series is at the participant level.

Figure 10 shows time series graphs for RSA and SUDS by participant, for the same subset of participants that were reported in Figure 9, and similarly reporting RSA and SUDS averages by condition. As with PEP, the RSA signal can be extremely variable from one observation to the next, so for the sake of comparability, the RSA value in the graphs reflects a moving average of observations reported over 37 cardiac cycles.

Figure 10: RSA and SUDS Time Series by Participant

Figure 10a: Participants in the Highest Quartile for Questionnaires Indexing Adaptive Functioning

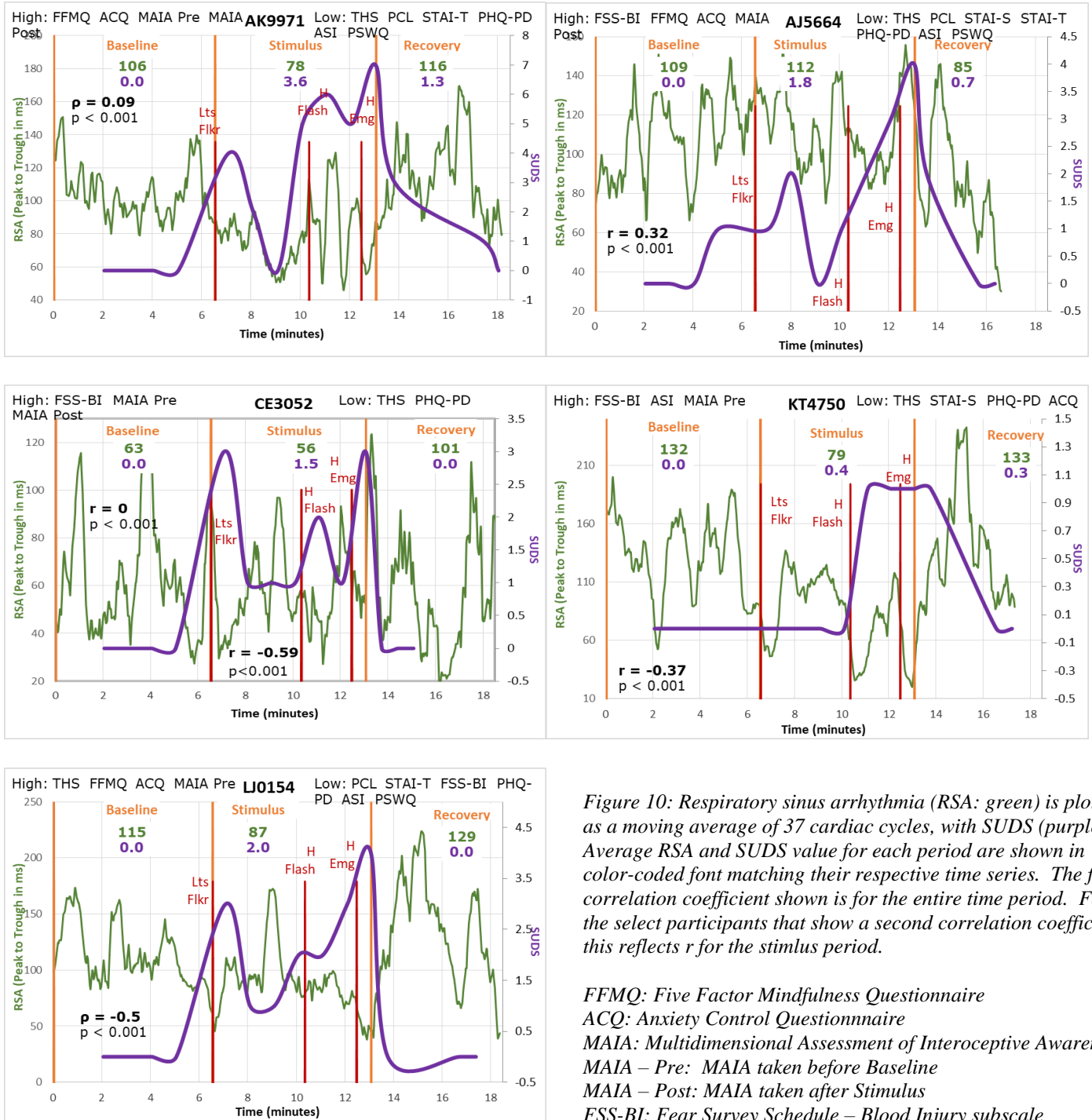


Figure 10: Respiratory sinus arrhythmia (RSA: green) is plotted as a moving average of 37 cardiac cycles, with SUDS (purple). Average RSA and SUDS value for each period are shown in color-coded font matching their respective time series. The first correlation coefficient shown is for the entire time period. For the select participants that show a second correlation coefficient, this reflects  $r$  for the stimulus period.

- FFMQ: Five Factor Mindfulness Questionnaire
- ACQ: Anxiety Control Questionnaire
- MAIA: Multidimensional Assessment of Interoceptive Awareness
- MAIA – Pre: MAIA taken before Baseline
- MAIA – Post: MAIA taken after Stimulus
- FSS-BI: Fear Survey Schedule – Blood Injury subscale
- THS: Trauma History Screen
- PCL: PTSD Checklist
- STAI: State Trait Anxiety Index
- STAI-T: Trait subscale of the STAI
- STAI-S: State subscale of the STAI
- PHQ-PD: Patient Health Questionnaire – Panic Disorder
- ASI: Anxiety Sensitivity Index
- PSWQ: Penn State Worry Questionnaire
- DISS: Dissociative experiences from the THS

Figure 10b: Participants in the Highest Quartile for Disorder-Related Questionnaires

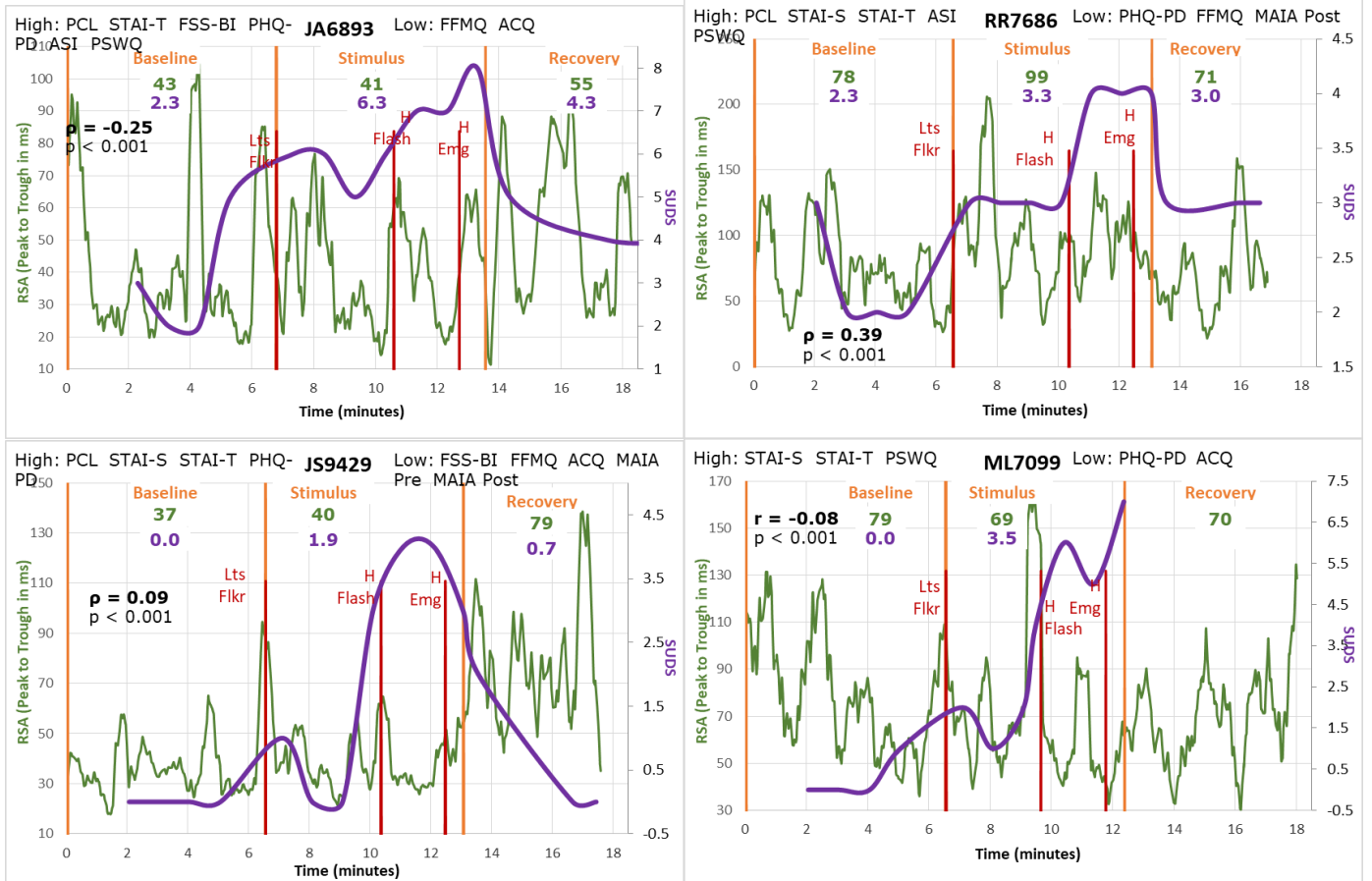
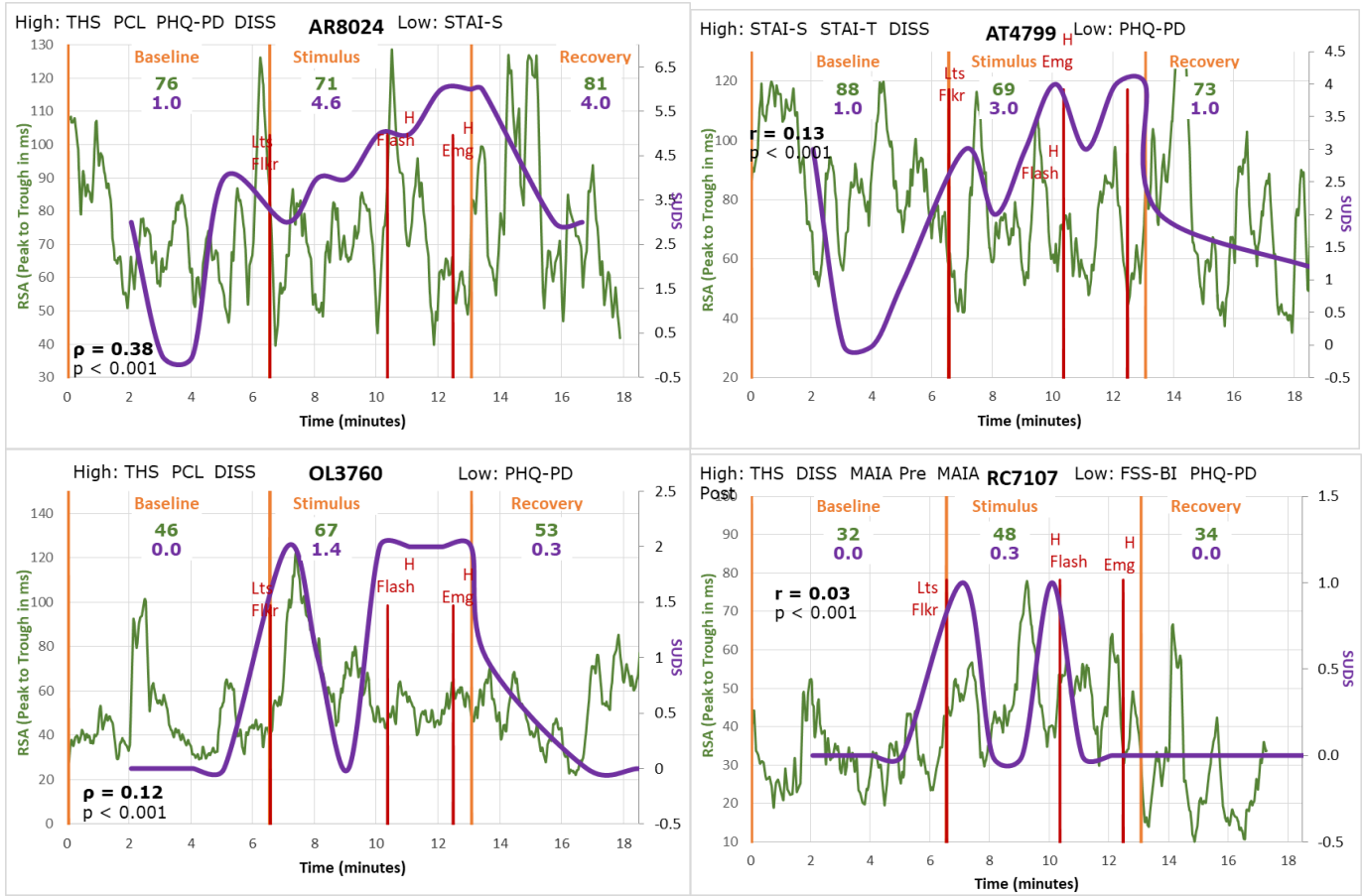


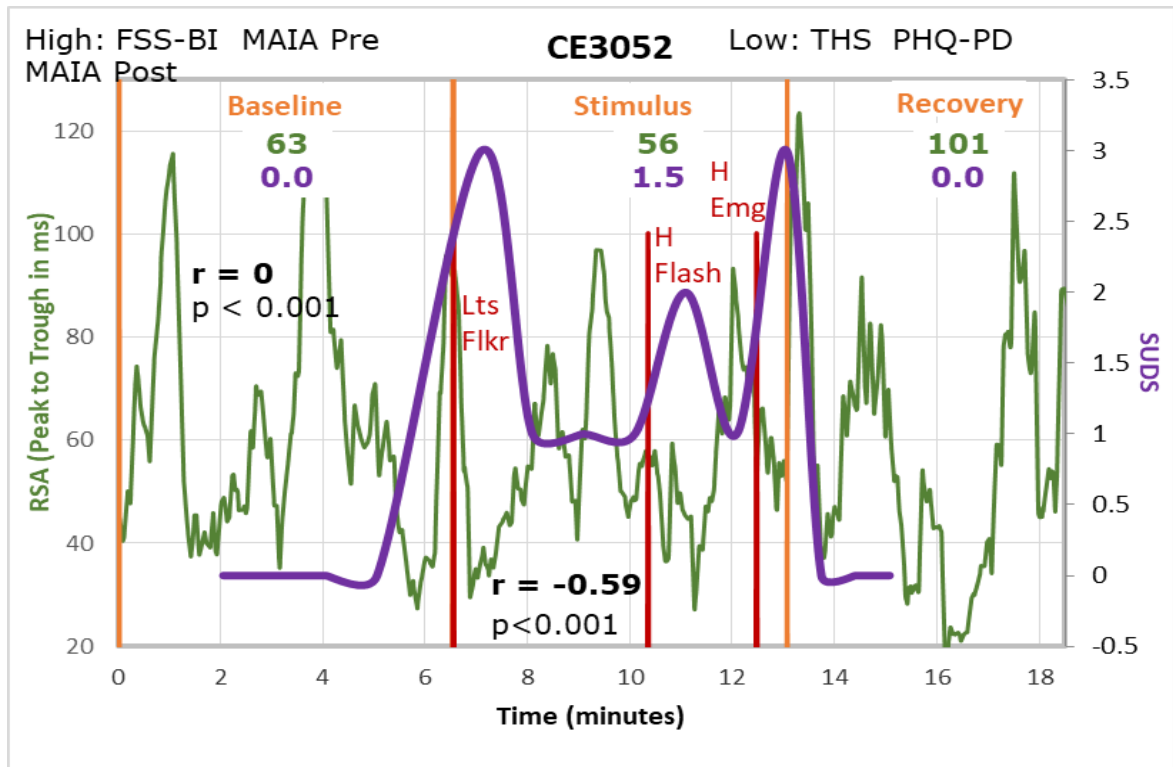
Figure 10c: Participants in the Highest Quartile for Dissociative Scores





Returning to the textbook participant, CE3052 is reproduced in Figure 10d.

Figure 10d: 37-cycle moving average of RSA with SUDS for participant CE3052.



CE3052's overall correlation between RSA and SUDS was zero. However, recall that this number is likely diluted by the unchanging string of 0 SUDS values throughout the entire baseline and recovery periods. Once again, the stimulus period provides a strong correlation – this time reflecting a significantly negative correlation between PNS and SUDS. This is as expected, since moderate levels of distress (SUDS peaking at 3) tend to lead to primary reciprocal engagement of the SNS. The RSA and SUDS time series lines for this participant illustrate an excellent example of a near perfect mirror image of one another.

### ***RSA vs. PEP Time Series***

Finally, RSA and PEP are plotted together in a time series in Figure 11. Both reflect a moving average of observations reported over 37 cardiac cycles. Once again, the scale for PEP is reversed, such that an apparent drop in PEP reflects a drop in SNS activation, while reported

correlation coefficients remain in the original direction. So time series that appear to be negative mirror image of one another should reflect a positive correlation coefficient.

Taken altogether these RSA and PEP time series are the empirical data that comes the closest to mapping the physiological temporal dynamics of the defense cascade. The analysis that follows will attempt to characterize these autonomic dynamics by questionnaire group.

Figure 11: RSA and PEP Time Series by Participant

Figure 11a: Participants in the Highest Quartile for Questionnaires Indexing Adaptive Functioning

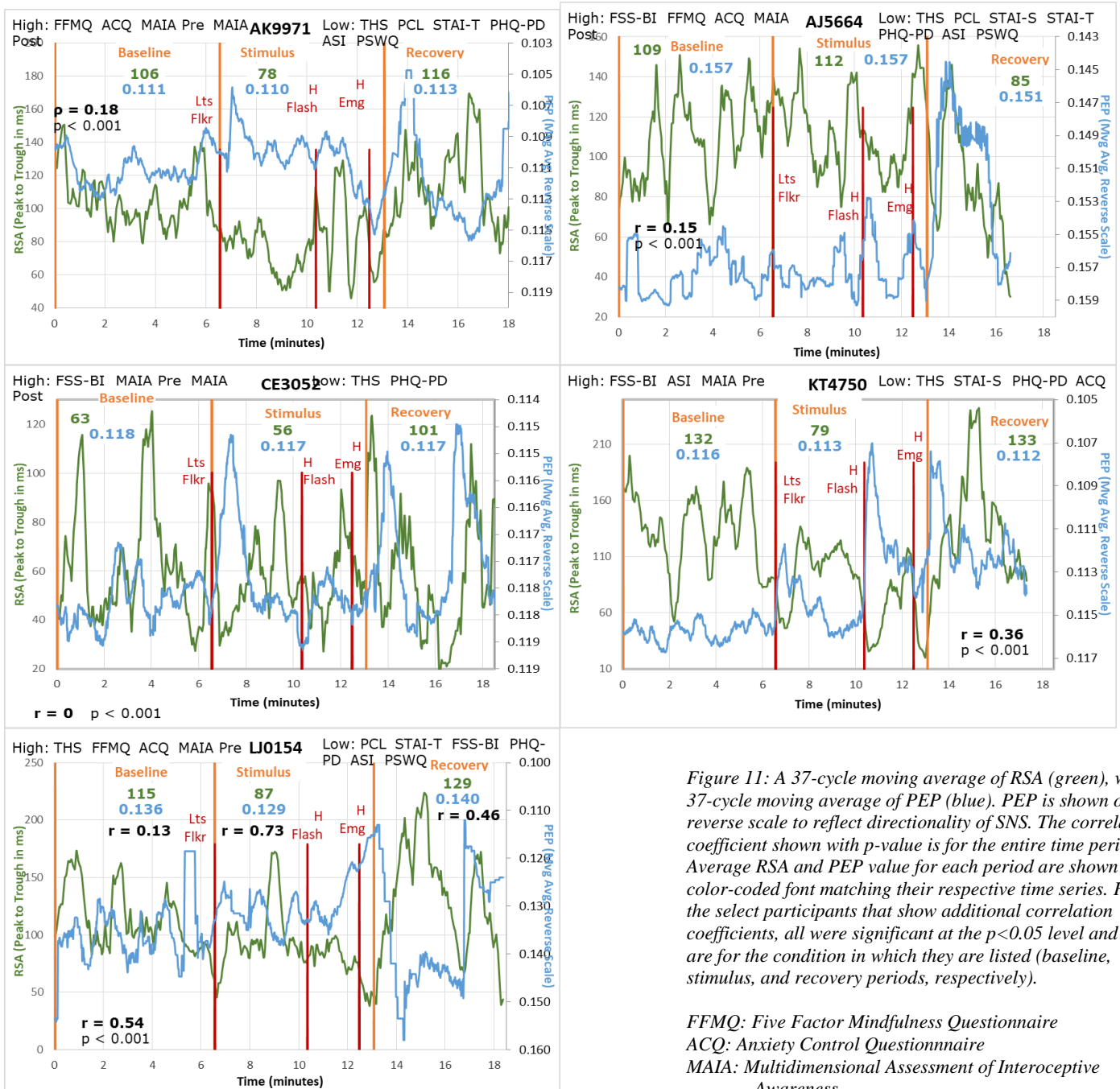


Figure 11: A 37-cycle moving average of RSA (green), with 37-cycle moving average of PEP (blue). PEP is shown on reverse scale to reflect directionality of SNS. The correlation coefficient shown with p-value is for the entire time period. Average RSA and PEP value for each period are shown in color-coded font matching their respective time series. For the select participants that show additional correlation coefficients, all were significant at the  $p < 0.05$  level and they are for the condition in which they are listed (baseline, stimulus, and recovery periods, respectively).

- FFMQ: Five Factor Mindfulness Questionnaire
- ACQ: Anxiety Control Questionnaire
- MAIA: Multidimensional Assessment of Interoceptive Awareness
- MAIA – Pre: MAIA taken before Baseline
- MAIA – Post: MAIA taken after Stimulus
- FSS-BI: Fear Survey Schedule – Blood Injury subscale
- THS: Trauma History Screen
- PCL: PTSD Checklist
- STAI: State Trait Anxiety Index
- STAI-T: Trait subscale of the STAI
- STAI-S: State subscale of the STAI
- PHQ-PD: Patient Health Questionnaire – Panic Disorder
- ASI: Anxiety Sensitivity Index
- PSWQ: Penn State Worry Questionnaire
- DISS: Dissociative experiences from the THS

Figure 11b: Participants in the Highest Quartile for Disorder-Related Questionnaires

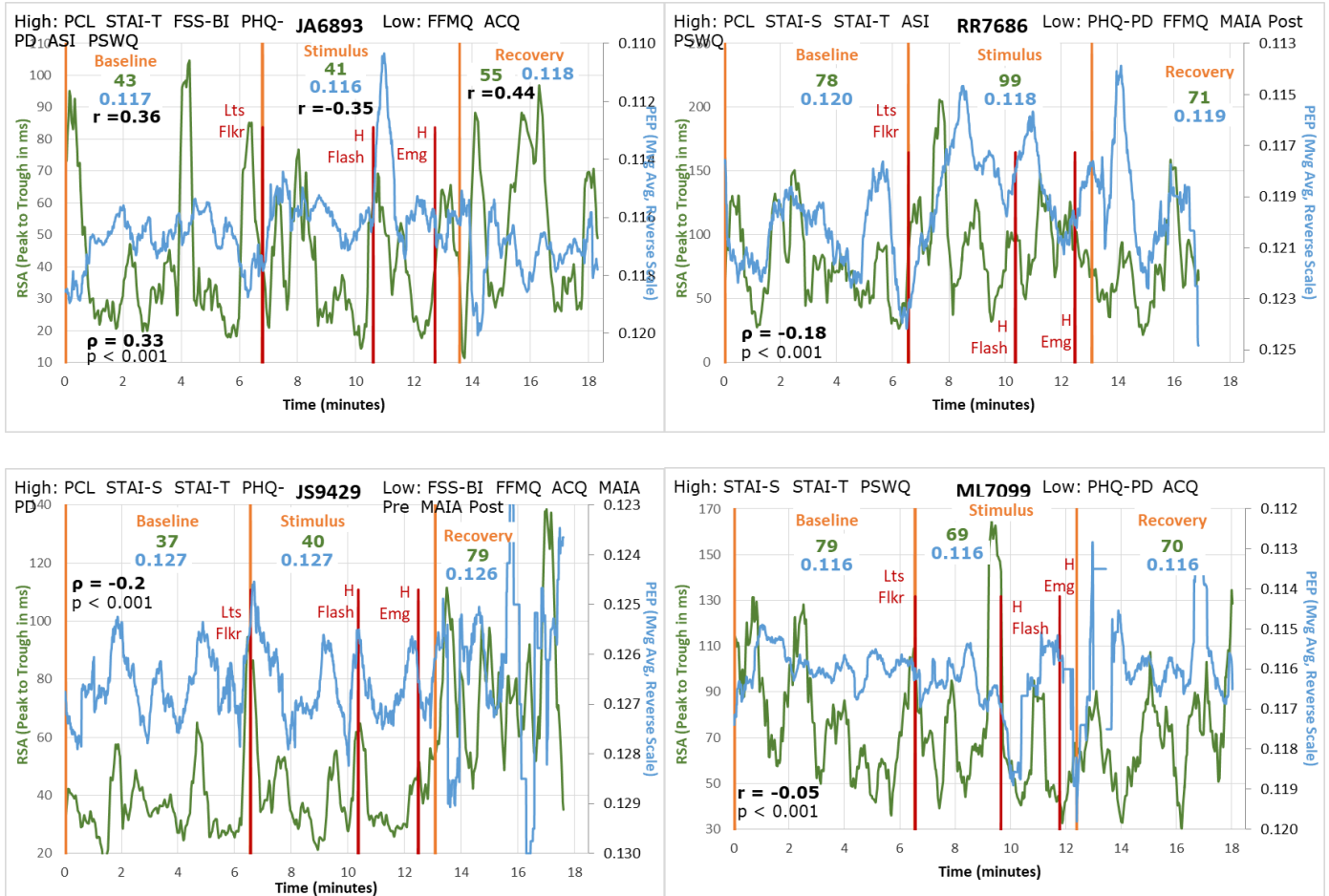
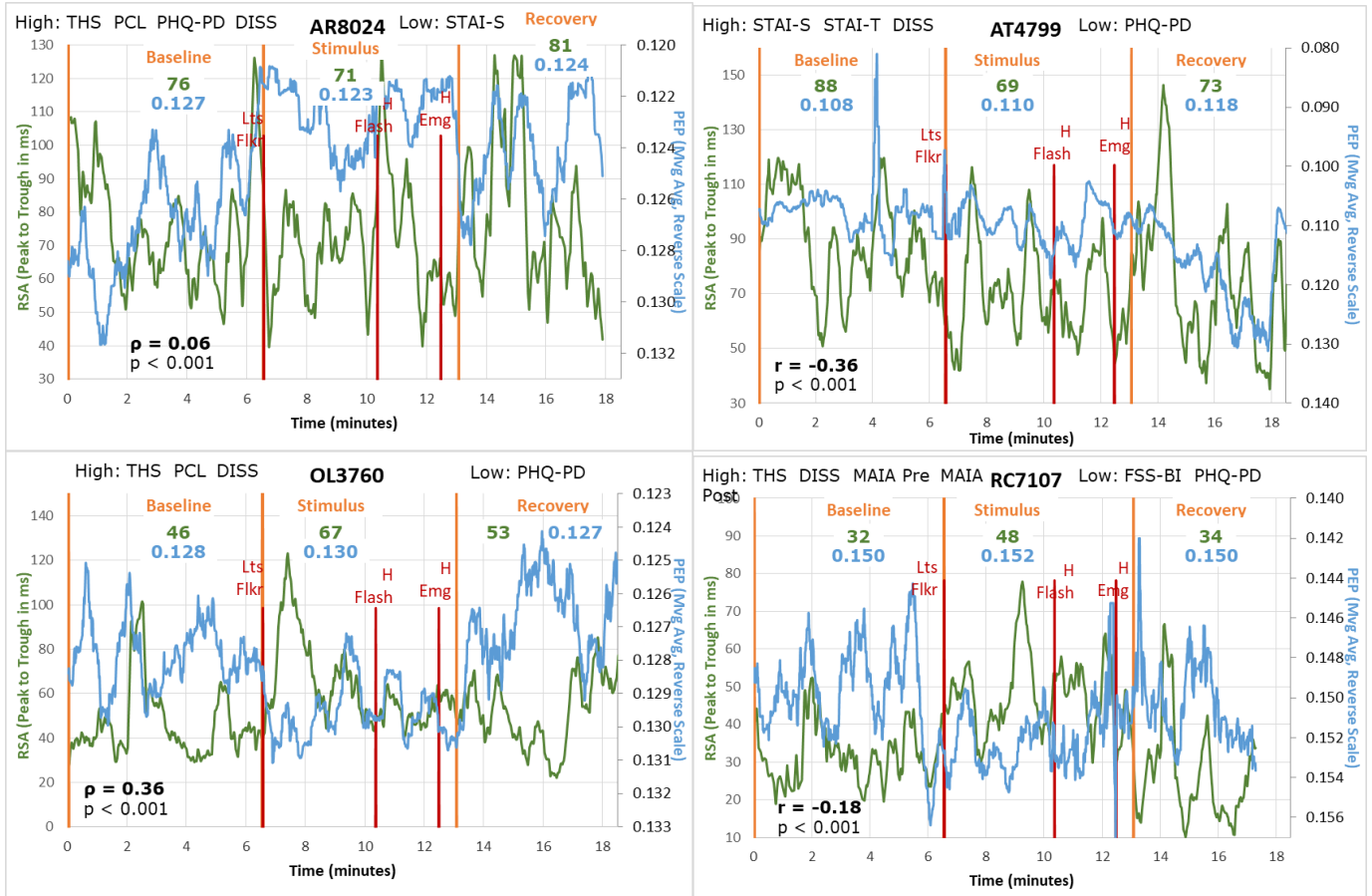


Figure 11c: Participants in the Highest Quartile for Dissociative Scores



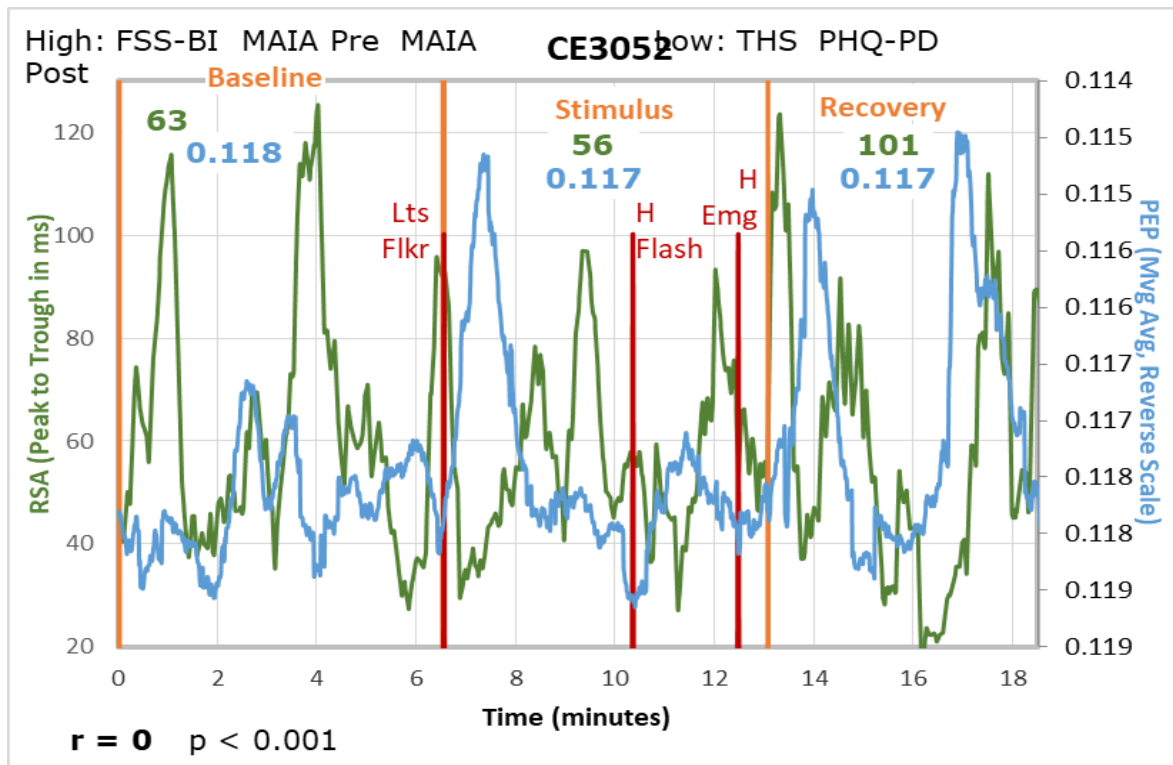
Looking at the “adaptive” group as an aggregate, this group showed primarily RSA (PNS) dominance during the baseline period. However, there was no clear group trend towards PEP (SNS) dominance as they moved into the stimulus period. One participant showed this trend; another showed continued RSA dominance; the other three seemed to fluctuate back and forth between RSA and PEP dominance. The recovery period was similarly without consensus. Two participants showed a restoration of PNS dominance, one continued to fluctuate between PNS and SNS, while the other two fell into a pattern of PNS-SNS coactivation.

The “disorder” group showed fewer discernable patterns. The group appeared to have a tonically higher level of SNS throughout the study, but whether this was reciprocally antagonistic with PNS is unclear. Some of this group’s SNS activation seemed clearly coactive, as PNS

tracked with the SNS movement. In between, there was much fluctuation between PNS and SNS dominance every two minutes throughout the entire study. The other participant, who scored high on both the state and trait version of the STAI, showed consistent SNS activation throughout both baseline and stimulus before dissolving into a static of PNS and SNS noise during recovery. The “dissociation” group showed essentially no patterns at the group level.

Figure 11 does however introduce the possibility of determining each participant’s individual sequence of autonomic activation. Returning to participant CE3052 this time in Figure 11d, there appears to be a PNS dominance during baseline, followed by a brief SNS dominance for the first two minutes of the stimulus, followed by a return to PNS dominance through the end of the stimulus.

Figure 11d: 37-cycle moving average of RSA and PEP for participant CE3052



The recovery period seems to change patterns from largely reciprocal activation what appears to be coactivity of the PNS and SNS. This series of observations brings into focus the need to examine further the correlational dynamics between SNS and PNS.

At first glance one sees an apparently strong negative correspondence between RSA and PEP, noting any number of large dips and spikes that happen in opposition to one another. And once again, the overall participant correlation coefficient delivers a disappointing  $r = 0$ . The fallacy of averaging over too much data emerges once more. When isolated to just the stimulus period alone, the correlation powers up to a strong  $r = -0.43$  ( $p < 0.001$ ).

Returning to the graph, another surprise: the recovery period reverses this pattern with RSA and PEP suddenly to falling back into lock-step with each other. Indeed, the correlation coefficient for this period turns strongly positive  $r = 0.33$  ( $p < 0.001$ ). Examining these two periods alone, it is not hard to understand now how the overall correlation coefficient for this participant was 0 – the strong positive and strong negative correlations simply cancelled each other out.

Keeping in mind the cancelling-out theme, one can now examine the baseline period for this participant. Depending on where the eye first falls, one could either get a sense of strongly negative correlation or a positive one. The first two minutes shows the classic mirror image, followed by the next two minutes during which the two series fall into lock step, followed by the remaining 2½ minutes where they flip once more and show a mirror image again. Based on this simple description, it is not hard to predict the slightly negative correlation that balanced out for the overall baseline period:  $r = -0.18$  ( $p < 0.001$ ).

After examining several graphs in such excruciating detail, it becomes apparent there must be a simpler way to net out the correlational dynamics of the SNS-PNS relationship. The

next section will introduce such an approach using scatterplots to help visualize the correlations for each condition.

### **Correlational Analysis**

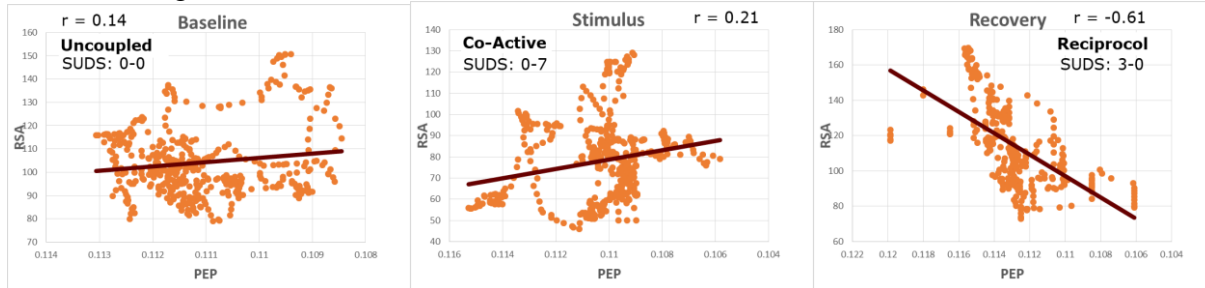
RSA and PEP are plotted together in correlational scatterplots by participant, by condition in Figure 12. The scale for PEP is reversed and the reported correlation coefficients are multiplied by -1. All correlations reported were significant at  $p < 0.001$ . An algorithm was developed for this analysis to attempt to map the raw correlation values to a meaningful indicator of ANS space. Specifically, correlations greater than 0.2 have been assigned the label “Co-Active; correlations between -0.2 and 0.2 are labeled “Uncoupled”, and correlations less than -0.2 are labeled “Reciprocal”. As a manipulation check for the meaning of these reported correlations, each participant’s SUDS range for each condition is reported in the direction in which they varied.



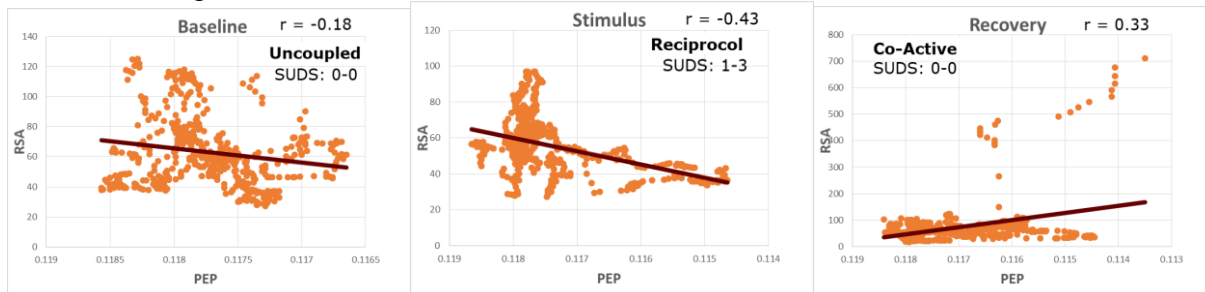
Figure 12: RSA-PEP Scatterplots by Condition by Participant

Figure 12a: Participants in the Highest Quartile for Questionnaires Indexing Adaptive Functioning

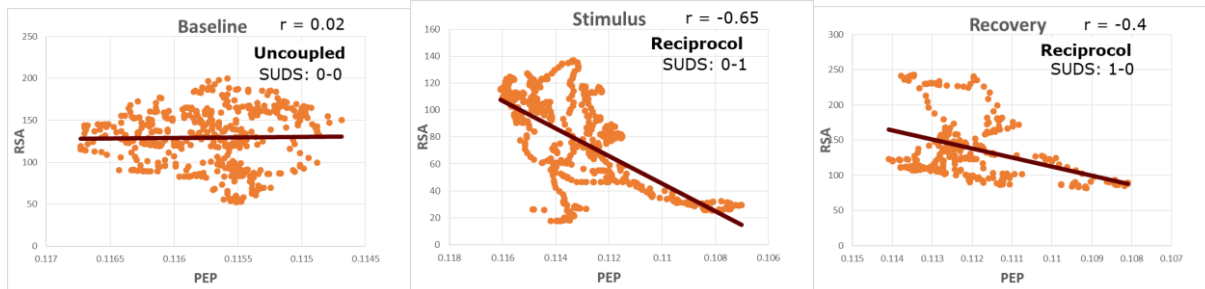
AK9971: High: FFMQ ACQ MAIA-Pre MAIA-Post Low: THS PCL STAI-T PHQ-PD ASI PSWQ



CE3052: High: FSS-BI MAIA Pre MAIA Post : Low: THS PHQ-PD



KT4750: High: FSS-BI ASI MAIA Pre : Low: THS STAI-S PHQ-PD ACQ



LJ0154: High: THS FFMQ ACQ MAIA Pre : Low: PCL STAI-T FSS-BI PHQ-PD ASI PSWQ

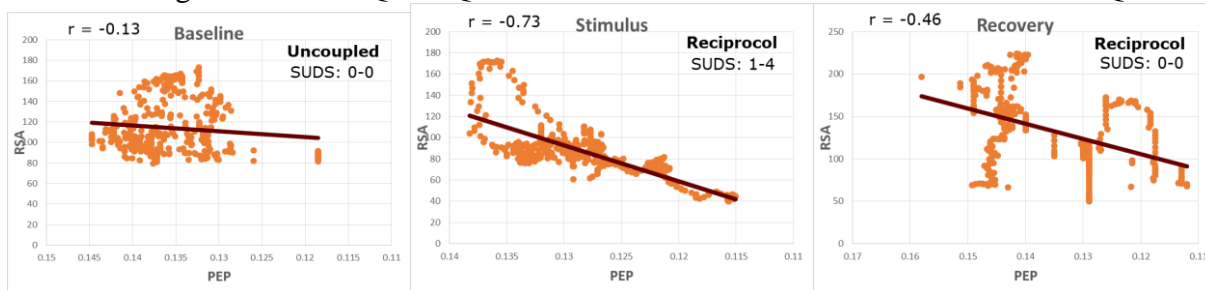
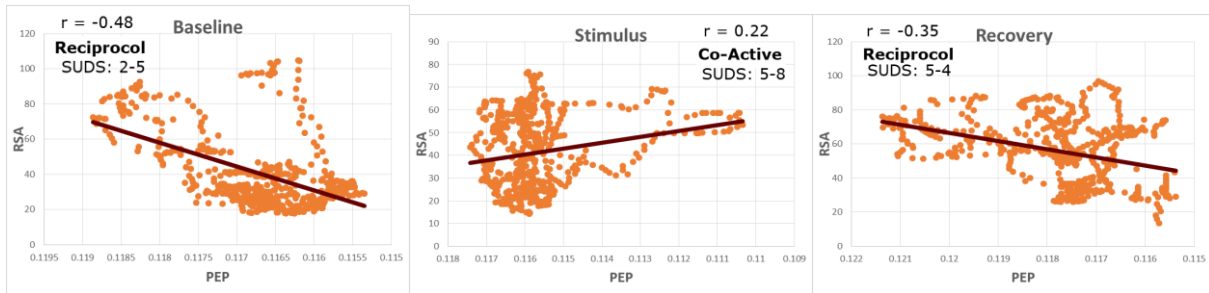


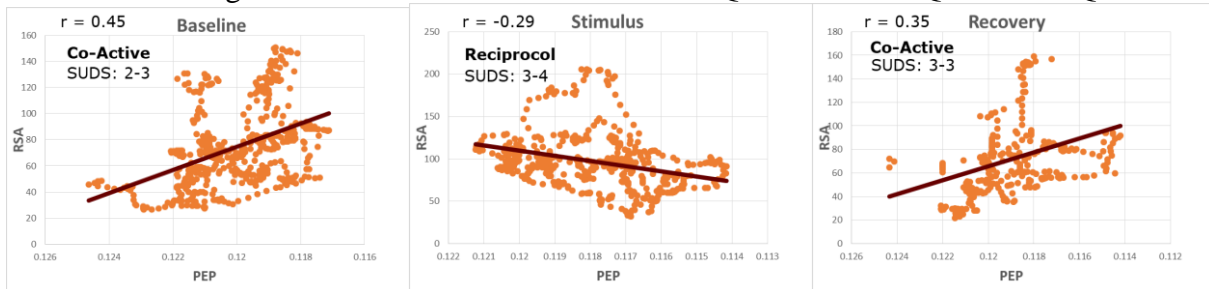
Figure 12 a: RSA-PEP scatterplots by condition for participants who scored mostly in the highest quartile on questionnaires indexing adaptive traits. PEP is shown on reverse scale, and correlation coefficients are multiplied by -1 to reflect directionality of the SNS. All correlations reported were significant at  $p < 0.001$ . Correlations  $> 0.2$  are labelled Co-Active,  $> -0.2$  and  $< 0.2$  Uncoupled, and  $< -0.2$  Reciprocal.

Figure 12b: Participants in the Highest Quartile for Disorder-Related Questionnaires

JA6893: High: PCL STAI-T FSS-BI PHQ-PD ASI PSWQ Low: FFMQ ACQ



RR7686: High: PCL STAI-S STAI-T ASI PSWQ Low: PHQ-PD FFMQ MAIA Post



JS9429: High: PCL STAI-S STAI-T PHQ-PD Low: FSS-BI FFMQ ACQ MAIA Pre MAIA Post

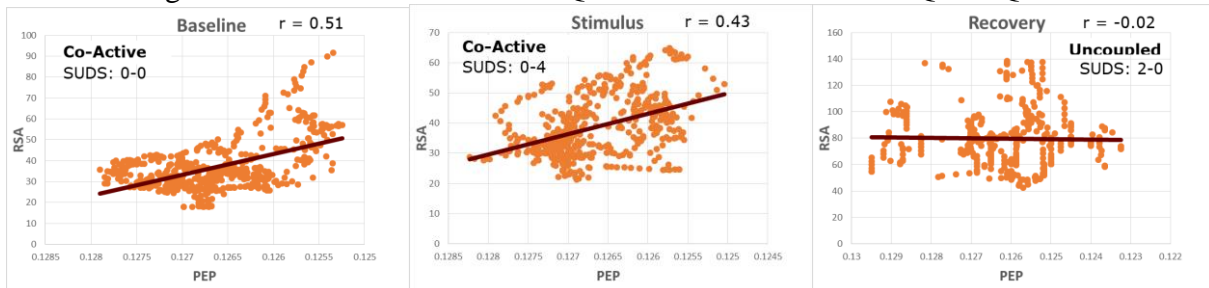
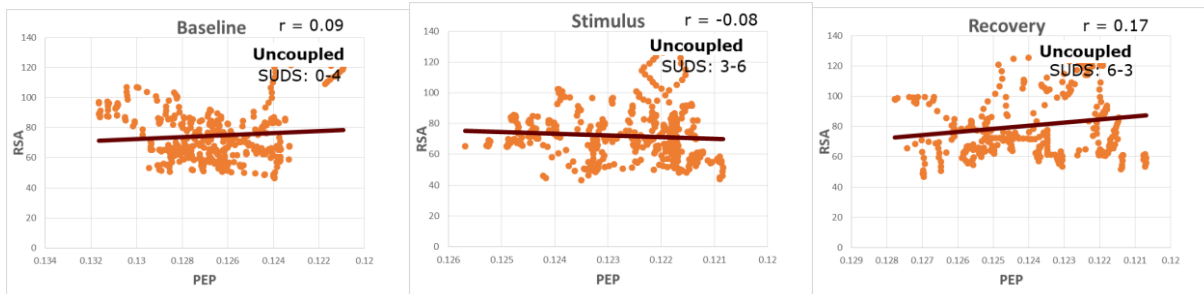


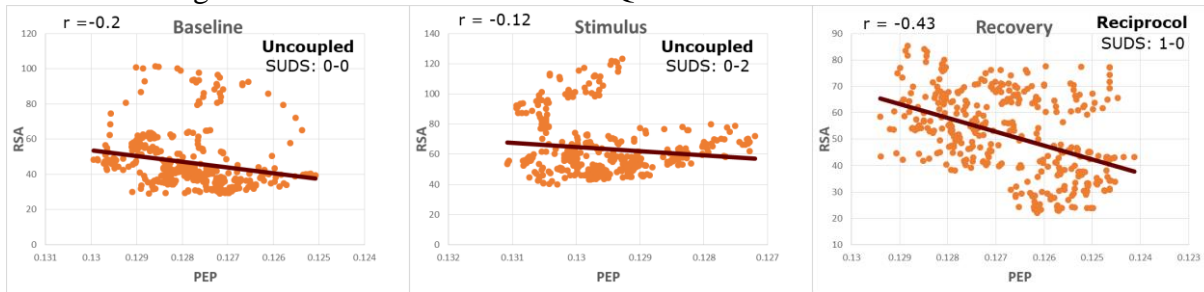
Figure 12 b: PEP-RSA scatterplots by condition for participants who scored mostly in the highest quartile for disorders and the lowest quartile for mindfulness, anxiety control, and interoceptive awareness. PEP is shown on reverse scale, and correlation coefficients are multiplied by -1 to reflect directionality of the PNS-SNS relationship. All correlations reported were significant at  $p < 0.001$ .

Figure 12c: Participants in the Highest Quartile for Dissociative Scores

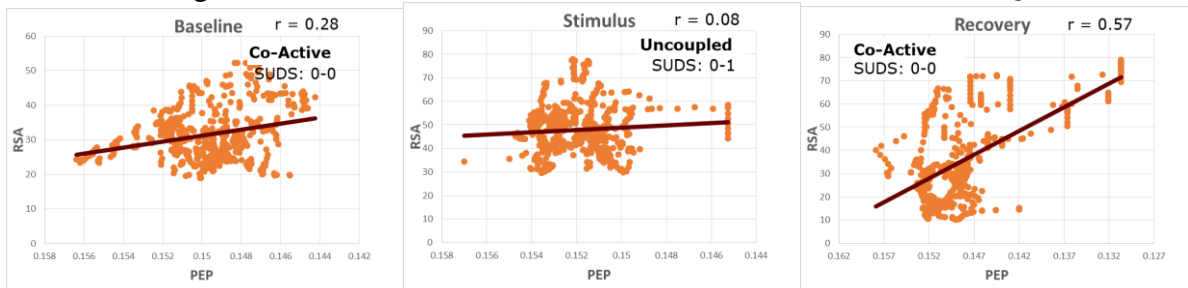
AR8024: High: THS PCL PHQ-PD DISS : Low: STAI-S



OL3760: High: THS PCL DISS Low: PHQ-PD



RC7107: High: THS DISS MAIA Pre MAIA Post : Low: FSS-BI PHQ-PD



AT4799: High: STAI-S STAI-T DISS : Low: PHQ-PD

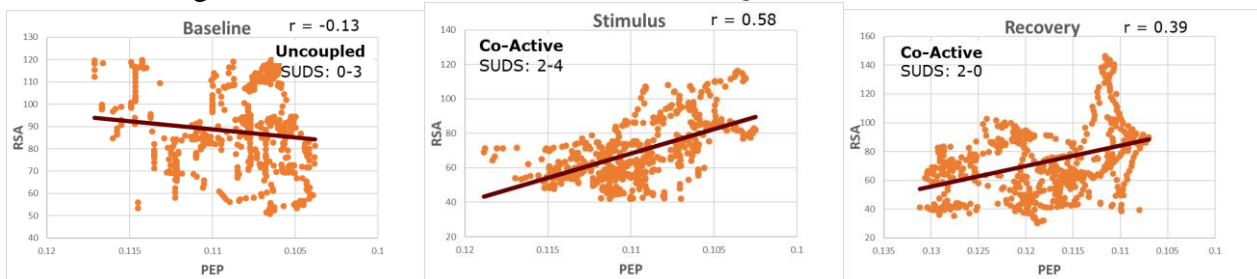


Figure 12 c: PEP-RSA scatterplots by condition for participants who scored mostly in the highest quartile for disorders and the lowest quartile for mindfulness, anxiety control, and interoceptive awareness. PEP is shown on reverse scale, and correlation coefficients are multiplied by -1 to reflect directionality of the PNS-SNS relationship. All correlations reported were significant at  $p < 0.001$ .

The graphs in Figure 12 finally bring into clearer focus what the graphs in Figure 11 were alluding to – each participant’s pattern of ANS activation is dynamic and most of them are surprisingly unique. Nonetheless, with the clarity that Figure 12 brings, it is now possible to review ANS activation for patterns across participants. Two small themes emerge. First, viewing all participants from the “adaptive” category in Figure 11a, the “Uncoupled” model of ANS activation appears to dominate during the baseline period. In addition, all but one participant in the “dissociative” group also showed this dimension. There was no such discernable pattern in the high disorder group.

This uncoupled baseline pattern provides evidence for the hypothesis proposed in the introduction, that uncoupled activation is not an “intermediate” between reciprocal and coactive states. Rather, it appears to be engaged in relatively low threat situations. Further, this provides an interesting revision to the idea that PNS dominance during low threat situations is a reciprocal dominance. This introduces a nuance not directly addressed in Berntson et al. (2008) which is the idea of two different modes for how PNS dominance might look. The traditionally-conceptualized version shows PNS dominance reciprocally suppressing SNS levels. Returning to our participant CE3052’s graph in Figure 11d, one can see largely PNS “dominance”, but the Figure 12a graph confirms that this dominance occurs regardless of the underlying SNS activation. The nearly flat trend line demarcates “high” and “low” levels of RSA, and one can see that the accompanying PEP levels span the range from low to high.

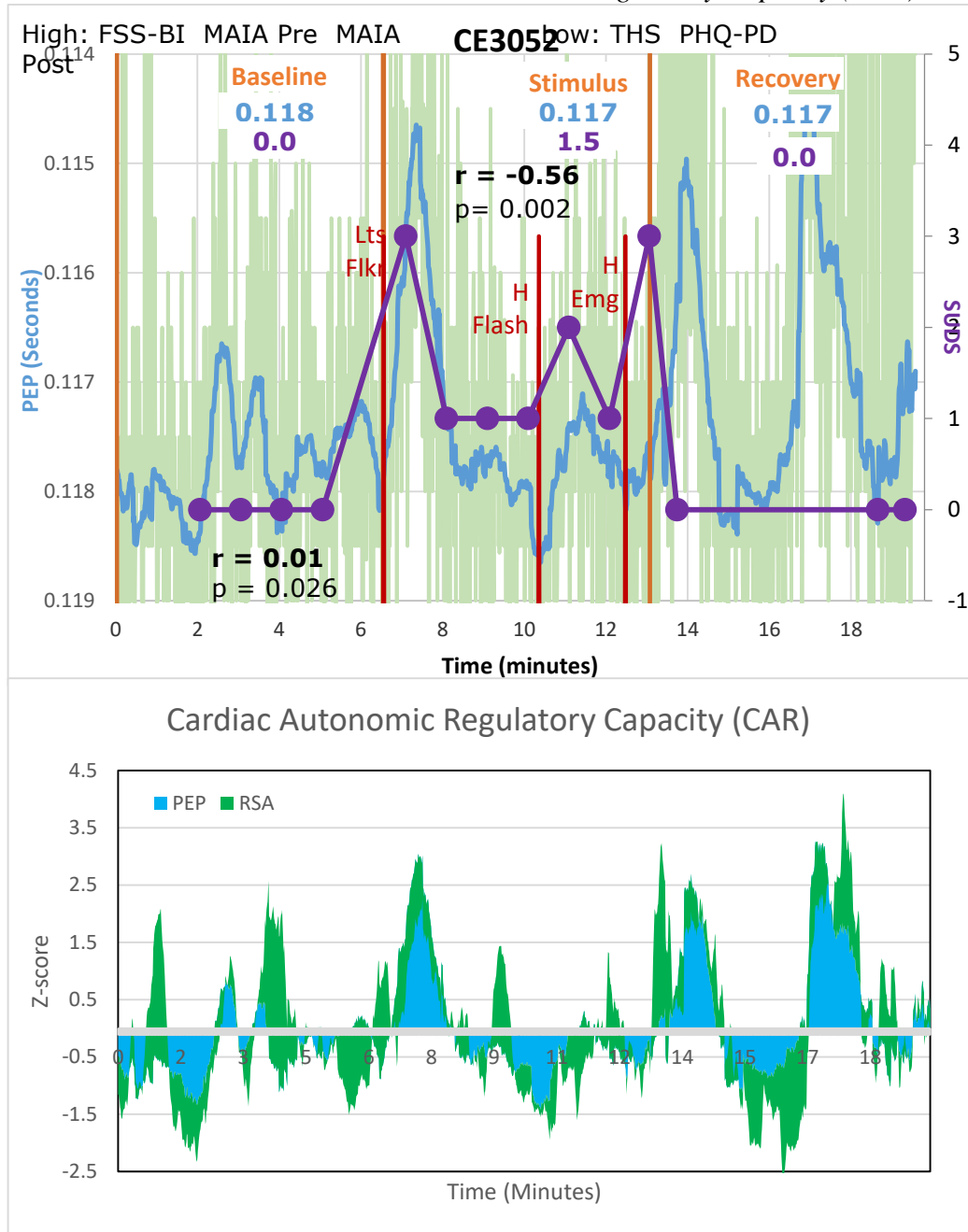
The dissociative group showed one more interesting trend: all but one showed a surprisingly uncoupled PNS-SNS pattern during the expectedly challenging stimulus period. This did not seem to be affected by SUDS ratings, as the highest SUDS reporter in this group showed one of the two lowest PNS-SNS correlations:  $r = -0.08$ . Beyond this observation, few

other patterns come clear across participants, and their unique ANS strategies seem to be the strongest take-away.

The apparently dynamic nature of the graphs in Figure 12 inspires the possibility of viewing the ANS patterns in a time series. Revisiting the research from Berntson et al. (2008), Figure 19 takes the autonomic space concept and adds two novel features; it combines the concept of CAB and CAR into one visual, and then plots it as a time series. The original methodological approach to calculating CAB and CAR prescribed normalizing the SNS and PNS indices by taking their Z-scores. The foundational research used PEP as the SNS index and HF-HRV as the PNS index. HF-HRV worked for their purposes, since they were calculating a simple mean number over time. However, in this research, RSA is substituted for HF-HRV so that a time series analysis is possible.

The top graph in Figure 13 is basically a reprint of the graph from Figure 9a that was discussed previously, using the textbook participant CE3052. This graph provides the anchor points of the conditions, events, and SUDS reports to help inform the dynamics of the bottom graph.

Figure 13: PEP-SUDS Time Series and Cardiac Autonomic Regulatory Capacity (CAR)



Cardiac Autonomic Regulatory Capacity (CAR) reflecting the summed Z-scores for PEP and RSA. Cardiac Autonomic Balance (CAB) is visualized by the unique contributions of PEP and RSA to the overall peaks and valleys shown in the graph.

Cardiac Autonomic Regulation (CAR; also known as Cardiac Autonomic Regulatory Capacity) is displayed in the bottom graph of Figure 13 as the sum of the Z-scores for PEP and RSA (again, PEP is multiplied by negative 1 to convey accurate directionality of the SNS it

indexes). The overall peaks and troughs of this graph represent the combined effect of both SNS and PNS: CAR. Within those peaks and troughs, PEP and RSA can still be visualized indicating the relative proportions of SNS and PNS (indicating the Cardiac Autonomic Balance: CAB).

Several of the ANS CAR peaks and troughs during the baseline appear to be strictly driven by PNS tone, reflected in the undiluted green hue of these features. However, there are other features in the baseline period that show nearly an equal proportion of blue SNS (PEP). This dynamic range of autonomic interactions for the baseline period contrasts sharply with the singular characterization of this period as “Uncoupled” in CE3052’s Baseline graph in Figure 18a. Again, times series can reveal what averages cannot.

The stimulus period provides another opportunity to map ANS dynamics back to responses theorized in the defense cascade. The stimulus period starts with a sudden loud sound, hoping to induce a freeze and orienting response. Figure 19 reveals a strong SNS-PNS coactivation in response. The tall peak immediately following the light flickering is composed of more blue SNS than green PNS, but still plenty of both. Beyond this initial freeze response, it does not appear as though even the “flight” stage was achieved, as CAR and PEP individually dropped after this and did not return to such levels until the recovery period.

Stepping away from the defense cascade model, it should be noted that the most extreme features throughout the study appear to show coactivity: when blue SNS was peaking upward, green PNS was moving with it, adding a modest, but measurable green to the height of each ANS “mountain”. Referring back to Figure 18a, this netted out to an averaged “Reciprocal” activation designation during the overall stimulus.

The recovery period introduced some of the most dramatic peaks and troughs of the entire experience for CE3052. It is unclear what precipitated these responses, although a review

of the video revealed that the moments of above-average activation corresponded with required social interactions with the opposite sex researcher to answer questions about SUDS and to go through the debriefing. A best guess might be that the participant reacted autonomically to these interactions. Without the benefit of questionnaires indexing social anxiety, speculation about social sensitivities will be reserved. The CAR trough in the middle of the recovery period did however, happened during the non-social time during which the participant was completing the MAIA-Post survey on the computer. The significant contribution of both SNS and PNS to the peaks and troughs netted out to a rating of “Coactive” for the recovery period with a sizable  $r = 0.3, p < 0.001$ .

Reviewing this time series CAR graph, one cannot help but sense that something of Berntson’s original conceptualization of CAR is missing, even though his formula was faithfully reproduced for this analysis. Because the Z-score transformation was needed to make PEP and RSA units comparable to one another, the graph in Figure 19 is anchored around the average Z-score of zero, which is really just the mean of each index reset to zero using standard deviation units.

Berntson’s original conceptualization of CAR was a number that indicated magnitude of collective autonomic effort, by combining the power from SNS and PNS influences. But the Z-score transformation about the mean ensures that roughly half of any data set will show as below average and the other half will show as above average. It is not clear if this accurately reflects the person’s general daily mean. The lowest Z-score within the study itself might actually be reflective of high overall ANS power for that person. It could be more informative to calculate Z-scores using the pre-study values.



Therefore, a pre-study analysis was conducted, using the roughly 20 minutes of pre-baseline physiological data that had already been gathered from the participant. The challenge with this approach became clear, as Z-scores are calculated from the mean and standard deviation for each variable. The pre-baseline mean was similar to the study mean, but the pre-baseline variability as measured by standard deviation was strikingly different. PEP standard deviation was 75% greater during the period before the study began ( $SD = 0.0035$ ) as it was during the study itself ( $SD = 0.0020$ ). Comparatively, RSA standard deviation was only 18% larger during the pre-study period ( $SD = 59.9$ ) vs. study period ( $SD = 50.8$ ). Because standard deviation is a critical component of Z-score calculations, pre-study values for baseline Z-score setting actually would have made the resulting study CAR values less variable. Further investigation is warranted to find a time series approach that resembles a more cardinal number measurement of CAR, rather than deviation-about-the-mean approach.

## Discussion

The aim of this study was to clarify the temporal physiological dynamics of the defense cascade. Based on previous studies, I hypothesized that freeze and fright stage would show ANS coactivation and that flight and fight stages would show reciprocal SNS activation. Flag and faint stages were not targeted.

Analysis of the stimulus period provided no clear patterns of ANS activation across the participants, making general conclusions impossible. An unexpectedly different picture emerged: patterns of autonomic activation appeared to be almost as unique as the individual. Baseline results showed a trend toward uncoupled PNS activation during the baseline period for participants scoring high on “adaptive” questionnaires. Although this period was designed to capture participants at rest without threat (pre-defense cascade model), results suggested higher levels of SNS than expected. The recovery period showed perhaps the most variability, with participants showing nearly every variation of activation from reciprocal, to coactive, to uncoupled, and well as variation from SNS to PNS dominance.

This individual physiological variation is consistent with the “idiodynamic” approach to cardiac reactivity (Friedman & Santucci, 2003), inspired by the work of Saul Rosenzweig (1986). Accordingly, the analysis for this study had to adapt by examining the data at a much more granular level: that of the individual. This analysis of individual response patterns yielded some of the most valuable insights in the study.

For example, because the defense cascade is a theoretical model, it was challenging to determine “where” in the model participants was at any given moment. Group level analysis only added uncertainty to the analysis, but the individual examination of participant CE3052 yielded some clarity. It appeared that the lights flickering invoked a state akin to freezing for

this participant, based on simultaneously high levels of both CAR and CAB observed in the CAR time series.

Extending Berntson et al.'s (2008) model of Cardiac Autonomic Balance and Cardiac Autonomic Regulatory Capacity to a time series introduced a new tool for assessing temporal dynamics in any study, but was especially useful in the context of defensive behavior. It would be revealing to map every participant's CAR/CAB during the stimulus period to look for similar evidence across participants. Also, an even more fine-grained correlational analysis could be used to unpack the changing picture of ANS strategies in response to the changing VR threat over the course of the stimulus period.

Participants showed significant improvement on two measures of interoceptive awareness from pre-intervention to post-intervention, and they approached significance for a third subscale. Noticing and attention regulation significantly improved and self-regulation approached significance. While it cannot be concluded that the SUDS ratings alone influenced these significant improvements, the findings nonetheless lends support to my hypothesis that SUDS may have inherent therapeutic value.

The SUDS ratings were also beneficial as a research tool – they provided a valuable anchor to nearly every aspect of the analysis. When the physiological data grew murky, SUDS provided clarity. Even when the physiological data were clear, their interpretation was aided by cross-referencing SUDS. For example, SUDS helped in attributing reciprocal activation to a higher levels of distress.

The aggregation analysis yielded some significant albeit confusing results about the extent to which PNS tone was restored following the stimulus period. Extracting any statistical significance from such a small sample size ( $n = 19$ ) is difficult, and by convention allows

cautious generalizations to be offered. However, with the benefit of the qualitative analysis of the time series data to fill in the details, the utility of the averaged data decreased.

Even without averaging across participants, the downside of averaging within a participant became apparent in the PEP vs. SUDS analysis of participant CE3052. This participant went through a dramatic surge in PEP and SUDS ratings in response to the stimulus onset of the sudden flickering, buzzing, and expiration of the lights in the VR experience. A slow recovery followed over the ensuing three minutes, with the PEP SNS index even dropping below average levels, only to be jarred into another PEP-SUDS spike with the sudden appearance of the inkblot monster in the face, its glowing eyes staring down the participant. The quick disappearance of the monster allowed another PEP-SUDS cooldown until finally the monster climbed out of the movie screen and stalked straight for the participant's neck, grunting all the while, producing a final surge of both PEP and SUDS.

The "averaged" effect of this stimulus period was a weak PEP stimulus mean of 0.117 and a SUDS mean of 1.5, static numbers that do not reflect dynamic experience. Although statistically accurate, these values are misleading in terms of understanding the physiological and the emotional levels they are supposed to index. By analogy, the experience of a riding a roller coaster ride cannot be conveyed by reporting the mean height of the tracks. Averaging physiological and self-reported variables obscures the dynamic nature of phenomena such as the defense cascade.

Clinical psychologists who use SUDS ratings to aid exposure therapy sessions would not typically not report an average SUDS rating to characterize the session. The whole session was not likely best summed up with a mean SUDS of "4". It would typically start at a low number,

climb to a much higher number, and hopefully come back down. Those movements are where the work gets done.

The qualitative analysis of the time series data thus provided a clearer picture of the trajectory of each participant. It also conferred the benefit of statistical power far and above that of the cross-sample averages: the number of observations for each participant for each condition averaged around  $n = 400$  for PEP and  $n = 100$  for RSA. The number of SUDS ratings per participant was 14, although feedback from both participants and researchers suggest that future studies would be improved by polling for SUDS twice as often – around once every 30 seconds, especially during the stimulus condition when distressing experiences are coming and going much more quickly.

One final point in favor of time series assessment: they can safeguard against scientific fraud, because time series data are typically harder to fudge and invent than averaged data. For example, data for this study were sampled at 2000 samples per second for most of the raw data channels. At nearly 20 minutes per file, this would require the daunting task of inventing over two million data points per participant. All derived and calculated channels are fully traceable back to the original two million data points per channel.

In examining the changing nature of subjective experiences, Lang's 1967 tripartite model of fear as examined through Allen et al.'s (2015) synchrony study brings an interesting perspective. Lang's tripartite model proposes three main components of fear: a subjective experience, a physiological component, and a behavioral manifestation of avoidance. Allen, et al., (2015) examined the extent to which these three fear components were synchronized, as defined by significant changes in each component before and after a one-session treatment for phobia. Subjective experience (SUDS) and behavioral avoidance dropped significantly, while

the study's measure of physiology (IBI) did not. After the roller coaster of SUDS and PEP values observed in the present study, it is intriguing to imagine how a time series of each participant in the synchrony study might have looked.

Returning to the conceptualization of fear, it is important to recognize that fear is just one of several related emotions that might have been indexed by elevated SUDS and physiology in this study. Anxiety is another emotion that would show elevated SUDS and physiology. Öhman (2008) suggested that fear involves behavioral coping in the actual presence of a threat. By contrast, he suggested that anxiety arises when coping behaviors are not available and when a threat is not actually present. I propose an additional nuance: anxiety carries with it uncertainty about whether a threat will actually materialize.

This definition makes room for a related but distinct emotion: dread. Such an emotion would likely also come with elevated subjective distress and physiology, but dread tends to occur when there is some certainty that a threat will materialize. I speculate that at least some participants were experiencing more dread than fear or anxiety: they knew they had signed up for a study about physiology under threat; they just had to sit and wait for the threat to appear.

Somewhat relatedly, a body of research into positive emotions shows that the anticipation of reward is more reinforcing than the reward itself (e.g., Knutson et al., 2008). Whether this also applies to the anticipation of aversive states is speculative but intriguing. Perhaps the anticipation of threat in a state of dread is more distressing than the threat itself. In this case, uncertainty could certainly add to the distress. The comment from one participant adds color to this idea. When asked overall how distressing the experience was the participant replied, "I thought it was going to be worse."

Beyond the restrictions imposed by the pandemic lockdown, this study had some limitations on the generalizability of the results. First, the accelerometer was placed on top of the head in hopes of capturing postural changes, but subsequent behavior analysis suggested that head movements corresponded more with curiosity than with fear-driven responses. The sternum would be a better location to affix the transducer for an accelerometer.

Each participant's state of distress before baseline began was not determined. Because SUDS became such a guide throughout the study, it would have been helpful to determine a SUDS level before the study got underway. This could have helped draw more precise conclusions about the ANS patterns reviewed in the correlational analysis.

The lack of consistency in the recovery period made it more difficult to generalize about changes from stimulus to baseline. Ideally, a protocol should be developed to ensure that the recovery period was of equal duration to the baseline and stimulus periods, ensuring time consistency across epochs. Time series data can be biased when there is unequal epoch sampling across subjects (Porges & Bohrer, 1991).

Dissociation was an independent variable of interest in this study, but the only measure used to track this was the reported dissociation experiences from the THS. The experience of having dissociated in the past has been shown to be a predictor of dissociative tendencies (Ross, Joshi, & Currie, 1991), but a more direct measure of dissociative tendencies might be a better way to index dissociation. Replications would be needed that included questionnaires dedicated to dissociation, such as the Dissociative Experiences Scale – II (Carlson & Putnam, 1993).

In designing the study, there was a concern that each request for participant SUDS ratings would be a significant distraction from the stimulus. In fact, participants did not appear to struggle with these requests. Further, this participant "task" seemed to help them stay more

engaged with the experience, which was needed especially for the reportedly “draggy” baseline experience. Because distraction concerns about the SUDS did not materialize, it would have been desirable to increase the number of SUDS data points. Debriefing inquiries with the participants suggested that SUDS could be requested every 30 seconds, rather than every 60 seconds, to obtain more real-time information about the participant experience.

Also, the actual SUDS prompts remained on the screen for ten seconds; both participants and researcher assistants agreed that this time was too long. This interval was so long that several participants changed their ratings before the end of the ten seconds to respond to changing stimulus conditions in the VR. Future studies would benefit from a maximum 5 second SUDS prompt.

SUDS ratings were not conceived as a number that was meant to be averaged over time. Nonetheless, it was averaged in the present study to provide consistency and comparability with the similarly averaged physiological measures of PEP, HF-HRV, and RSA.

Although this study was conceived as having two measures of SNS activation, the second measure, EDA was not calibrated correctly during data collection to allow it to provide a full time series. Phasic responses have the potential to be extracted but were omitted due to their limited benefit for this analysis. An EDA time series would have provided another estimate of SNS.

The stimulus itself was a multidisciplinary achievement, but it can be improved in future studies. The animation of the inkblot monster lacked a certain biological realism and did not provide a sufficiently threatening experience for several participants, as indexed by their maximum SUDS ratings of 1. The participants were part of an age cohort that has been raised on the horror genre as an entertainment staple. Expectations for fearful stimuli were therefore high,



especially in light how the study was advertised. For many participants, the actual experience did not produce the degree of threat that was targeted.

The advertisement for the study may have inflated participant expectations of the feared stimulus beyond what was delivered. (Future development is underway to increase the threat potency of the VR stimulus.) After 17 minutes of high fear expectations, when the inkblot monster finally emerged and perhaps disappointed in his lack of intimidation, it is likely that some horror-acclimated participants actually experienced disappointment or perhaps even amusement. Others less accustomed to horror may have experienced relief. A few additional follow-up questions could have helped determine how this played out, and a question about participant's prior exposure to and affinity horror films could help. Relatedly, one participant remarked to the researchers that she "enjoys [her] nightmares". Not surprisingly, her SUDS ratings were one of the lowest of the participants. Future related studies could also ask how much the person feared the experience before, how fearful it actually made them, what they felt at the conclusion of the study, in hindsight (relief, disappointment, annoyance, etc.).

One final stimulus-related limitation is that the threat may not have escalated at the same pace as an evolutionary threat would. In preparation for this study, I conducted a qualitative review of documentaries depicting predator attacks – most encounters were over within less than a minute. The challenge for this study was thus a methodological one: most PNS indices such as HF-HRV and RMSSD are recommended to consist of a minimum of 5 minutes for accurate calculations comparisons across studies (Laborde, Mosley, & Thayer, 2017; Malik, 1996; Shaffer & Ginsberg, 2017). The cinema consultant is to be commended for increasing the duration of a one-minute threat while finding a way to make it sufficiently fear-inducing for most participants.

This was the first known study to explore the temporal dynamics of physiological measures of the defense cascade (DC) using metrics that are known to differentiate sympathetic from parasympathetic influences. This study introduced a number of additional novel features that can further the precision with which defense cascade and psychophysiological research proceeds. First, it expanded the ANS measures of RSA and PEP to a time series format, allowing significantly more information to be conveyed about the experience of each participant. The use of participant-level analysis augmented this process. Also, this study introduced a time series approach to the CAR/CAB framework (Berntson et al., 2008).

Another enhancement to the physiological time series was the addition of a time series of participant mobility. Accelerometry time series can simultaneously provide a manipulation check on the participant response to a stimulus, a behavioral report to temper apparently extreme physiological readings (movement can create noisy data), and a dependent variable in its own right. Dynamic mobility data can indicate critical stage transitions in the defense cascade. A modification was recommended to this study's application of the accelerometer equipment: measuring accelerometry from the sternum rather than the top of the head would better insulate mobility estimates from head movement related to visual exploration rather than the desired stimulus-related postural changes.

A custom threat was designed for this study using virtual reality. This VR stimulus demonstrated the ability to generate stronger physiological responses, facilitated by the superior ecological validity of VR. Further, the immersion created by the placement of the VR headset, ensured no visual distractions from non-stimulus features in the environment.

Finally, the video capture procedure in this experiment introduced a novel approach to the simultaneous analysis of physiological and behavioral variables. Video capture alone is not

new in psychological research, but creating a recording that simultaneously displays both real-time streaming physiology and participant behavior is an innovation. To complete the picture, the stimulus imagery that the participant was experiencing was also part of the video feed, being continuously screen casted directly behind the participant, but in view to only the researchers, thanks to the participant VR headset.

This advance in video capture provided behavior information to better interpret several elements of this study. First, it allowed for the observation that periods of high participant mobility typically coincided with exploration of the VR environment, rather than the originally-intended flight or fight mobility for the study. Second, erratic variations in the voltage in the ECG channel were cross-referenced to be movement artifact during the lights flickering stimulus. Relatedly, video review indicated that the light flicker stimulus acted as a jump scare for many participants, revealing the unexpected degree of potency of this stimulus. Finally, the video provided a behavior check on apparently aberrant respiration patterns. For example, several respiratory cycles seemed impossibly long, extending 3 to 4 times longer than the average cycle time. Review of the video revealed the participant attempting to stifle a yawn. Other unusually short respiration cycles coincided with the participant talking aloud or coughing. Relatedly, yawning and coughing can produce artifacts in the respiration that should be cleaned before RSA calculation, but this step was not taken for this analysis. Future studies leveraging RSA should ensure the removal of such artifacts.

The time series from this study provided a valuable picture into the moment-to-moment changes in the participant physiology during a threat stimulus. The use of SUDS ratings provided the anchor to this time series information. The most meaningful results from this research can be summed up in a word: variability. There was vastly more variability across

participants than was hypothesized. There was also more variability within a participant's response over time than that predicted in the defense cascade model, and no participant showed a clean replication of the autonomic predictions from the defense cascade. While variability can sometimes confound the research process, it is a hallmark of human adaptability. It is also what makes the roller coaster worth the ride.

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**Appendix A**Mind-Body Laboratory Health History Questionnaire (HHQ)

A very brief medical history must be obtained as part of the experimental protocol. It is very important that you be completely honest. This information will be kept strictly confidential.

1. What is your age, height, weight, and gender?

Age: \_\_\_\_\_ years

Height: \_\_\_\_\_ feet, \_\_\_\_\_ inches

Weight: \_\_\_\_\_ pounds

Sex: \_\_\_M \_\_\_F

2. Since birth, have you ever been hospitalized or had any major medical problems?

\_\_\_ Yes \_\_\_ No

If Yes, briefly explain:

3. Have you ever experienced a concussion or lost consciousness due to a blow to the head?

\_\_\_ Yes \_\_\_ No

If Yes, briefly explain:

4. Have you ever had problems that required you to see a counselor, psychologist, or psychiatrist?

\_\_\_ Yes \_\_\_ No

If Yes, briefly explain:

5. Do you use tobacco products of any kind?

\_\_\_ Yes \_\_\_ No

If Yes, describe what kind how often/much:

6. Have you ever been diagnosed with a psychological disorder?

\_\_\_ Yes \_\_\_ No

If Yes, briefly explain:

7. Do you currently have or have you ever had any of the following?

- Yes  No Strong reaction to cold weather  
 Yes  No Circulatory problems  
 Yes  No Tissue disease  
 Yes  No Skin disorders (other than facial acne)  
 Yes  No Arthritis  
 Yes  No Asthma  
 Yes  No Lung problems  
 Yes  No Cardiovascular disorder/disease  
 Yes  No Auditory deficiency that noticeably affects your ability to hear  
 Yes  No Tinnitus (ringing in your ears)  
 Yes  No Pulsatile tinnitus (hearing “whooshing” sound in timing w/ your pulse)  
 Yes  No Diabetes  
 Yes  No Hypoglycemia  
 Yes  No Hypertension (high blood pressure)  
 Yes  No Hypotension (low blood pressure)  
 Yes  No Hepatitis  
 Yes  No Neurological problems  
 Yes  No Epilepsy or seizures  
 Yes  No Brain disorder  
 Yes  No Stroke

If you responded Yes to any of the above conditions, briefly explain:

8. Have you ever been diagnosed as having:

- Yes  No Learning deficiency or disorder  
 Yes  No Reading deficiency or disorder  
 Yes  No Attention deficit disorder  
 Yes  No Attention deficit hyperactivity disorder  
 Yes  No Autism spectrum disorder or Asperger syndrome

9. Have you ever been diagnosed with:

- Yes  No Claustrophobia (extreme fear of small closed spaces)  
 Yes  No Blood phobia (extreme fear of needles or blood)  
 Yes  No Fear of medical settings (e.g. hospital or doctor)  
 Yes  No Health anxiety (extreme fear of serious but undiagnosed medical condition)  
 Yes  No Phobia of any type (if Yes, briefly explain:)  
 Yes  No Generalized anxiety disorder  
 Yes  No Social anxiety disorder  
 Yes  No Post-traumatic stress disorder  
 Yes  No Panic disorder  
 Yes  No Obsessive Compulsive Disorder  
 Yes  No Anxiety disorder of any type (if Yes, briefly explain:)

If you responded Yes, briefly explain here:

10. Have you ever been diagnosed with:

Yes  No Major depressive disorder

Yes  No Bipolar disorder

Yes  No Seasonal affective disorder

Yes  No Affective disorder of any type

If you responded Yes, briefly explain here:

11. Do you currently take selective serotonin reuptake inhibitors (SSRI's)? (Some examples: Prozac, Paxil, Zoloft, Luvox, Lexapro, Celexa).

Yes  No

If you responded Yes, briefly explain here:

12. Do you currently take hormonally-based contraception (including birth control pills, skin patches, or vaginal rings)?

13. List any over-the-counter or prescription medications you are currently taking:

14. If any medications are listed in Question 13, list the symptoms that these drugs are treating

15. List any other medical conditions that you have or have had in the past:

16. What is your average daily caffeine consumption (approximate number of cups/glasses of coffee, tea, or caffeinated soda)?

17. What is your average weekly alcohol consumption (approximate number of alcoholic beverages)?

18. How many hours of sleep do you average per night?

19. On average, how often do you engage in physical activity for at least 30-minute sessions? (Circle one)

a- Never;      b- Rarely;      c- One to two times per month;      d-One to two days per week;  
e-Three to four days per week;      f-Five to six days per week;      g-Seven days per week

20. Do you regularly use a FitBit or other similar device to keep track of your heart rate?

21. Have you ever fainted? If so, explain. (When, what was likely to have caused it, how often does this occur?)

## Appendix B

Scores on certain questionnaires strongly correlated with participant PEP variance across conditions. Participants scoring in the top quartile on the Trauma History Screen (THS) showed the greatest difference in PEP changes versus participants scoring in the bottom THS quartile, with High THS participants showing more robust drop in PEP from B-to-S and a more robust rebound in PEP from S-to-R. The THS produced a larger High-vs.-Low quartile difference than all other questionnaires and for their respective High-Low splits. The Penn State Worry Questionnaire (PSWQ) and the post-stimulus Multidimensional Assessment of Interoceptive Awareness (MAIA) also produced large High-Low quartile disparities in PEP condition changes. See Table B1 for details.

Questionnaires that most effectively created a dispersion in RMSSD variance included the Fear Survey Schedule – Blood Injury subscale (FSS-BI), the pre-stimulus MAIA and the post-stimulus MAIA (Table B2), with those in the top quartile for all three questionnaires showing RMSSD dropping from B-to-S. While this same high quartile group increased their RMSSD from S-to-R, their increase was significantly less than that of the low quartile group.

Questionnaires creating the largest dispersion of RSA variance were the Penn State Worry Questionnaire and the post-stimulus MAIA (Table B3) although their effects were in the opposite direction. The high worry group increased RSA from B-to-S, while the low worry group decreased for this condition. Both groups increased RSA from S-to-R, but the low worry group increased by a significantly larger margin. Those scoring in the top quartile on the post-stimulus MAIA expectedly decreased RSA from B-to-S, while their bottom quartile counterparts increased for this condition. Both groups increased RSA from S-to-R, but the high post-stimulus interoceptive group increased by a significantly larger margin than their low scoring counterparts.

Questionnaires most strongly dispersing HF-HRV FFT and HF-HRV AR variance were the FSS-BI, the Anxiety Sensitivity Index, and the post-stimulus MAIA (Table B4 and B5). The

high fear and anxiety groups showed an expected reduction in HF-HRV measures from B-to-S while their low fear/anxiety counterparts showed an unexpected increase. Both high and low fear/anxiety groups showed an unexpected decrease in both measures of HF-HRV from S-to-R, with the low fear/anxiety group decreasing significantly more. Both top and bottom quartile scorers on the post-stimulus MAIA showed an unexpected increase in HF-HRV measures from B-to-S, and an unexpected decrease from S-to-R, with the low quartile scorers showing a significantly more robust response in both conditions. Additionally, for the HF-HRV AR measure, the State-Trait Anxiety Index – State subscale (STAI-S) dispersed results: those with high state anxiety increased HF-HRV AR from B-to-S, while the low anxiety decreased in the same condition. Both groups decreased in the S-to-R condition, with the high anxiety group decreasing measurably more.

The State-Trait Anxiety Index produced the strongest spread in SUDS responses between high- and low-scoring participants (Table B6) with high anxious participants showing an average of 1.2 additional SUDS increase from B-to-S over low anxious participants and an average of 1.7 SUDS additional decrease from S-to-R.



Table B1: PEP Changes in High and Low Quartile Participants

			THS	PCL	STAI-S	STAI-T	FSS-BI	PHQ-PD	ASI	PSWQ	FFMQ	ACQ	MAIA Pre	MAIA Post
HI-LO	B-S		2.2%	0.5%	1.5%	1.4%	0.9%	0.9%	0.5%	1.8%	1.2%	1.4%	0.3%	1.1%
Difference	S-R		<u>5.5%</u>	<u>1.4%</u>	<u>2.5%</u>	<u>0.2%</u>	<u>2.3%</u>	<u>2.2%</u>	<u>0.8%</u>	<u>3.4%</u>	<u>1.4%</u>	<u>0.1%</u>	<u>1.4%</u>	<u>3.2%</u>
Total Difference:			<b>7.8%</b>	<b>2.0%</b>	<b>4.0%</b>	<b>1.6%</b>	<b>3.1%</b>	<b>3.0%</b>	<b>1.3%</b>	<b>5.2%</b>	<b>2.6%</b>	<b>1.5%</b>	<b>1.7%</b>	<b>4.3%</b>

Table B2: RMSSD Changes in High and Low Quartile Participants

			THS	PCL	STAI-S	STAI-T	FSS-BI	PHQ-PD	ASI	PSWQ	FFMQ	ACQ	MAIA Pre	MAIA Post
HI-LO	B-S		6%	24%	24%	23%	14%	8%	6%	33%	19%	6%	26%	38%
Difference	S-R		<u>128%</u>	<u>92%</u>	<u>35%</u>	<u>12%</u>	<u>447%</u>	<u>188%</u>	<u>63%</u>	<u>79%</u>	<u>19%</u>	<u>146%</u>	<u>588%</u>	<u>524%</u>
Total Difference:			<b>134%</b>	<b>116%</b>	<b>59%</b>	<b>36%</b>	<b>461%</b>	<b>196%</b>	<b>69%</b>	<b>112%</b>	<b>38%</b>	<b>152%</b>	<b>614%</b>	<b>561%</b>

Table B3: RSA Changes in High and Low Quartile Participants

			THS	PCL	STAI-S	STAI-T	FSS-BI	PHQ-PD	ASI	PSWQ	FFMQ	ACQ	MAIA Pre	MAIA Post
HI-LO	B-S		1%	27%	13%	13%	21%	5%	8%	21%	23%	9%	30%	29%
Difference	S-R		<u>7%</u>	<u>9%</u>	<u>8%</u>	<u>7%</u>	<u>0%</u>	<u>1%</u>	<u>7%</u>	<u>39%</u>	<u>15%</u>	<u>4%</u>	<u>3%</u>	<u>28%</u>
Total Difference:			<b>8%</b>	<b>37%</b>	<b>20%</b>	<b>19%</b>	<b>21%</b>	<b>5%</b>	<b>15%</b>	<b>60%</b>	<b>38%</b>	<b>13%</b>	<b>33%</b>	<b>58%</b>

Table B4: HF-HRV FFT Changes in High and Low Quartile Participants

			THS	PCL	STAI-S	STAI-T	FSS-BI	PHQ-PD	ASI	PSWQ	FFMQ	ACQ	MAIA Pre	MAIA Post
HI-LO	B-S		21%	2%	25%	8%	50%	11%	49%	13%	37%	1%	11%	23%
Difference	S-R		<u>3%</u>	<u>23%</u>	<u>18%</u>	<u>28%</u>	<u>26%</u>	<u>29%</u>	<u>23%</u>	<u>13%</u>	<u>0%</u>	<u>19%</u>	<u>5%</u>	<u>29%</u>
Total Difference:			<b>24%</b>	<b>25%</b>	<b>43%</b>	<b>36%</b>	<b>76%</b>	<b>41%</b>	<b>72%</b>	<b>27%</b>	<b>37%</b>	<b>20%</b>	<b>17%</b>	<b>52%</b>

Table B5: HF-HRV AR Changes in High and Low Quartile Participants

			THS	PCL	STAI-S	STAI-T	FSS-BI	PHQ-PD	ASI	PSWQ	FFMQ	ACQ	MAIA Pre	MAIA Post
HI-LO	B-S		4%	0%	25%	14%	22%	8%	11%	18%	6%	13%	3%	29%
Difference	S-R		<u>6%</u>	<u>2%</u>	<u>12%</u>	<u>6%</u>	<u>19%</u>	<u>9%</u>	<u>26%</u>	<u>4%</u>	<u>8%</u>	<u>2%</u>	<u>2%</u>	<u>19%</u>
Total Difference:			<b>10%</b>	<b>3%</b>	<b>38%</b>	<b>20%</b>	<b>41%</b>	<b>17%</b>	<b>37%</b>	<b>22%</b>	<b>14%</b>	<b>15%</b>	<b>6%</b>	<b>48%</b>

Table B6: SUDS Changes in High and Low Quartile Participants

			THS	PCL	STAI-S	STAI-T	FSS-BI	PHQ-PD	ASI	PSWQ	FFMQ	ACQ	MAIA Pre	MAIA Post
HI-LO	B-S		0.2	0.5	1.2	0.8	0.1	0.6	0.5	0.3	0.0	0.5	0.4	0.2
Difference	S-R		<u>0.4</u>	<u>0.0</u>	<u>1.7</u>	<u>0.8</u>	<u>0.0</u>	<u>0.0</u>	<u>0.2</u>	<u>0.6</u>	<u>0.1</u>	<u>0.6</u>	<u>0.1</u>	<u>0.8</u>
Total Difference:			<b>0.6</b>	<b>0.6</b>	<b>2.9</b>	<b>1.6</b>	<b>0.1</b>	<b>0.6</b>	<b>0.7</b>	<b>0.8</b>	<b>0.1</b>	<b>1.0</b>	<b>0.5</b>	<b>0.9</b>

Tables B1 - B6: Comparison of the highest and lowest quartile participants with respect to their averaged changes in the noted variable from the Baseline to Stimulus period (B-S) and from the Stimulus to Recovery period (S-R). The overall change accounted for by each variable was calculated by summing the absolute value of the changes.