Neural Correlates of Threat and Emotion Processing in Panic Disorder

by

Isabel Taake

M.A., Simon Fraser University, 2009

B.A., Simon Fraser University, 2007

Thesis Submitted in Partial Fulfillment of the

Requirements for the Degree of

Doctor of Philosophy

in the

Department of Psychology

Faculty of Arts and Social Sciences

© Isabel Taake 2018

SIMON FRASER UNIVERSITY

Summer 2018

Approval

Name:	**Isabel Taake**
Degree:	**Doctor of Philosophy (Psychology)**
Title:	**Neural Correlates of Threat and Emotion Processing in Panic Disorder**
Examining Committee:	**Chair: John McDonald** **Professor**

Mario Liotti
Senior Supervisor
Professor Emeritus

Alexander Chapman
Supervisor
Professor

Urs Ribary
Supervisor
Professor

Maureen Hoskyn
Internal Examiner
Associate Professor
Department of Education

Geoffrey Potts
External Examiner
Associate Professor
Department of Psychology
The University of South Florida

Date Defended/Approved: June 14, 2018

Ethics Statement

The author, whose name appears on the title page of this work, has obtained, for the research described in this work, either:

    a.   human research ethics approval from the Simon Fraser University Office of Research Ethics

or

    b.   advance approval of the animal care protocol from the University Animal Care Committee of Simon Fraser University

or has conducted the research

    c.   as a co-investigator, collaborator, or research assistant in a research project approved in advance.

A copy of the approval letter has been filed with the Theses Office of the University Library at the time of submission of this thesis or project.

The original application for approval and letter of approval are filed with the relevant offices. Inquiries may be directed to those authorities.

Abstract

Early and rapid attention to threat is an adaptive response in healthy individuals; however, in persons with clinical or sub-clinical levels of anxiety, this mechanism appears to be exaggerated and maladaptive. Specifically, anxious individuals seem to be distinctively sensitive to threatening information in the environment, which is called an attentional bias. To better understand the neural mechanisms underlying hypervigilance to threat in clinical and subclinical anxiety, scalp-recorded brain electrical activity (EEG) was recorded while participants performed supraliminal and subliminal versions of an emotional Stroop task where threat and neutral, or positive and neutral words were presented in blocks. Participants responded either to the color of the presented word (supraliminal task), or to the colour of a mask (subliminal task). Study 1 found enhanced early frontal responses (200-320 ms, the EAP) to task-irrelevant threat words relative to neutral words in healthy individuals with high Anxiety Sensitivity (AS) in the supraliminal task, confirming early stage, likely pre-attentive processing of consciously perceived threat information. More interestingly, the high AS group additionally exhibited an enhanced, even earlier frontal effect (starting at 130 ms) in the subliminal Stroop task, indexing pre-conscious processing of threat stimuli below perception threshold. Study 2 extended the analysis of hypervigilance to threat signals to a group of Panic Disorder patients. There was <u>no</u> evidence of early differential frontal ERP modulation to threat words (EAP and P150) in either the supraliminal or subliminal eStroop task.

We concluded that in high trait anxiety, early frontal effects may index a *hyperactive* early threat detection system located in medial PFC or insular cortex. In Panic Disorder patients, on the other hand, threat information triggering a hyperresponsive amygdala would fail to cause sufficient top-down emotion regulating activity from vmPFC, resulting in the absence of a sizeable early EAP. The inverse correlation between symptom severity and prefrontal activation supports the hypothesis that PFC hypoactivation could be a component of the transition of dispositionally high anxiety to disordered anxiety. We propose that presence or absence of the EAP could provide a marker able to quantify such transition.

Keywords: Attentional bias; Event-Related Potentials; Anxiety; Panic Disorder; Emotional Stroop Task; Subliminal Processing

Dedication

To my parents: Hans-Georg Taake and Doris Taake.

And my children: Nicolas and Leonor

Acknowledgements

First of all, I would like to acknowledge my primary supervisor Dr. Mario Liotti for his mentorship and breadth of knowledge that has shaped this project, and also would like to thank him for his kindness and friendship over the last 11 years during which we worked together. He will be considerably missed at Simon Fraser University, but I wish him great success and all the best for his new position and future endeavours. I would also like to thank the other members of my supervisory committee, Drs. Alexander Chapman, Urs Ribary, Maureen Hoskyn, and Geoffrey Potts for their advice and for their time serving as my examiners.

Thanks to my fellow lab mates of LADN, whose input and help in various ways was instrumental for this project, particularly: Patrick, Killian, Fern, Jen, and Reg. A special big thank you to my friend and brilliant programmer Calen Walshe; without your help the coding and design of my tasks would have been a huge headache. To all the volunteers who contributed to this project and gave their time as participants, the SFU undergraduates, LADN volunteers, and of course, most importantly, the patients who made it up to the SFU campus despite the various challenges they face.

My parents: Without their emotional and financial support, I would have never finished this degree. I owe you everything. To Camilo, my best friend and partner for the last 20 years.

Table of Contents

List of Tables

List of Figures

Preface

The study of how emotion influences perceptual processing and attentional selection has been an increasingly popular topic over the last two decades (LeDoux, 1986; Williams & Gordon, 2007a; Pourtois, Schettino, & Vuilleumier, 2013). To understand the mechanisms of these affective processes in the human brain and how emotionally salient stimuli in the environment undergo preferential selection, and gain access to more elaborate processing and consciousness is of particular interest (Schupp et al., 2004). It helps us understand the dynamic interplay between the environment and the healthy individual, with stimulus detection being governed by both objective and subjective factors, but this knowledge might also shed light on the modulatory effects of individual state-trait characteristics (Carretie, 2014), and on exaggerated or maladapted attentional mechanisms in clinical populations (Lipka, Hoffmann, Miltner & Straube, 2014; Pergamin-Hight, Naim, Bakersmans-Kranenburg, van Ijzendoorn, & Bar-Haim, 2014).

In general, research suggests that humans give precedence to signals of threat or danger when processing incoming environmental information (Smith, Cacioppo, Larsen, & Chartrand, 2003). One of the most important functions of the mechanisms underlying the fear system is *rapid* detection of threatening information in the environment, as it allows the organism to appropriately respond to potential danger, and therefore enhances survival. This early and rapid attention to threat is an adaptive response in healthy individuals (LeDoux, 1986; Oehmann, Flykt, & Esteves, 2001). However, in persons with clinical or sub-clinical levels of anxiety, such process seems to be exaggerated and maladaptive. Specifically, anxious individuals seem to be distinctively more sensitive to threatening information in the environment and display a tendency to allocate attention preferentially to these stimuli (Williams, Mathews, & MacLeod, 1986; Taake, Jaspers-Fayer & Liotti, 2009; Eysenck, Derakshan, Santos, & Calvo, 2007). This hypervigilance to threat takes the form of an attentional bias, and is thought to occur at early, automatic stages of processing, prior to conscious awareness. This increased processing of threatening information has been demonstrated in a variety of tasks in both clinically and non-clinically anxious individuals, and appears to be largely content-specific to the particular symptoms of the various anxiety disorders (Pergamin-Hight, Naim, Bakermans-Kranenburg, van Ijzendoorn & Bar-Haim, 2015), and is

believed to play a role in the etiology and maintenance of clinical anxiety (e.g. Bar-Haim, Lamy, Pergamin, Bakermans-Kranenburg, & van Ijzendoorn, 2007; MacLeod, Matthews, & Tata, 1986; Mogg, Mathews, & Eysenck, 1992).

A critical distinction emerging in the affective processing literature, and specifically in studies investigating anxiety, is the difference in neural responses to supraliminal versus subliminal presented emotional stimuli. Subliminal stimuli refer to what is presented below the threshold of conscious awareness, and compared to supraliminal stimuli, are weak and of low intensity (Meneguzzo, Tsakiris, Schioth, Stein & Brooks, 2014). Threatening stimuli such as fearful and angry faces, predatory and poisonous animals, and weapons have been found to capture spatial attention, both when presented unmasked and masked, in healthy and clinical populations (Carlson et al., 2012; Pourtois, Grandjean, Sander & Vuilleumier, 2004; Carlson & Reinke, 2008; Fox, 2002; Blanchette, 2006; Beaver, Mogg & Bradley, 2005). There is also some evidence that subliminal *positive* prime words can increase liking scores for targets following these prime words if primes score high on arousal dimensions (Gibbons, 2009).

EEG offers a particularly powerful means of studying affective processing in the human brain by recording event-related potentials (ERPs) to affective stimuli from the scalp (Hajcak, MacNamara, & Olvet, 2010; Eimer & Holmes, 2002; Williams, Palmer, Liddell, Song & Gordon, 2006), and such knowledge can provide unique information about the stage of information processing at which the attentional bias to emotional stimuli may occur.

The goals of this project were to examine supraliminal and subliminal affective processing in healthy adults using EEG and well-controlled variations of a supraliminal and a subliminal version emotional Stroop (eStroop) task to document when these effects are taking place. Building upon these results, another primary goal of this research was to provide insight into the electrophysiological correlates of attentional bias to physical threat words in individuals with high and low anxiety sensitivity. Furthermore, we wanted to extend the analysis of hypervigilance to threat signals to a group of Panic Disorder patients, to find out more about the origin of typical physical anxiety symptoms. It is plausible that ERP abnormalities in response to threat stimuli may represent a risk factor for the development of clinical anxiety. Moreover, to further explore automatic

processing of threat, the subliminal eStroop task was conducted to examine the unconscious influence of threat signals.

Identifying underlying neural differences in how threat stimuli are processed will help us to more completely understand this deficit in clinical anxiety and when it breaks down at the neural level. Of specific interest in the Panic Disorder group were neural responses over frontal scalp sites, given a handful of recent studies that converge to suggest possible dysfunction in frontal regions of the brain in this population (Dresler et al., 2012; Pauli, Amrhein, Muehlberger, Dengler & Wiedemann, 2005; Taake et al., 2009).

Chapter 1. Neural correlates of Threat and Emotion Processing in Healthy Adults with High and Low Anxiety Sensitivity

1.1. Introduction

1.1.1. Affective processing in healthy individuals

Research on emotion regulation has become a popular topic in all disciplines of psychology over recent years, and specifically the use of neurophysiological and neurobiological measures in this research field has expanded rapidly. The area of neuroscience attempts to shed light on the *neural structures* underlying emotions, whereas clinical psychologists are often interested in *dysregulation* of emotion, which seems to play a key role in various psychopathologies (Hajcak et al., 2010). From an evolutionary perspective, the attentional selection or orienting to salient stimuli in the environment is crucial for flexible and adaptive behaviours that enhance survival (Pourtois et al., 2013; Carretie, 2014). As emotional events are by definition salient for an individual, a comprehensive understanding of emotional processing and how it relates to attention seems crucial (Carretie, 2014). Pourtois et al. (2013) even propose that the emotional and motivational significance of a stimulus for an individual provides the same influence on attentional processing as the three classic mechanisms identified in the attention literature (exogenous, endogenous, and object-based attention).

Emotional attention seems to be characterized by the following distinct features: Firstly, an enhancement of the neural response to an emotional stimulus in several sensory pathways, which makes it a very efficient mode of processing. Secondly, emotional effects have been observed at very early latencies, which suggests a different spatio-temporal dynamic compared to other attention mechanisms. And lastly, emotional attention may occur in parallel to the other attentional processes, and therefore constitutes at least a partly independent system (Pourtois et al., 2013). Altogether, exogenous, endogenous, and emotional attention may constitute a multiple systems architecture, and the combined influence of these systems determines attention selection.

1

Different stimuli such as emotional pictures, words and faces have been used in various experiments on emotional processing. The majority of studies found some indication of greater attention to emotional than to neutral stimuli (Carretie, 2014). There is some indication that healthy individuals have an enhanced response to positive relative to neutral material (Bradley et al., 1997; Keogh, Dillon, Georgiou, & Hunt, 2001), but typically a negativity bias has been observed (Smith, Cacioppo, Larsen & Chartrand, et al., 2003), with enhanced responses to negative compared to positive and neutral stimuli, manifested in faster reaction times (Hansen & Hansen, 1988; Oehmann, Flykt & Esteves, 2001) or comparatively larger ERPs to negative stimulus materials (Ito, Larsen & Smith, 1998; Smith et al., 2003). These results are consistent with the idea that humans have evolved to give early precedence to signals of potential threat to avoid harm (Williams et al., 2006), and positive stimuli are attended to only once signals of threat have been addressed and safety is assured. These enhanced neural responses to negative stimuli can be observed along several sensory pathways in the brain, including category specific selective regions, and non-specific regions, such as early sensory cortex or fronto-parietal attention networks (Kober et al., 2008). Furthermore, the time-course of emotional effects suggests relatively early responses in limbic regions, such as the amygdala, anterior cingulate cortex, and orbitofrontal cortex (Carretie et al., 2014).

Event-Related Potential manifestations of emotional attentional processing in healthy individuals

Scalp recorded electrical activity of the brain, the electroencephalogram (EEG), is a neurophysiological method often used to study the neural underpinnings of emotion. The EEG can be time-locked to a certain event, for example, the presentation of a stimulus, and the resulting changes in voltage can be measured over time. These changes are then described as event-related potentials (ERPs). They can be differentiated according to their timing, scalp topography, polarity of their deflection, and latency, among other criteria (Hajcak et al., 2010). By recording event-related potentials (ERPs) from the scalp, neural mechanisms underlying affective processing can be studied with a high degree of temporal resolution since ERPs, unlike fMRI, directly reflect neural activity. Such knowledge can provide unique information about the stage of information processing at which an attentional bias to emotional stimuli may affect information processing.

Several ERP components have been identified as being sensitive to emotional processing. The following is a review of those components that are of relevance to this project.

A fronto-central positivity peaking between 80 ms and 150 ms has been reported by several authors to be more pronounced to threat compared to neutral stimuli, in most cases to fearful faces (Eimer & Holmes, 2002; Taylor, Batty & Itier, 2004; Williams et al., 2006; Eimer, Kiss, & Holmes, 2008). This component has been named by some the P150 and has also been shown to distinguish aversive visual scenes from pleasant and neutral scenes, in a time interval of 120 ms to 170 ms (Kawasaki et al., 2001). Since these authors recorded directly from single neurons in a human undergoing neurosurgery, the source of these effects could be traced directly to ventromedial prefrontal cortex. The researchers concluded that prefrontal neurons directly participate in encoding the aversive value of threatening stimuli. Overall, it has been suggested that the P150 indicates a very early appraisal of signals of potential danger prior to detailed perceptual processing, consistent with the view that indicators of threat are given precedence (Williams et al., 2006).

Another frontal positivity following the P150 has been reported by several studies in the time range of 200 ms to 350 ms over bilateral frontal scalp (e.g. Taake et al., 2009; Carolan, Jaspers-Fayer, Asmaro, Douglas & Liotti, 2014; Asmaro et al., 2012). This effect has been referred to as the Early Anterior Positivity (EAP) and has been shown to be sensitive to emotionally salient stimuli. For instance, Taake et al. (2009) explored behavioural and electrophysiological correlates of threat-related emotional processing in non-clinical individuals varying for levels of trait-anxiety for physical symptoms (Anxiety Sensitivity) using a modified eStroop task. We reported an EAP in response to threat words, which peaked as early as 200 ms post-stimulus over anterior frontal scalp in both healthy and highly anxious participants. Two other studies (Li, Zinbarg & Paller, 2007; Pauli et al., 2005) found an EAP to threat words relative to neutral words in similar time ranges (300 ms to 500 ms, and 200 ms to 400 ms). Finally, another study identified a frontal positivity (180-292 ms) to both positive and negative words in relation to neutral ones (Schacht & Sommer, 2009). The EAP might reflect a first response to emotional salience and evaluation of threat in prefrontal cortex (Taake et al., 2009).

One of the most widely studied ERP components is the P300, a broad positivity that is maximal at parietal recording sites and occurs between 300 ms and 500 ms post stimulus onset. Target stimuli in oddball tasks elicit larger P300s than standard stimuli, but amplitude of the ERP depends on the participant's attention to the stimulus. Overall, Hajcak (2010) concluded that "the P300 appears to reflect the allocation of capacity-limited resources toward motivationally salient environment stimuli" (p.131). In oddball tasks, the demands of the task itself predefine which stimuli are relevant. Interestingly, when tasks contain emotional stimuli, several studies indicate that these stimuli capture attention automatically, hence, emotional stimuli in the P300 context seem to be automatically processed as task-relevant. Quite a few researchers have reported increased P300 amplitude to emotional compared to neutral pictures (Hajcak et al., 2010), but also to emotional adverbs (e.g. Thomas, Johnstone, & Gonsalvez, 2007; Naumann, Bartussek, Diedrich, & Laufer, 1992). Whereas earlier studies focused on modulations of the P300, more recent work has focused on what is called the Late Positive Potential (LPP). The LPP arises 300 ms after stimulus onset, is a rather sustained positive reflection that can last well beyond 1000 ms, and typically presents with increased amplitude to negative and positive pictures (e.g. Foti & Hajcak, 2008) and words (e.g. Dillon, Cooper, Grent-t-Jong, Woldorff, & LaBar, 2006), compared to neutral stimuli. Interestingly, amplitude of the LPP has been shown to be larger for more intense, arousing and threatening stimuli, and does not seem to habituate over repeated presentation of the stimulus. Also, most available data suggests that the LPP requires conscious recognition (Hajcak et al., 2010). A frontal LPP with bilateral scalp distribution has also been described to negative stimuli (e.g. Pauli et al., 2005). Elaborative processing of these threatening stimuli is thought to be reflected in this frontal component (Pauli et al., 2005).

In summary, whereas the P300 seems to indicate "a phasic increase towards attention to task-relevant stimuli, the LPP appears to track the sustained increase in attention toward, and processing of, intrinsically motivating stimuli" (Hajcak, 2010, p. 134).

1.1.2. Emotional Processing in Sub-clinical and clinical anxiety

Anxious individuals seem to be distinctively sensitive to threatening information in the environment and display a tendency to allocate attention preferentially to these

stimuli (Williams, Mathews, & MacLeod, 1986; Taake, et al., 2009; Eysenck, Derakshan, Santos, & Calvo, 2007). This hypervigilance to threat takes the form of an attentional bias. In the dot-probe task, which involves the presentation of pairs of stimuli that vary in emotional significance (e.g. one threat, one neutral) followed by a dot appearing in the location of one of the presented stimuli, participants with clinical and non-clinical levels of anxiety have been shown to display faster reaction times to targets appearing at the location of a threat relative to a neutral stimulus, which is indicative of an attentional bias towards threat (e. g. Mueller et al., 2009; Bar-Haim, Lamy, Pergamin, Bakermans-Kranenburg & van Ijzendoorn, 2007). Another commonly used laboratory measure revealing such attentional biases is the emotional Stroop (eStroop) task, where emotional and neutral words are presented in different ink colours, and participants are instructed to respond to the *colour* of the word, while ignoring the word meaning. Anxiety disorder patients and healthy individuals with high trait or state anxiety have been found to have slower reaction times to content-specific threat words relative to positive or neutral words (e.g. Taake et al., 2009; Becker, Rinck, Margraf, & Roth, 2001; Williams, Mathews, & MacLeod, 1996), which has been interpreted as interference caused by the impact of the emotional word.

When looking at electrophysiological evidence of these attentional biases to threat in anxious participants compared to healthy controls, differences in timing, particularly of early stages of information processing of threat stimuli, can be expected in anxious participants, according to the model of hypervigilance to threat, and earlier ERP findings employing emotional faces and words have supported this construct (e.g. Williams et al., 1996; Williams et al., 2007b, Taake et al., 2009). For instance, in a study conducted in our own laboratory (Taake et al., 2009), we found an ERP correlate of a block RT effect (with threat blocks slower than positive blocks) in the form of a *negative* ERP modulation over fronto-central scalp that was present only in the high anxiety group (threat vs. positive blocks), peaking around 380 ms (which we called AN380), which was similar in timing, polarity and scalp topography to the N450 observed in the classical Stroop task (Liotti, Woldorff, Perez & Mayberg, 2000) and interpreted as the result of cognitive conflict. We also reported an earlier anterior ERP effect (200-300 ms) over bilateral frontal scalp that took the form of a *positive* polarity enhancement of threat relative to neutral words, which was present in both AS groups (the EAP). The latter positivity may index an early affective response to the aversive nature and self-relevance

of the presented stimuli, possibly reflecting automatic detection and the first stages of evaluation (the EAP) of a threat stimulus before a choice is made about whether or not to implement a behavioural change (e.g., fight or flight).

Even earlier ERP effects with a similar frontal distribution have been recently reported in response to near-threshold presentations of masked fearful vs. neutral faces (as early as 80 ms), in a group of anxious relative to non-anxious participants (Williams et al., 2007b). A later frontal positivity has also been reported in a blocked version of the eStroop task (300-500 ms), which was also independent of anxiety status (high vs. low behavioral inhibition; Li et al., 2007).

These results seem to support the proposal that *hypervigilance* to threat in heightened anxiety is reflected in early activity in frontolimbic structures (Williams et al., 2006; 2007b; Etkin et al., 2004). In line with these interpretations on the role of anxiety in enhancing vigilance to threat (Williams et al., 2007b), Taake et al. (2009) proposed that trait-anxiety (as exemplified by anxiety sensitivity) modulates early, more automatic stages of processing of threat.

1.1.3. Supraliminal versus subliminal presented stimuli

In the available literature, stimuli have been defined as subliminal if they are "attended to by the brain, but not consciously perceived" (Brooks et al., 2012, p. 2963). When subliminal stimuli are presented, a typically short exposure is followed by a mask, to prevent further processing of the stimulus by visual cortex which is normally required for conscious detection. Nevertheless, subliminal stimuli have been repeatedly found to influence future conscious action without awareness, for example "priming" responses to following neutral or emotional stimuli (Brooks et al., 2012).

There is now evidence in the literature that brain regions involved in conscious and non-conscious processes may overlap, but that these processes originate in different areas of the brain and can occur independently. Meneguzzo, Tsakiris, Schioth, Stein and Brooks (2014) conducted a recent meta-analysis on fMRI studies that used both subliminal and supraliminal presentations of the same stimuli in healthy participants. They found that unconscious processing uniquely activated primary visual areas and insular cortex (responsible for interoceptive awareness). In contrast, Anterior Cingulate Cortex was significantly activated for both unconscious and conscious presentations-possibly integrating the two levels of processing. Some portions of ACC were more

active for conscious processing. Another broad distinction that could be inferred from this meta-analysis is the fact that supraliminal stimuli mostly seem to activate the *left* hemisphere, whereas subliminal presentations often evoked activity in the right hemisphere (Meneguzzo et al., 2014). Another meta-analysis of fMRI studies using only subliminal presentations in healthy populations partly confirmed these findings (Brooks et al., 2013). Across all studies, which also included auditory and lexical stimuli presentations, the brain regions most consistently activated in unconscious processing were the amygdala, hippocampus, bilateral anterior cingulate cortex, insular cortex, and primary visual cortex. The authors concluded that "subliminally presented arousing stimuli activate core brain regions associated with somatosensory, emotional, memory, and visual brain regions" (Brooks et al., 2013, p. 2969). Specifically, the brain area mostly associated with subliminal *face* presentations was the right amygdala, whereas conscious perception of faces has been shown to mostly activate the fusiform gyrus (Meneguzzo et al., 2014). Physiological stimuli, such as subliminal rectal stimulation, and unperceived acid stimulation of the esophagus, activated the bilateral anterior cingulate and bilateral insular cortices in the Brooks et al. (2013) meta-analysis, whereas similar physiological stimuli presented supraliminally evoked activity in anterior cingulate cortex in the Meneguzzo et al. (2014) study. This supports the notion that anterior cingulate cortex is involved in both conscious and non-conscious processing and perhaps integrates and evaluates the two, fulfilling a "gateway" function. The anterior cingulate has also been shown to be involved in several primal emotions in studies on awareness of breathlessness (Liotti et al., 2001), where ACC plays a major role in the awareness of physiological state. It has been proposed that in Panic Disorder, there may be an altered alarm center which could be located in ACC.

What about processing of subliminally presented *emotiona*l stimuli? There is accumulating evidence that non-conscious threat signals activate the amygdala and that the amygdalo-ACC network modulates visual cortical processing at the location of masked threat. It has also been proposed that there is a fast processing route through which subcortical limbic structures, such as the amygdala and thalamus, rapidly send signals of threat directly to regions of the frontal cortex (LeDoux, 1996; Liddell et al., 2005; Williams et al., 2006). This feedforward, subcortical network might be characterized by a great number of excitatory connections between the amygdala and medial prefrontal cortex. For instance, PTSD patients relative to controls have been

found to have positive connectivity between the right amygdala and medial-prefrontal cortex (mPFC), which is consistent with the idea that the amygdala can modulate mPFC activity (Bryant et al., 2008). However, Portois et al. (2013) argue that fast processing of emotional stimuli could also follow a fast feed forward cortical route, through direct occipito-frontal connections (the OccipitoFrontal fasciculus). This is corroborated by recent visual ERP studies showing early visual responses in frontal areas for neutral stimuli rapidly following the visual P1 and N1 (Berchicci, Spinelli, DiRusso, 2016), consistent with the view that posterior and anterior brain areas may be rapidly activated through recursive re-entrant processing during visual processing.

Healthy versus clinical populations

Brain regions involved in conscious and non-conscious processes of emotional stimuli might be different in healthy versus clinical populations. For example, hyperactivation in the amygdalo-ACC network in response to subliminal threat stimuli is associated with individual differences in anxiety (Etkin et al., 2004; Monk et al., 2008). Enlarged amygdala volumes have been found in patients with generalized anxiety disorder (Etkin, Prater, Schatzberg, Menon & Greicius, 2009), and Carlson et al. (2012) recently reported evidence that elevated anterior cingulate cortex gray matter volume is correlated with greater attentional biases to threat. Moreover, it has been suggested that attentional biases to threat in anxiety are mostly prominent when stimuli are being presented at an unconscious level (Williams et al., 2006). These effects seem to occur relatively early within the processing stream, and at frontal brain sites (Windmann, Sakhavat, & Kutas, 2002). For example, Pauli et al. (2005) found that only Panic Disorder patients compared to healthy controls displayed ERP positive potentials triggered by panic-related compared to neutral subliminally presented words in early time windows (200-400 ms after stimulus presentation). Another recent study reported that PTSD patients displayed reduced mPFC activation to fear stimuli only during conscious stimulus processing; when stimuli were presented subliminally, mPFC activity together with amygdala responsivity was, in fact, heightened (Bryant et al., 2008). Also, in an ERP study comparing overt and covert processing of fear stimuli in both healthy and anxious participants, high anxiety individuals were characterized by both a speeding and exacerbation of neural signals to fear in the covert condition, which further underlines the automatic nature of an attentional bias in anxiety (Williams et al., 2007).

1.1.4. Current Project

The current experiments examined behavioural and electrophysiological correlates of supraliminal and subliminal emotional word processing using an eStroop task in non-clinical individuals varying for levels with anxiety sensitivity.

Anxiety Sensitivity (AS) can be considered a subtype of trait-anxiety where the concerns are centered on the experience of the physical manifestations of anxiety. It describes the extent to which people fear a future situation in which they may experience symptoms of physical anxiety (e.g., palpitations, sweating, choking; McNally, 2002). A recent analysis of the psychometric properties of AS suggested that this construct "is broadly applicable to a range of anxiety-related phenomena" (Deacon & Abramowitz, 2007, p. 852). More importantly, however, AS has been recognized as a risk factor for the development of Panic Disorder, in which such symptoms are prevalent (Schmidt & Lerew, 1997), as there is a very strong relationship over time among AS, panic attacks, and Panic Disorder (Maller & Reiss, 1992). AS may also be predictive of the amount of time patients are in an acute episode of Panic Disorder (Perez Benitez al., 2009). Earlier studies on this association have recently been confirmed by neuroimaging results that showed that neural activity in response to emotional stimuli in the cortico-limbic network is correlated to the extent of anxiety sensitivity in Panic Disorder patients (Poletti et al., 2015). Furthermore, AS has been found to be a unique construct of Panic Disorder, as it describes an individual's response to anxiety symptoms - and physical concerns lie at the core of Panic Disorder (Naragon - Gainey, 2010; Olatunji & Wolitzky-Taylor, 2009). In support of this notion, a recent meta-analysis showed that panic among all of the anxiety disorders is most closely related to both the physical and cognitive components of AS. Hence, we chose to use healthy participants with high and low anxiety sensitivity for study 1, since one of the goals was not only to replicate the results of our Taake et al. (2009) study, but also to compare the results of study 1 to those of study 2, in which we wanted to include Panic Disorder patients.

The paradigms employed in this project were unique in its examination of supraliminal and subliminal emotional word processing by using variations of the emotional Stroop task and employing the same stimuli across supraliminal and subliminal tasks. This allowed a within-subjects design, and direct comparison of subliminal and supraliminal processing of emotion. Of specific interest in this study were

frontal ERP components in the 130-320 ms latency, including the EAP, due to the currently equivocal state of the field regarding these components. The LPP, recorded maximally over parietal-temporal sites approximately 350 ms post-stimulus onset and the later frontocentral positivity components were also examined to document the time course of emotional word processing.

Hypotheses

In the behavioural analysis, we hypothesized that specific slowing of reaction time to threat words would only be present in the supraliminal task, and such emotional interference would be detected in the high anxiety sensitive individuals only, as we (Taake et al., 2009) had found in our 2009 study. We did not suspect reaction time differences in the subliminal task, as most studies have not found RT interference in the subliminal eStroop task in healthy controls and high trait anxiety individuals (e.g. Li et al., 2007).

The following hypotheses were formulated with regard to the electrophysiological correlates of the eStroop tasks:

In the supraliminal task, we expected to replicate previous findings from Taake et al. (2009) for early frontal positivities (EAP) to threat stimuli in the high AS participants, and the AN380 indicating greater conflict for threat compared to positive blocks in the high AS group. Later LPP modulation to threat words – reflecting conscious processing of threat information- would be present only in the supraliminal task independent of AS group, in line with the emotional literature, and Taake et al. (2009).

In the subliminal task, we predicted that the detection of threat stimuli below perceptual threshold would similarly elicit frontal positivities, possibly even earlier than the EAP, consistent with hypervigilance to threat in high anxiety individuals. We did not expect the AN380 to be present in the subliminal task due to the lack of conscious conflict generated by threat stimuli. Due to the lack of conscious awareness in the subliminal task, we also did not expect differences in the amplitude of the posterior LPP.

1.2. Methods

The study involved two phases: A screening and experimental session. The Simon Fraser University Ethics Board approved all aspects of these experiments. All participants gave their informed and written consent before participating in this study and received a small monetary remuneration for their involvement.

1.2.1. Screening Session

284 undergraduate students took part in a screening session in which they completed the Anxiety Sensitivity Inventory (ASI) and a demographic questionnaire. The ASI is a 16-item scale used to reveal the fear of anxiety related sensations (Reiss, Peterson, Gursky & McNally, 1986). Scores were distributed as such: Mean=22.31 ± 9.3, range 4-50. Cut-off points were made at 1 standard deviation; those with ASI scores > 31 were classified as high AS, while those with ASI scores < 13 were labeled as low AS. Participants in the two subgroups who reported in the demographic questionnaire being native English speakers were contacted by email and invited to participate in the second study session.

1.2.2. Experimental Session

A total of 44 participants agreed to take part in the experimental session (21 low AS and 23 high AS participants) in exchange for course credits or a small monetary compensation. On the day of the EEG session, they first completed four additional questionnaires, a medical screening form, the Spielberger State Inventory (STAI-S; Spielberger, Gorsuch & Lushene, 1983), the Anxiety Sensitivity Index as a re-test, and two visual analogue scales assessing subjective ratings of current anxiety and sadness, respectively (on a scale 0-10). All participants reported normal colour vision, normal or corrected-to-normal visual acuity, and admitted being free of current and previous neurological and psychiatric disorders, and current use of psychotropic medications (as measured by the self-administered medical questionnaire). Participants were then tested in a sound-attenuated room facing a computer monitor. Firstly, their individual perception threshold to word stimuli was determined. Then, their electrical brain activity was recorded while they performed variations of the eStroop task to explore the presence of subliminal and supraliminal attentional bias to threat words. Upon completion, they filled

out two more questionnaires, the Beck Depression Inventory, version 2 (BDI-II; Beck, Steer & Carbon, 1988), and the Spielberger Trait Inventory (STAI-T; Spielberger et al., 1983). Lastly, recognition of the subliminally presented threat words was checked behaviourally to ensure that the stimuli had been indeed presented sub-threshold and had not been recognized during the experimental session.

Threshold determination phase

The threshold determination phase was modeled after Pauli et al, (2005). A set of 8 neutral and 8 threat words different from the stimuli used in the experimental session were used to determine each participant's individual perception threshold in the following procedure: Each of the words was presented repeatedly with increasing exposure times until it was identified correctly. The base presentation time was set according to the screen refreshment rate, which was 13 ms, and multiples of 13 were used thereafter. For example, if the first stimulus was not correctly identified, it was presented again for 26 ms and so on, until the participant correctly named the word (Pauli et al., 2005). If the word was not recognized at an exposure time of 130 ms, the next trial commenced, but this rarely happened. The set of stimuli was being presented to each participant repeatedly until there was one presentation time at which 6 words were correctly identified. The perception threshold used in the subliminal task for this participant was then calculated as 13 ms less than that presentation time.

1.2.3. Behavioural task

The experimental tasks were programmed in E-Prime 2.0 Professional (Psychology Software Tools, Inc., Pittsburgh, PA). Modified versions of an eStroop task were employed that included physical threat, positive, and neutral words presented in one of four colors (red, blue, green, or yellow). In the subliminal task, stimuli were shown one at a time at the participant's individual perception threshold and then immediately followed by a mask consisting of a string of letters and symbols that was presented for 300 ms minus the participant's individual perception threshold (see Figure 1-1). The interstimulus interval was randomly jittered between 1.7 and 2.3 sec. There were three different kinds of experimental blocks. In the *Threat Blocks*, physical threat and neutral words were randomly intermixed, with equal probability. In the *Positive Blocks*, positive valence and neutral words were shown. Within each block, emotional words were

12

individually matched to neutral words for frequency of use and length (Kucera and Francis, 1067; see word stimuli in Appendix A). In the Mask Blocks, the mask alone was presented for 300 ms. There was a total of six experimental runs (2 Threat, 2 Positive, 2 Mask Blocks), each comprised of 120 stimuli. Order of presentation was counterbalanced across participants. Participants were instructed to maintain central fixation and to discriminate the ink color of the mask, and to respond as quickly and accurately as possible by pressing one of four response keys on a game pad controller. Average reaction time was calculated for each trial type and emotional block. Only reaction times from correct hits were used in the following analysis.

The supraliminal task was identical, except there was no mask being shown. Words were presented for 300 ms, and participants were instructed to respond to the color of the word (see Figure 1-2). There was a total of four blocks (two threat blocks, two positive blocks), and there were no mask blocks. A different set of words was shown in the supraliminal task.

After the experimental session, the participants received a list of words that included the 12 threat words and 12 neutral words from the subliminal task, as well as 12 different neutral and 12 threat words with the instruction to indicate which words, if any, they had seen during the study.

1.2.4. ERP measures

The electroencephalogram (EEG) was recorded from 64 standard 10-20 scalp sites mounted in an elastic cap with active Ag/AgCl electrodes (Biosemi Active Two system, Amsterdam, NL). Six additional channels included four eye movement channels (two at the external canthi and two below each orbit) and two reference electrodes placed on each mastoid bone. EEG and EOG channels were digitized at a sampling rate of 512 Hz. Off-line processing was performed in BESA 5.3 (Brain Electric Source Analysis, Gräfelfing, Germany). Filter settings included a highpass zero-phase filter set at 0.1Hz (12dB/oct), and a low-pass filter of 30 Hz. ERPs were timelocked to stimulus onset and baseline corrected to the mean voltage of the 200 ms pre-stimulus interval. Trials containing blinks and eye movements were removed prior to ERP averaging by applying a semi-automated artifact detection routine based on amplitude at eye movement channels implemented in BESA (thresholds were individually adjusted). A

total of four participants (two for each group) were excluded for either excessive blinking or technical difficulties while recording behavioural performance, leaving a total of 21 high AS and 19 low AS participants for the supraliminal analysis, and a total of 17 high AS and 19 low AS for the subliminal analysis.

1.2.5. Behavioural Data Analysis

Mixed design ANOVAs were employed on the mean correct RT (RTs < 150 ms or > 1500 ms were excluded). The between group factor was AS status (high vs. low). Within group factors were emotional block (threat vs. positive), and type of trial (emotional vs. neutral). Statistical threshold was set at $p < 0.05$. This design allowed testing for the significance of both between-block differences ("slow" effects) and within-block differences ("fast" effects). Furthermore, mixed design ANOVAs were run on the mean number of correctly and falsely recognized stimuli from the post-experimental recognition test. The between group factor was AS status (high vs. low). Within group factors were presence of stimulus in the study (hit vs. false alarm), and word type (threat vs. neutral).

1.2.6. ERP data analysis

Firstly, distinct ERP averages were obtained for all four trial types (physical threat and neutral words in the threat blocks, positive and neutral words in the positive blocks), time-locked to word onset. Secondly, two collapsed ERP averages were calculated for all words within the threat blocks and all words within the positive blocks. Only ERPs to correct RT responses (hits, 150-1500 ms) were included. All channels were referenced to the average of the two mastoid electrodes. ERP amplitudes were aligned to a 200 ms pre-stimulus baseline period. Grand averages were calculated across subjects for each trial type (emotional and neutral words), emotional block (threat and positive) and group (high vs. low AS). To help isolating effects of interest, within group difference waves were calculated. To facilitate visualization, scalp voltage topographic distributions were obtained using spherical spline interpolation. After inspection of grand-average waveforms and scalp topography distributions for each emotional block and each word type and the various difference waves, several effects of interest were identified.

1.2.7. Statistical analyses

For the statistical analyses, mixed-design ANOVAs were employed with between factor being group (high anxiety vs. low anxiety), and within factor being emotional block (threat vs. positive), and trial type (emotional vs. neutral). Global analyses were followed by more restricted ANOVAs and t-tests upon significance of interactions, or when a priori hypotheses for group differences were present. For all analyses, the critical p-value was set at 0.05, and degrees of freedom were corrected with the Greenhouse–Geisser epsilon method to correct for sphericity (Greenhouse and Geisser, 1954).

1.3. Results

1.3.1. Clinical and demographic variables

Demographic characteristics of the participant groups are summarized in Table 1-1. The high and low AS participants did not differ on age [$t(1\ 41)= -1.26$, $p=.216$)], education [$t(1\ 41)= -.83$, $p=.411$)], or handedness [$t(1\ 42)= .093$, $p=.926$)]. However, there was a greater number of women overall (29 female vs 15 male participants, $\chi^2\ (1) = 4.46$, $p = 0.035$), as well as within the high AS group (18 female vs 5 male participants, $\chi^2\ (1) = 7.35$, $p = 0.007$), but not within the low AS group $\chi^2\ (1) = 0.048$, $p = 0.827$. High AS participants reported significantly higher scores on the ASI at screening [$t(1\ 42)= 20.89$, $p=.000$)], on the ASI re-test [$t(1\ 42)= 10.32$, $p=.000$)], on the BDI [$t(1\ 41)= 4.51$, $p=.000$)], on the STAI-S [$t(1\ 42)= 4.65$, $p=.000$)], on the STAI-T [$t(1\ 42)= 5.66$, $p=.000$)], on the Anxious VAS [$t(1\ 42)= 4.46$, $p=.000$)], and on the Sad VAS [$t(1\ 42)= 4.26$, $p=.000$)], compared to the low AS participants.

1.3.2. Behavioural results

Threshold Determination Phase

Table 1-2 depicts separately for high AS and low AS participants the mean perception thresholds and frequency of which each perception threshold occurred within each group. The two groups did not differ on the mean perception threshold [$t\ (1,34)=-2.02$, $p=.052$]. The most common threshold was 13 ms.

Supraliminal task reaction times

Average reaction time and standard error for each combination of emotional block and word type in the high and low AS groups in the supraliminal Stroop task are depicted in Table 1-2 and Figure 1-3. In the mixed design ANOVA, there was a significant main effect of emotional block [$F_{(1,41)}$ =4.10 p=.050]. All participants responded on average 13.42 ms slower to threat blocks than to positive blocks. Such slowing in the threat blocks relative to the positive blocks was independent from type of stimulus, being present for both threat and neutral words. This slow RT effect is similar to what was reported in Taake et al (2009). In spite of the RT interference being on average larger for the high AS than the low AS group (20 msec vs. 8 msec), the group x emotional block interaction did not approach significance [$F_{(1,41)}$=.779 p=.383], and t tests run within each group failed to reach significance (high group: t[(1,21)=1.86, p=.075], low group: t[(1,20)=.917, p=.370].

Subliminal task reaction times

Average reaction times and standard error for each combination of emotional block and word type in the high and low AS groups in the subliminal Stroop task are depicted in Table 1-3 and Figure 1-4. In the mixed design ANOVA, there was no significant main effect of emotional block, type of trial, or any significant interactions (for all p >.10).

Recognition task

Percentages of correctly recognized words and false alarms as a function of Group and Word Type in the subliminal Stroop task are depicted in Table 1-4. As can be seen, the percentage of words correctly recognized from the subliminal task was very small, and therefore it is very unlikely that the Hits exerted a great influence onto the behavioural effects or ERP effects.

1.3.3. ERP results

Supraliminal task

Block differences

AN450 (350-500 ms)

The evoked response to words in the threat blocks relative to words in the positive blocks revealed greater amplitude of a negative wave with frontal distribution which was greater on the left, peaking between 350-550 ms (here called AN450, anterior negativity peaking at 450 ms). Such effect was only observed in the high AS group (see Figure 1-5). For the AN450 (350-550 ms), an anterior lateral region of interest (ROIs) was selected for the left hemisphere, by averaging together sites (AF7, AF3, F7, AFz).

In the global analysis, the interaction of block x group approached significance [F(1,39)=3.94 p=.054]. Based on our previous findings, we proceeded with more restricted Anova's within groups. In the high group, threat blocks elicited more negativity than positive blocks [F(1,39)=4.91 p=.038]. In the low group, there was no difference between blocks [F(1,39)=.22 p=.65]. This effect is illustrated in Figure 1-5.

Within-Block ERP Analysis

Within the threat blocks, ERPs to physical threat words were characterized by greater amplitude of an early slow wave with positive polarity (here called the Early Anterior Positivity or EAP) relative to neutral trials. This wave extended for several hundred ms, its voltage difference peaking between 200-320 ms from onset of the threat word. Such effect also had a focal scalp distribution over frontal scalp, more prominent on the left, and was only present in the high AS group. In contrast, an EAP was not evoked by the emotional words in the positive block for either group. Secondly, it was noticed that the ERPs to the physical threat words also appeared to show an even earlier difference for the high AS group, with an earlier frontal modulation (as early as 150 ms, called here P150). This unexpected effect was analysed on an exploratory basis. For the EAP (200-320 ms), an anterior lateral region of interest (ROIs) was selected for each hemisphere, by averaging together four neighbour electrode sites where the voltage difference was maximal (sites AF7/AF8, AF3/AF4, F3/F4 and F5/F6). For the P150 (130-200 ms), an anterior lateral region of interest (ROIs) was selected for

17

each hemisphere, by averaging together three neighbour electrode sites where the voltage difference was maximal (sites AF7/AF8, F5/F6, and F7/F8).

Thirdly, a later latency, posteriorly distributed positive slow wave (LPP, peaking around 450 ms over parietal scalp) displayed greater amplitude for threat than neutral words. However, such LPP modulation appeared to be present in both groups independent of AS status. For the later LPP window (350-550 ms), a posterior parietal ROI was chosen, collapsing sites P1/P2 and P3/P4. The within-block effects are depicted in Figure 1-6.

P150 (130-200 ms)

There was no hint of a significant main effect of group, emotional block, or trial type, or any significant interactions (for all p>.10).

Early Anterior Positivity (EAP 200-320 ms)

Threat words elicited greater amplitude EAP than neutral words over left frontal scalp, resulting in a main effect of emotional trial (emotional words: 4.01 ± .604 µV; neutral words: 3.05 ± .61 µV) [F(1,39)=8.19 p=.007]. There was also a significant interaction of emotional trial x AS group [F(1,39)=6.20, p=.017]. In the high AS group, mean amplitude of the EAP over left frontal scalp was greater in response to threat words relative to neutral words in the threat blocks (threat words: 4.61± .89 µV; neutral words: 2.80 ± .78 µV) t[(1,20)=3.47, p=.002]. In contrast, in the low AS group, EAP amplitude did not differ as a function of emotional trial (threat words: 3.41 ± .80 µV; neutral words: 3.23 ±.93 µV) t[(1 18)=.301, p=.78]. These effects are illustrated in Figure 1-6. In the positive blocks, no EAP was present for emotional words relative to neutral words in either group (p>.20).

Parietal LPP (350-550 ms)

Mean amplitude of the LPP over posterior scalp was greater in response to threat than neutral words in both groups, yielding a significant main effect of type of trial [F(1,39)=8.46, p=.006], and a significant interaction of block x type of trial [F(1,39)=11.24, p=.002]. There was no significant main effect, or interactions involving group (for all, p > .10). Across groups, in the threat blocks, threat words were more positive than neutral words t [(1 39) =4.71, p=.000]. In contrast, in the positive blocks, there was no difference

between positive and neutral words t [(1 39) =-.38, p=.71]. These effects are illustrated in Figure 1-6.

Subliminal task

Block differences

Firstly, the evoked response to words in the threat blocks relative to words in the positive blocks revealed greater amplitude of a bilateral positive wave with frontocentral distribution, peaking between 130 and 200 ms (here called the P150), 200 and 320 ms (here called the Early Anterior Positivity, the EAP), and 350 and 550 ms (here called frontal LPP). Such effects were only observed in the high AS group. Anterior lateral regions of interest (ROIs) were selected for each hemisphere, by averaging together electrode sites for each effect where the voltage difference was maximal. For the P150, the following electrodes were averaged: AF3/AF4, AFz, Fz, F1/F2. For the EAP, sites AF7/AF8, AF3/AF4, F3/F4 and F5/F6 were used, and for the frontal LPP, the ROI consisted of electrodes AF3, F1, F3, F5, AF4, F2, F4, F6.

Secondly, the evoked response to words in the threat blocks relative to words in the positive blocks revealed greater amplitude of a bilateral positive wave with parietal distribution, peaking between 350-550 ms (here called the LPP). Such effect was also only observed in the high AS group. For this analysis, parietal lateral regions of interest were selected for each hemisphere, by averaging together electrode sites P3/P4, P1/P2.

P150 (130-200 ms)

In the global analysis, the main effect of emotional block was significant [F (1,34)=5.20 p=.029]. There was also a significant interaction of emotional block x AS group [F (1,34)=4.14, p=.050]. In the high AS group, mean amplitude of the P150 over frontal scalp was greater in response to threat blocks relative to positive blocks (threat blocks: 5.01± .66 µV; positive blocks: 3.87 ± .57 µV) [F (1,16)=6.79, p=.019]. In contrast, in the low AS group, P150 amplitude did not differ as a function of emotional block (threat blocks: 3.94 ± .70 µV; positive blocks: 3.88 ±.70 µV) [F (1,18)=.044, p=.837]. These effects are illustrated in Figure 1-7 and Figure 1-8. No other main effects or interactions approached significance (p>.10).

Early Anterior Positivity (EAP 200-320 ms)

In the mixed design ANOVA, there was a significant interaction of emotional block x AS group [$F_{(1,34)}$=6.37, p=.016]. In the high AS group, mean amplitude of the EAP over frontal scalp was greater in response to threat blocks relative to positive blocks (threat blocks: 3.43 ± .87 µV; positive blocks: 2.08 ± .64 µV) [$F_{(1,16)}$=6.79, p=.019]. In contrast, in the low AS group, EAP amplitude did not differ as a function of emotional block (threat blocks: 2.91 ± .86 µV; positive blocks: 3.18 ± .87 µV) [$F_{(1,18)}$=.472, p=.501]. These effects are illustrated in Figure 1-7 and Figure 1-8. No other main effects or interactions approached significance (p>.10).

Frontal LPP (350 – 550 ms)

In the global analysis, the main effect of emotional block was significant [$F_{(1,34)}$=5.31 p=.027]. The interaction of emotional group x block failed to reach significance [$F_{(1,34)}$=1.96 p=.171, but given our apriori hypothesis of a selective frontal enhancement to threat blocks in the high AS group, and based on visual inspection of the waveforms, we proceeded with within-group analyses. In the high AS group, there was a main effect of emotional block [$F_{(1,16)}$=6.55 p=.021], which was explained by greater amplitudes for threat blocks compared to positive blocks (threat blocks: 3.19 ± .94 µV; positive blocks: 1.69 ± .93 µV). In contrast, in the low AS group, frontal LPP amplitude did not differ as a function of emotional block (threat blocks: 3.12 ± .901 µV; positive blocks: 2.75 ± 1.05 µV) [$F_{(1,18)}$=.432 p=.519]. These effects are illustrated in Figure 1-7 and Figure 1-8. No other main effects or interactions approached significance (p>.10).

Parietal LPP (350-550 ms)

There was no hint of a significant main effect of group, emotional block, or trial type, or any significant interactions (for all p>.10).

1.4. Discussion

Behavioral and electrophysiological correlates of threat-related emotional processing were explored in non-clinical individuals varying for levels of trait-anxiety for physical symptoms (Anxiety Sensitivity) using a modified supraliminal and subliminal eStroop task.

In the behavioral analysis for the Supraliminal task, a specific slowing of reaction time to threat words was only present as a block effect (threat versus positive blocks), and such emotional interference was detected in both the high and low AS group. There was no interference effect present in any group in the subliminal Stroop task. Furthermore, a number of threat-specific ERP effects were found.

Supraliminal task: As predicted by our hypothesis, an ERP correlate of the block RT effect was found in the form of a *negative* ERP modulation over fronto-central scalp present only in the high AS group (threat vs. positive blocks), peaking around 450 ms (called AN450). Our hypothesis about frontal positivities was also confirmed: Threat-related ERP modulations within the threat blocks were identified over frontal scalp. However, such anterior ERP effect took the form of a *positive* polarity enhancement peaking earlier than the AN450 (EAP 200-320 ms), and again was present only in the high AS group. Given the absence of RT effects associated with the threat vs neutral contrast and their earlier latency, the EAP is being interpreted as reflecting a response to emotional salience rather than emotional conflict. Other threat-specific within-block effects were modulations of the late positive potential (LPP) over posterior scalp, being present in both groups. Such effect replicated previous findings in the literature.

Subliminal task: As hypothesized, threat-related ERP modulations for the threat blocks were identified over anterior bilateral frontal scalp. Such anterior ERP effects took the form of a *positive* polarity enhancement in response to threat blocks relative to positive blocks peaking at a very early time window of 130-200 ms (P150), and also between 200 and 320 ms (EAP), and again between 350 and 550 ms (frontal LPP). These effects were only present in the high AS group and suggest that early enhancement to threat material in high AS individuals can happen at a pre-conscious stage.

Behavioural findings

High and low AS participants were found to have similar perception thresholds as determined with neutral words before the two experiments: The most common threshold used in both groups was 13 msec. Furthermore, both groups were comparable in terms of the percentages of correctly recognized words and false alarms in the subliminal Stroop task, and both Hits and False Alarms were at chance rate in both groups.

Reaction times: All participants responded on average 13.42 ms slower to threat blocks than to positive blocks in the supraliminal Stroop task. Such slowing in the threat blocks relative to the positive blocks was independent of type of stimulus, being present for both threat and neutral words. This slow RT effect is similar to what we reported in the Taake et al. (2009) study, which found an interference effect to threat blocks versus positive blocks in the high AS participants only. In the present study, both groups displayed a similar tendency to respond slower to the threat blocks; however, low AS participants were on average only 7.55 ms slower, whereas the high AS participants showed an interference effect of 19.27 ms. This result is consistent with several studies conducted with anxiety disorder patients who displayed similar RT slowing to *blocks* of stimuli specific to the participants' disorder rather than mixed presentations of threat and non-threat words (Becker et al., 2001; Hope et al., 1990; van den Heuvel et al., 2005; see meta-analysis in Phaf & Kan, 2007). Our findings corroborate previous evidence that high AS individuals display a selective concern-specific cognitive bias towards physical threat words.

There was no specific slowing of reaction time detectable in either AS group in the subliminal Stroop task. This finding is in line with several studies in the current literature that conducted subliminal tasks involving threat stimuli with participants varying in trait anxiety and found no behavioural interference effects (e.g. Berggren & Derakshan, 2013; Gibbons, 2009; Li et al., 2007). For instance, Berggren and Derakshan (2013) concluded that behavioural differences in selective attention and cognitive control solely seem to modulate conscious but not unconscious processing. Note, however, that there are a few subliminal Stroop studies and one dot-probe study that did find an interference effect in the unconscious condition in participants with trait anxiety (e.g. Hunt, Keogh & French, 2006; Mogg, Kentish, & Bradley, 1993; Van den Hout, Tenney, Huygens, Merckelbach, & Kindt, 1995; Macleod & Hagan, 1992). This difference may be in part attributable to the fact that those studies used the same presentation time for each participant and did not determine individual perception thresholds. Furthermore, some of the studies used considerable longer presentation times than most of our participants' individual perception thresholds: For instance, Macleaod and Hagan (1992) presented all stimuli for 20 ms, and van den Hout et al. (1995), presented all stimuli at 30 ms. Perhaps the participants in those studies were

able to identify at least some of the stimuli, although this conclusion is, of course, speculative.

ERP findings

Anterior Negativity (AN450: 350-500 ms). In the high AS participants only, threat block words relative to positive block words elicited greater negative amplitude in the 350-500 ms time window over frontocentral scalp in the *supraliminal Stroop* task. This is a direct replication of our Taake et al. (2009) findings. Even though an interference RT effect was found in both groups in this study, we propose, in line with Taake et al (2009), that the AN450 represents an ERP correlate of the eStroop RT interference effect. Evidence for this comes from previous studies on the cognitive Stroop task that have found a similar negative wave around 400 ms with fronto-central distribution in response to incongruent relative to congruent color words (Liotti, Woldorff, Perez & Mayberg, 2000; West & Alain, 1999), which has been interpreted as indicating conflict detection (West, 2003). Furthermore, an emotional Stroop task by van Hoff, Dietz, Sharma and Bowman (2008) reported a similar frontal wave in response to negative relative to neutral stimuli, which was present only for those negative words which produced RT interference, and only if the intertrial interval was long (500 ms). In contrast, the frontal wave was not present if the intertrial interval was short (40 ms), albeit this was the condition that produced the greatest RT interference effect, suggesting that time pressure may be a factor in the latter. Lastly, a recent meta-analysis of fMRI studies of emotion-word and emotional counting Stroop paradigms identified a cognitive control neural circuit consisting of several regions in prefrontal cortex and parietal cortex to be involved in the Stroop effect, which is in line with the scalp distribution of the AN450 (Feng et al., 2018).

Late Positive Potential effects (LPP, 350-550 ms). Mean amplitude of the LPP over posterior scalp was greater in response to threat than neutral words in both groups in the *supraliminal Stroop* task. These results are in line with a large body of literature, showing that emotionality and valence of visual stimuli (words and faces) are reflected in modulations of this component: The LPP arises 300 ms after stimulus onset, is a rather sustained positive reflection that can last well beyond 1000 ms, and typically presents with increased amplitude to negative and positive pictures (e.g. Foti & Hajcak, 2008) and words (e.g. Dillon, Cooper, Grent-t-Jong, Woldorff, & LaBar, 2006; Fischer & Bradley,

2006, for a review of word studies), compared to neutral stimuli. Thomas et al. (2013) conducted the emotional Stroop task in Panic Disorder patients, obsessive-compulsive disorder patients, and healthy controls, and found increased P300 amplitudes to threat versus neutral stimuli in those participants across groups who showed Stroop interference. Also, another eStroop study comparing healthy participants with individuals high in anxious-apprehension and anxious arousal found no P300 differences between groups to threatening or emotionally arousing words (Sass et al., 2010). Lastly, Pauli et al. (2005) presented threat or neutral words to Panic Disorder and healthy participants in a study where emotion was task-irrelevant. Threat words elicited greater amplitude LPPs (400-600 ms) than neutral words, and the effect was present independent of clinical diagnosis. It therefore appears that late conscious evaluation of a threat stimulus (as indexed by the LPP) is not affected by anxiety status. In sharp contrast, there was no posterior LPP modulation in either group in the *subliminal Stroop* task. This is not surprising since most available data suggests that the LPP requires conscious recognition (Hajcak et al., 2010).

P150 (130-200 ms). In the subliminal task and in the high AS group, P150 amplitude over frontal scalp was greater in response to threat blocks relative to positive blocks. Such effect was not present in the low AS group. This suggests that early enhancement to threat material in high AS individuals can happen at a preconscious stage, and that this process operates automatically. Since our recognition test showed that discrimination of individual threat words was at chance level, hypervigilance to threat in the high AS individuals seems to manifest itself as amplified processing of all stimuli in the threat blocks (emotional and neutral), perhaps in order to foresee potential threat. This mechanism is reminiscent of sensitization in conditioning studies involving aversive stimuli. A study by Carretie et al. (2005) found similar results. The researchers presented visual stimuli containing spiders to spider-phobic participants below conscious detection threshold and found increased activity in ventromedial prefrontal cortex (vmPFC) at 150 ms after stimulus onset to pictures of spiders. A few other studies using conscious stimuli have also found fast reactions (100 ms to 150 ms) in vmPFC to stimuli that were perceived as dangerous (e.g. Kawasaki et al., 2001; Northoff et al., 2000; Simpson et al., 2000; Williams et al., 2006). Interestingly, vmPFC seems to be involved only in the response to danger, and not to other emotionally negative stimuli. Its function may be to regulate in a top-down fashion attention through its reciprocal connections

with parietal and visual cortex. vmPFC also receives input from the amygdala, which is specialized in danger detection (Carretie et al., 2005).

EAP (200-320 ms). In the threat blocks in the supraliminal task, ERPs to emotional words elicited an early, greater positive deflection than neutral words. Such effect was present in the high AS group only, whereas in the positive blocks, no effect was found in either group. This effect is a direct replication of our Taake et al. (2009) study, where we reported an EAP in response to threat words, which peaked as early as 200 ms post-stimulus over anterior frontal scalp in both healthy and highly anxious participants. The EAP has also been found by two other studies in the time range of 200 ms to 350 ms over bilateral frontal scalp that were conducted in our laboratory (Carolan et al., 2014; Asmaro et al., 2012). Moreover, another experiment identified a frontal positivity (180-292 ms) to both positive and negative words in relation to neutral ones (Schacht & Sommer, 2009). In a recent blocked version of the eStroop task threat words elicited a positive frontal modulation relative to neutral words in a 300-500 ms time window, which was independent of anxiety status (high vs. low behavioural inhibition). No significant emotional RT interference was reported in this study in either group (Li et al., 2007). All of these studies appear to share the fact that emotion was *covert*, or task-irrelevant. Please note that in the subliminal task, the EAP was present as a block effect (composed of threat as well as neutral words), once again in the high AS group only: ERP amplitude to threat blocks was greater relative to positive blocks. A similar result, however, was obtained in a study where physical threat and neutral words were randomly presented below individual perception threshold to Panic Disorder patients and healthy controls in a mixed-trial paradigm (Pauli et al., 2005). Subjects read each word after a 3-sec delay. A frontal positivity (200-400 ms) distinguished threat from neutral words in both groups, being more pronounced in Panic Disorder patients. The authors concluded that panic patients are characterized by an early automatic and elaborate processing of disorder relevant stimuli. In line with this interpretation, we propose here that the EAP may index rapid affective responses to the aversive nature and self-relevance of the presented stimuli, in the form of increased processing of all stimuli in the threat blocks in the case of our subconscious stimuli presentations. Our results suggest that there is affective processing of unconscious word stimuli, and that individual differences in anxiety modulate the susceptibility to subliminal evaluation of these stimuli.

<u>Frontal LPP (350 – 550 ms)</u>. In the high AS group, mean amplitude of the frontal LPP was greater in response to threat blocks relative to positive blocks. Such effect was not present in the low AS group. This positive late slow wave has been associated with elaborate processing (Pauli et al., 2005), and has been reported before in Panic Patients (at 400 ms – 600 ms) who were presented with physical threat words at perception threshold (Pauli et al., 2005).

<u>Proposed Functional dissociation of frontal ERP effects.</u> The high AS group showed a very early frontal positivity (P150) to threat-containing relative to positive-containing stimuli blocks, which persisted throughout the processing sequence (P150 through frontal LPP). However, no difference was present in the posterior LPP waves between the same blocks. In the absence of conscious discrimination of individual threat words, hypervigilance to threat in high AS individuals appears to take the form of an increased processing of potential threat for all stimuli in the threat blocks. Persistent frontal activity through time is consistent with evidence in PTSD of the involvement and feedback of interacting fear networks, one acting rapidly and one slowly, which may be re-activated throughout the sequence of threat processing (Williams et al., 2006). The present study suggests that distinct early frontal modulations may index separate components of emotional processing of threat words. The frontal positivities that were found in the absence of corresponding RT differences may index rapid affective responses to the aversive nature and self-relevance of the presented stimuli, possibly reflecting early automatic detection (the P150), and the first stages of evaluation (the EAP) of a threat stimulus before a choice is made about whether or not to implement a behavioural change (e.g., fight or flight). This notion is supported by some studies in the literature that proposed that *hypervigilance* to threat in heightened anxiety is reflected in early activity in frontolimbic structures (Williams et al., 2006; 2007b; Etkin et al., 2004). Electrical recordings from amygdala neurons in animals show activation to threat stimuli as early as 50 ms post-stimulus (Ledoux, 1996). Neuroanatomy provides evidence of a <u>direct</u> forward projection from the amygdala to medial prefrontal cortex, which may support automatic/unconscious detection of threat stimuli such as fearful faces (Williams et al., 2006; 2007b). In line with such findings and interpretations on the role of anxiety in enhancing vigilance to threat (Williams et al., 2007b), we propose that enhanced frontal P150 and EAP to subliminal threat words may represent a marker of hypervigilance to

26

threat with important implications for diagnosis and treatment in panic and other anxiety disorders.

1.4.2. Limitations

Sample size in the groups was rather small, albeit comparable to most published ERP studies. Nonetheless, further replication and extension of the study, particularly in relation to the P150, may be indicated.

Secondly, source analysis was not carried out in the present study, and therefore definite conclusions about differential brain localization of ERP effects cannot be drawn.

Results from this study may have limited generalizability. Individuals included in this study were screened based on specific inclusion and exclusion criteria (specific cut-offs on the ASI questionnaire) to form a low and high AS group. Individuals scoring one standard deviation below the mean on the ASI questionnaire to be classified as low AS may not be representing average or normal levels of anxiety, and therefore may not represent the most ideal control group. Furthermore, there were significantly more females in the high AS group (and males in the low AS group). Therefore, results may be skewed and not really represent males with high anxiety sensitivity. In addition, other more general gender differences in threat processing cannot be teased apart from the influence of AS status.

Regarding the statistical design, the two tasks could have been analyzed in one overall ANOVA, by including task (subliminal vs. supraliminal) as a within-subject factor in such analysis. We decided against this design to avoid the difficulty of teasing apart interactions with too many factors. Furthermore, we were using multiple statistical tests without correction for multiple comparisons, which increases the chance of erroneous inferences are to occur.

1.5. Study 1 Tables

Table 1-1:   Sociodemographic Characteristics of the Participants (high and low AS participants)

Characteristic	High AS (n=23)		Low AS (n=21)	
	n	%	n	%
Male	5	21.7	10	47.6
Female	18[§]	78.3	11	52.4
Handedness (right)	21	91.3	19	90.4
	Mean	**SD**	**Mean**	**SD**
Age (years) [a]	20	3.7	21.9	6
Education (years) [a]	13.7	1.2	14	1.3
ASI at screening[**]	36.5	5.3	10.3	2.4
ASI at test day[**]	35.5	10.5	10.1	4.3
BDI[**]	12.6	6.8	4.8	3.9
STAI-S[**]	43.3	9.5	31.8	6.6
STAI-T[**]	50.4	8.3	35.7	9
Anxious vas[**]	4.7	2.6	1.5	2.2
Sad vas[**]	2.7	2.4	4	8

[a] 1 participant was excluded due to incomplete questionnaires
[§] Within group significance p<.01 (Chi-square test)
[*]   p<.05
[**] p<.001

Table 1-2: Perception thresholds of the participants in the Subliminal Stroop task.

Group	High AS (n=17)		Low AS (n=19)	
Threshold used (msec)	n		n	
13	11		17	
26	4		2	
39	2		0	
	Mean	**SD**	**Mean**	**SD**
Mean perception threshold (msec)	19.1	9.3	14.4	4.1

Table 1-3: Average reaction time (RT) and Standard Error for each combination of Emotional Block and Word Type in the high and low AS group in the supraliminal Stroop task.

		Means in msec (SE)	
Emotional Block	**Word type**	**High AS**	**Low AS**
Threat	Threat	711.5 (23.3)	732.9 (24.0)
	Neutral	720.8 (23.5)	727.4 (24.0)
	Threat minus Neutral trial	-9.3 msec	5.5 msec
Positive	Positive	697.7 (23.0)	724.7 (23.5)
	Neutral	696.0 (21.6)	720.4 (22.1)
	Positive minus Neutral trial	1.7 msec	4.3 msec
Threat minus Positive Block	(Threat plus Neutral) minus (Positive plus Neutral)	19.3 msec	7.6 msec

Table 1-4: Average reaction time (RT) and Standard Error for each combination of Emotional Block and Word Type in the high and low AS group in the subliminal Stroop task.

		Means in msec (SE)	
Emotional Block	**Word type**	**High AS**	**Low AS**
	Threat	691.2 (22.6)	701.3 (26.3)
Threat	Neutral	697.0 (23.7)	694.5 (27.6)
	Threat minus Neutral trial	-5.8 msec	6.8 msec
	Positive	687.3 (20.7)	708.4 (24.1)
Positive	Neutral	679.6 (20.1)	705.9 (23.4)
	Positive minus Neutral trial	7.7 msec	2.5 msec
Threat minus Positive Block	(Threat plus Neutral) minus (Positive plus Neutral)	10.7 msec	-9.3 msec

Table 1-5: Percentages of correctly recognized words and false alarms as a function of Group and Word Type in the subliminal Stroop task.

Presence of stimulus	**Word type**	**High AS**	**Low AS**
Hits	Threat	3.5%	3.4%
	Neutral	1.7%	1.5%
	Overall	0.9%	0.8%
False Alarms	Threat	2.4%	1.7%
	Neutral	1.1%	0.8%
	Overall	0.6%	0.4%

1.6.   Study 1 Figures

The subliminal Emotional Stroop Task: Respond to the colour of the letters (mask)

Time (ms)

X&XX%XX%
+

CANCER
+

+

Fixation (1700–2300) ms

%&XX&X%%
+

Mask (300 ms minus individual perception threshold)

PANIC
+

Stimulus (individual perception threshold in ms
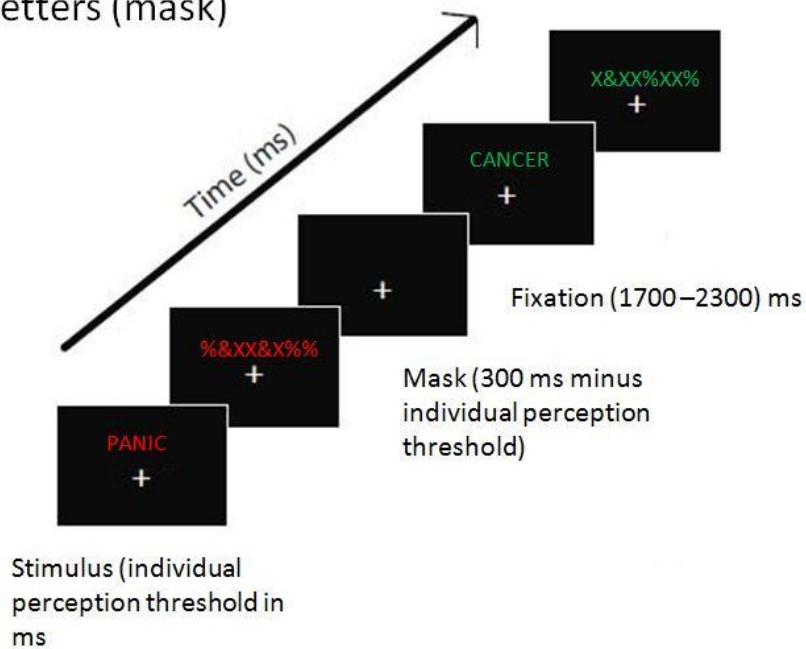
Figure 1-1:   Illustration of the subliminal emotional Stroop task.

The Emotional Stroop Task: Respond to the colour of the word

Time (ms)

PANIC
+

CLOCK
+

ULCER
+

+

JOURNAL
+

Stimulus (300 ms)

+

WORRY
+

+

Fixation (1700 –2300) ms

Threat Words

HELMET
+

Neutral Words
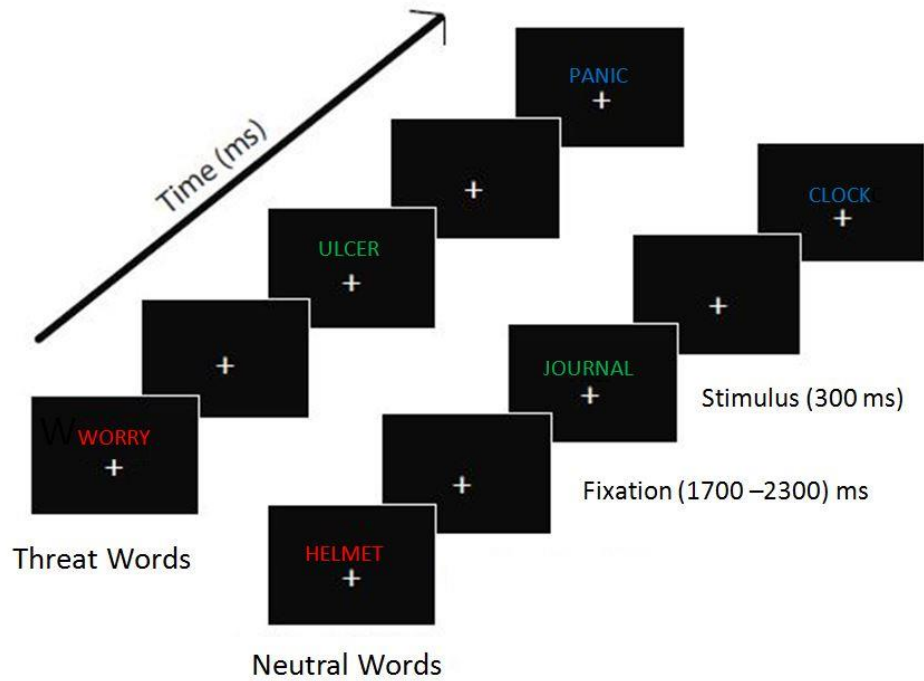
Figure 1-2:   Illustration of the supraliminal emotional Stroop task

Mean Reaction Times Supraliminal

■ Emotional Word ■ Neutral Word

High ASI group

Low ASI
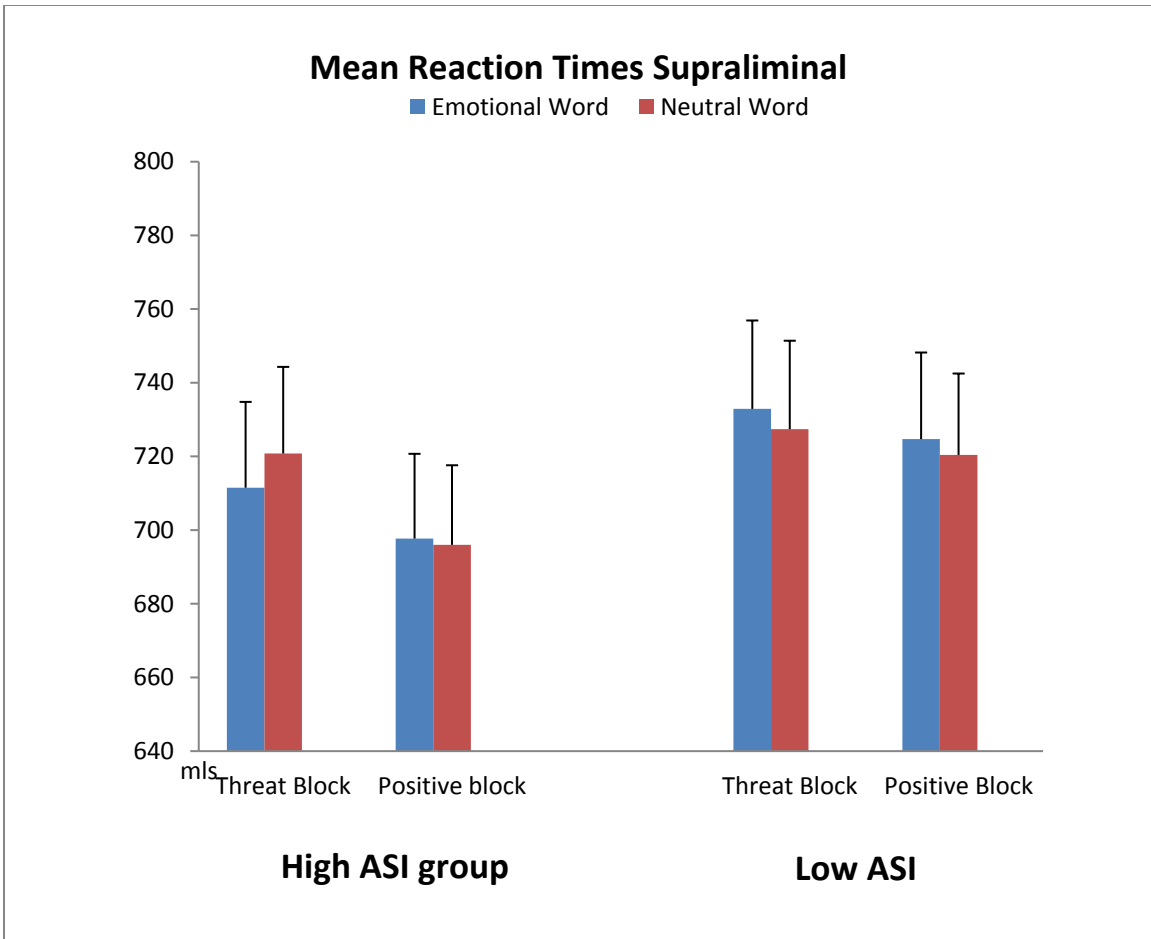
Figure 1-3: **Mean RT as a function of Group, Emotional Block, and Word Type in the supraliminal Stroop task.**
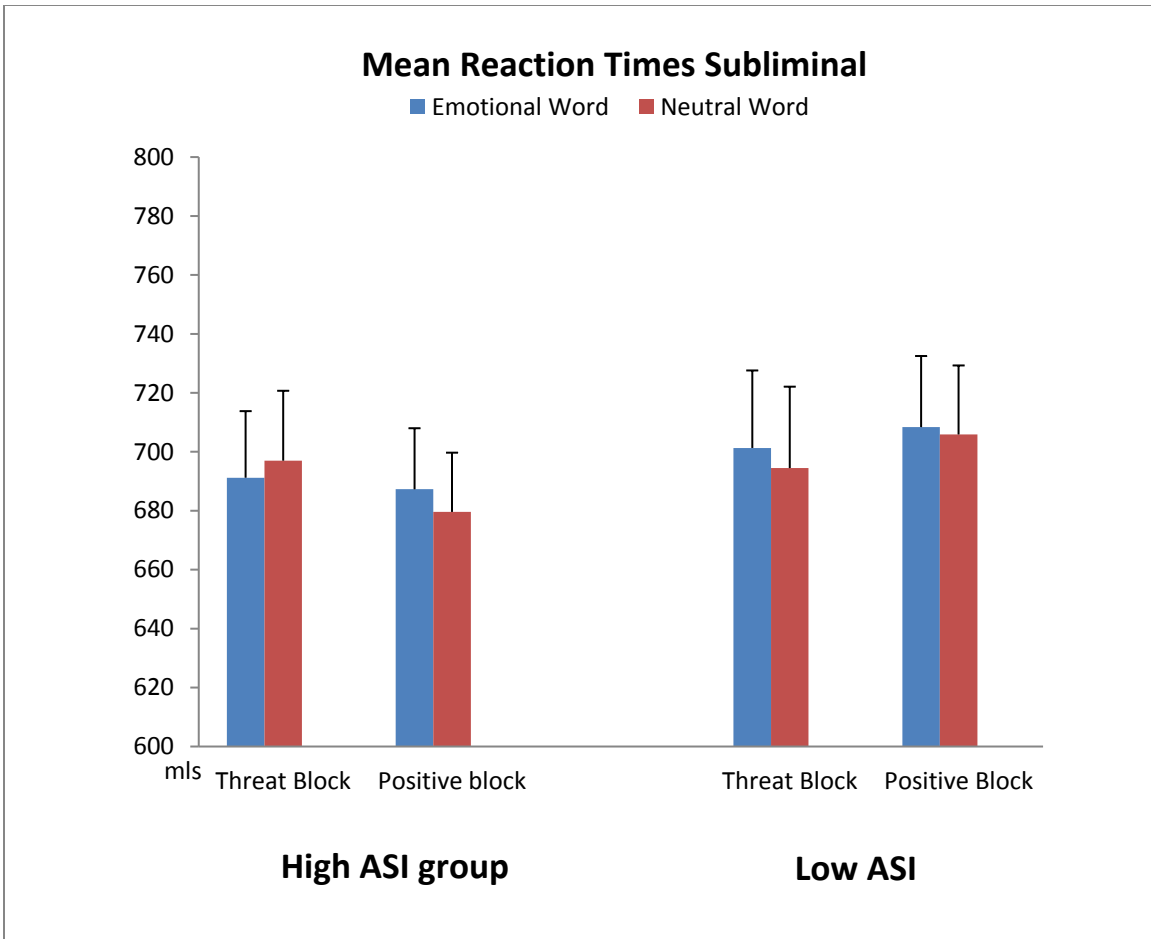
Figure 1-4: **Mean RT as a function of Group, Emotional Block, and Word Type in the subliminal Stroop task.**
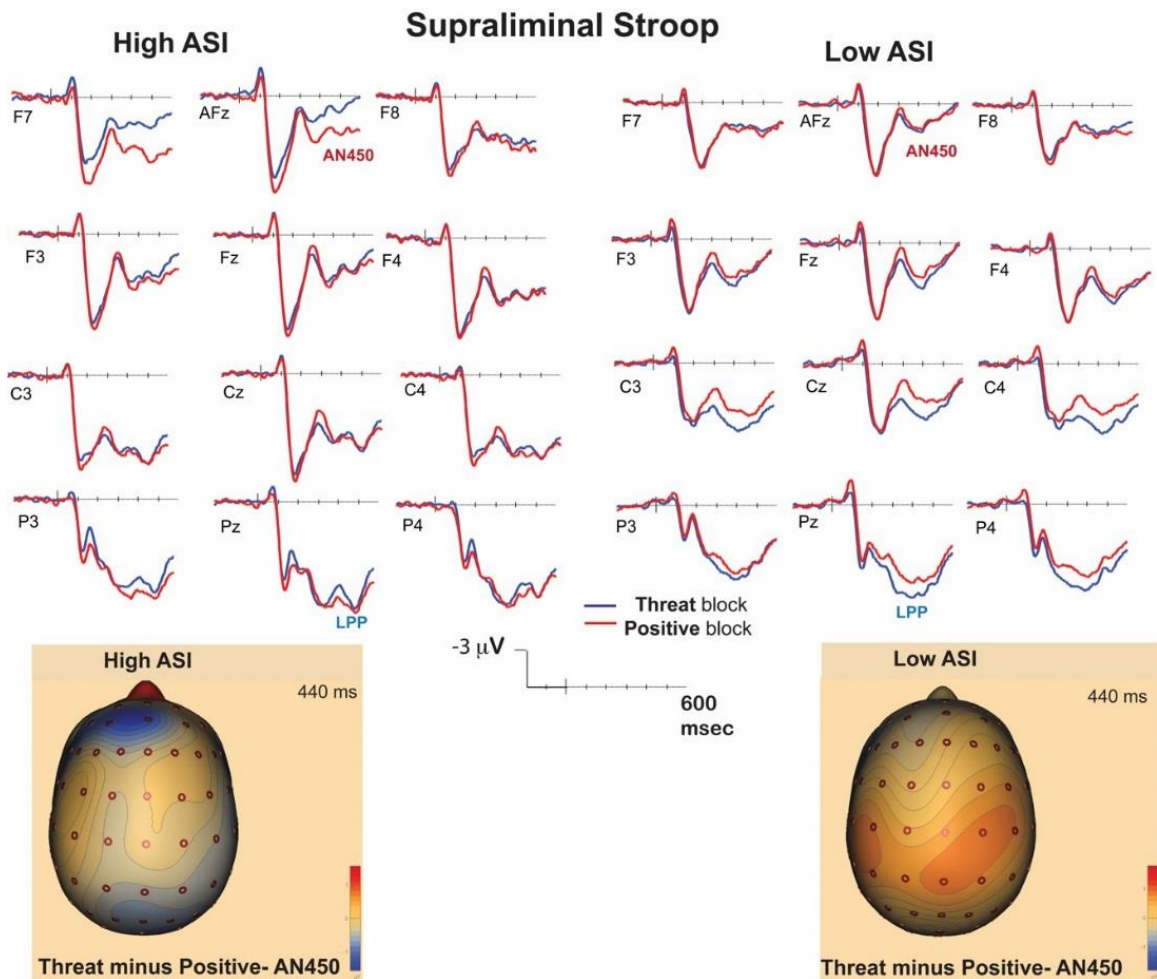
Figure 1-5: Grandaverage waveforms from the Supraliminal Stroop task for the Threat Blocks and Positive Blocks at 12 representative scalp sites over frontal, central, parietal, and occipital scalp for the High Anxiety Group and the Low Anxiety Group. Scalp topographical maps of the Threat minus Positive block difference wave in the 350 – 500 ms time window (AN450). Left: High Anxiety Group; Right: Low Anxiety Group.
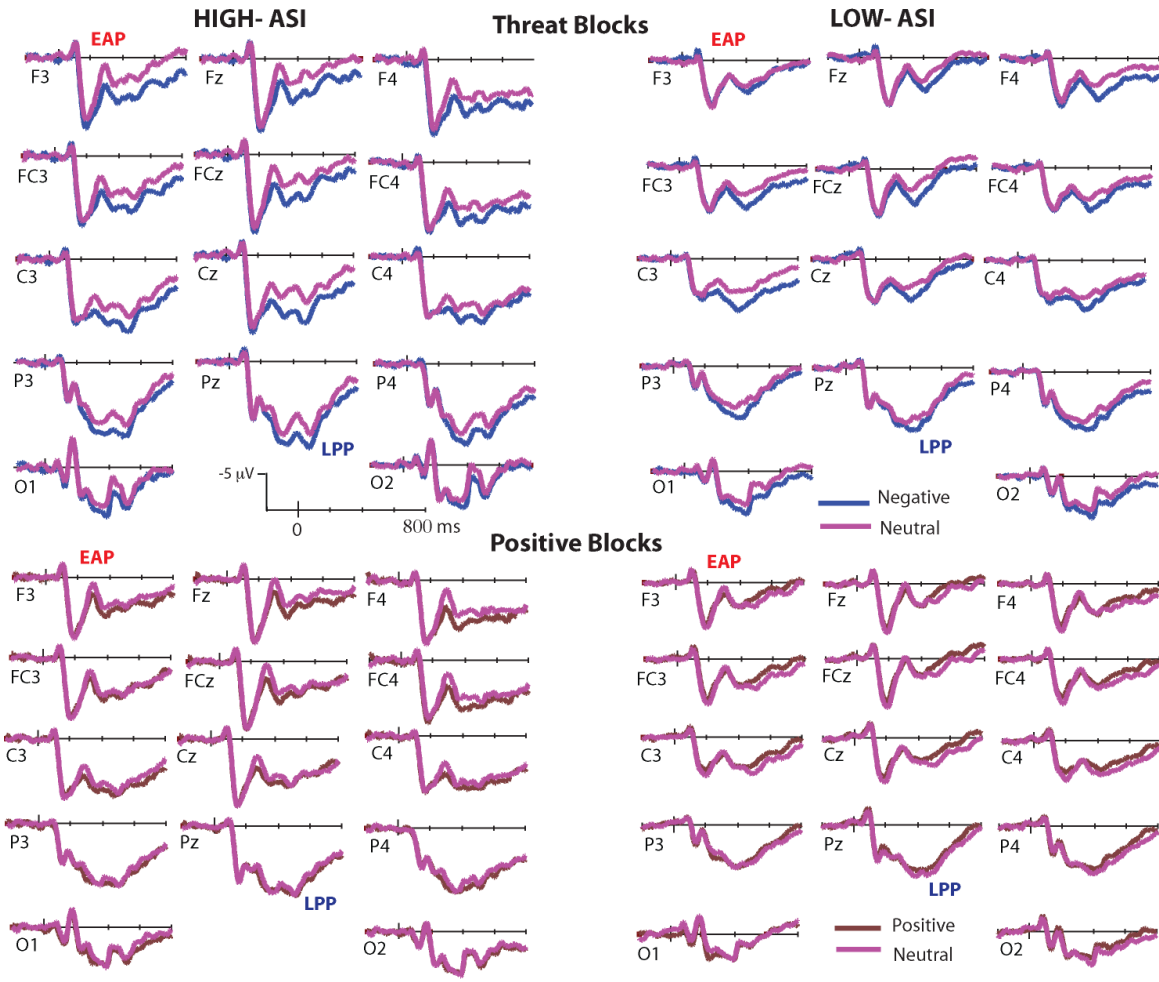
Figure 1-6: **Grandaverage waveforms from the Supraliminal Stroop task for the Threat Blocks and Positive Blocks at 14 representative scalp sites over frontal, central, parietal, and occipital scalp for the High Anxiety Group and the Low Anxiety Group.**

36
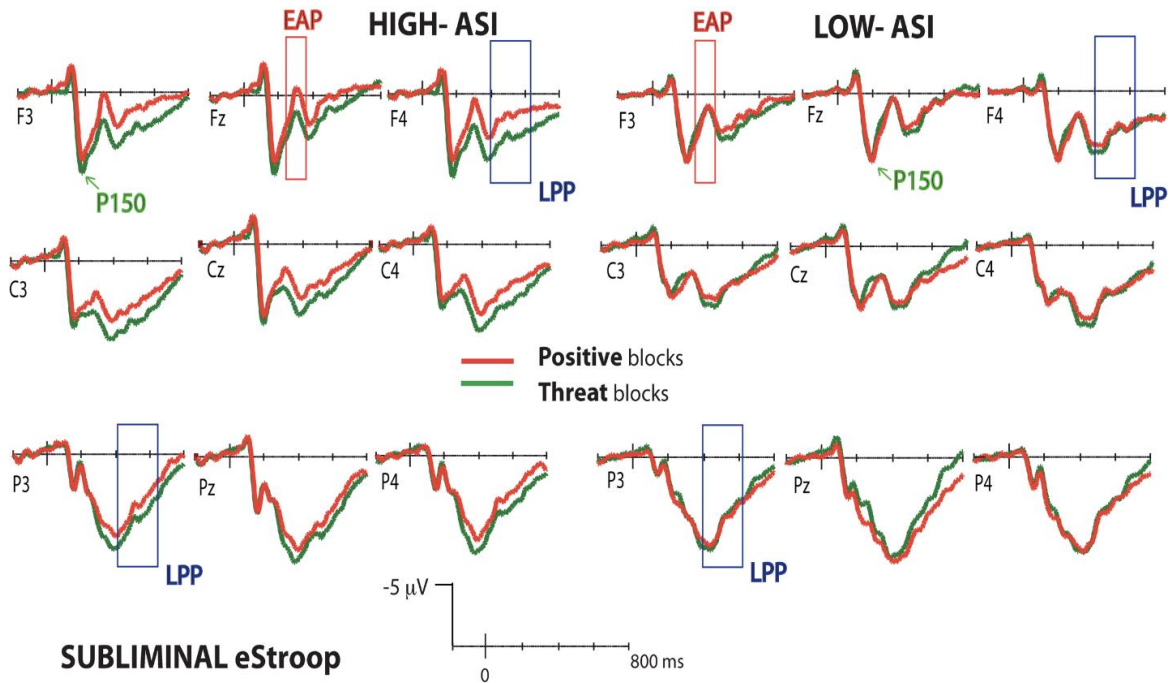
Figure 1-7: Grandaverage waveforms from the Subliminal Stroop task for the Threat Blocks and Positive Blocks at 9 representative scalp sites over frontal, central, parietal, and occipital scalp for the High Anxiety Group and the Low Anxiety Group.

Subliminal Stroop
Threat block minus Neutral block
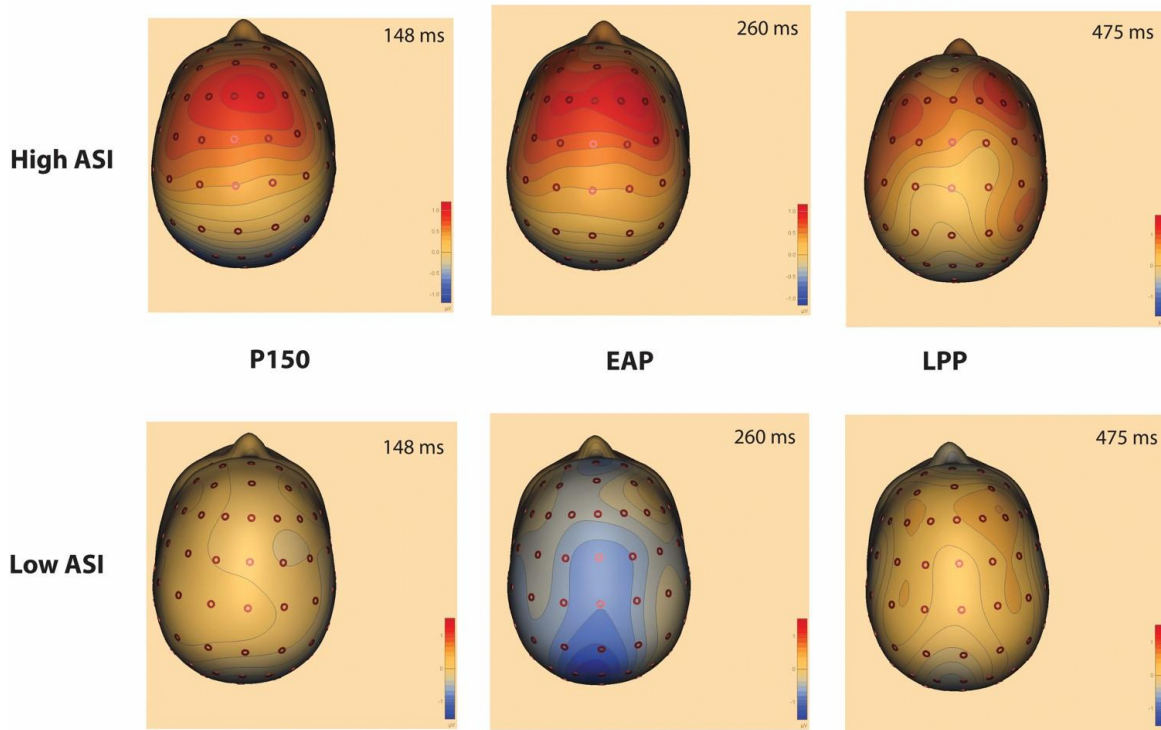
Figure 1-8: Scalp topographical maps of the Threat minus Neutral difference wave within the Threat Blocks in the subliminal Stroop task in the 130 – 200 ms time window (P150), 200-320 ms time window (EAP), and 350- 550 ms time window (LPP). Top: High Anxiety Group; Bottom: Low Anxiety Group

Chapter 2.   Neural correlates of Threat and Emotion processing in Panic Disorder

2.1. Introduction

2.1.1. Panic Disorder: An overview

Panic Disorder is a psychiatric disorder characterized by unpredictable attacks of intense fear accompanied by severe physical anxiety symptoms (e.g. palpitations), often followed by a persistent fear of having another attack in the future. It affects about 2% of the general population. Panic Disorder is a serious, chronic condition with a high degree of impairment on the patient's life. Since many of the symptoms of Panic Disorder resemble those of clinical diseases, Panic Disorder patients frequently use medical services and undergo costly diagnostic procedures to rule out conditions such as heart disease (Katon, 1996). This places not only a financial burden on the medical system, but also entails social and functional impairment of the affected individual. Patients show fear of bodily sensations (e.g. dizziness, breathlessness), as they have been characterized as being prone to misinterpreting harmless body feelings, but also external stimuli as dangerous (Katon, 1996).

A variety of different theories attempt to explain the cause(s) of Panic Disorder, ranging from psychodynamic theories, to cognitive hypotheses, to biological models of the disorder. *Psychodynamic theories* argue that susceptibility to Panic Disorder is determined by personality traits and the extent of one's defensive style, which act as a predisposition to anxiety. Other predispositions, or prerequisites can be early life events, and one's relationship and attachment style with early caregivers, which together give rise to an unconscious underlying conflict. According to this view, anxiety or a panic attack is then an attempt to avoid experiencing the underlying conflict (Fava & Morton, 2009).

There have been several theories developed within a *cognitive framework*. For example, *Clark* argues that a Panic Disorder patient misinterprets certain bodily sensations as impending danger, which then appear to provoke panic attacks. For instance, palpitations might be viewed as evidence of a heart attack. Within this framework, the trigger stimulus leading to a panic attack can be external (e.g. a certain

location) or internal (a body sensation). Other influential cognitive theories entail the model by *Bandura*, who focused on an individual's perceived ability (called self-efficacy) to cope with threat, and *Beck's* model, who focused on a person's sense of vulnerability, in addition to patients' catastrophic misinterpretations of body sensations (Fava & Morton, 2009). A recent new Integrative Cognitive Model of Panic Disorder by *Casey* tries to combine the theories by Bandura, Clark, and Beck, and focuses mainly on self-efficacy, and catastrophic misinterpretations of body sensations. According to this view, the level of self-efficacy can influence the susceptibility to high arousal, which starts the panic cycle (Fava & Morton, 2009). Finally, *McNally* has concentrated on the role of Anxiety Sensitivity in the development of Panic Disorder. As discussed previously, AS scores predict future panic attacks and panic symptoms, and have been shown to be a dispositional variable elevated in people with Panic Disorder. According to McNally, AS is the fear of anxiety sensations per se, and therefore catastrophic misinterpretations of body sensations are not necessarily part of the vicious cycle of Panic Disorder in McNally's model (Fava & Morton, 2009).

Biological models of the causes of Panic Disorder usually focus on heritability and neurological abnormalities. Panic Disorder seems to run in families, but it seems that the predisposition for Panic Disorder is being inherited, and not the disorder itself. The Neuroanatomical hypothesis of Panic Disorder proposes that a fear network in the brain, consisting of the amygdala, prefrontal cortex, insula, thalamus, brainstem and hypothalamus, is reacting oversensitively in Panic Disorder patients which results in the symptoms of the disorder (Fava & Morton, 2009). Abnormalities in this fear network might be further underlined by the irregular functioning of several neurotransmitters and their systems, in particular the noradrenergic system, the neuropeptide Orexin, Neuropeptide S, and Adenosine, which are all linked to the Locus coeruleus, which in turn has connections to the attention and memory systems in the brain (Geiger, Neufang, Stein & Domschke, 2014).

The most important treatments with empirical support for Panic Disorder are Cognitive Behavioural Therapy (CBT) and pharmacotherapy (Fava & Morton, 2009). The combined treatment of these two has been shown to be superior to any treatment alone: While CBT seems to address the underlying cause of Panic Disorder, namely focusing on the misunderstanding of body symptoms and apprehension of panic, pharmacological treatments affect the fear network in the brain including its neurotransmitters.

2.1.2. Panic Disorder and emotion processing: Behavioural results

Behaviourally, Panic Disorder patients exhibit attentional biases to negative face and word stimuli (e.g. Asmundson et al., 1992; Reinecke, Cooper, Favaron, Massey-Chase, & Harmer, 2010; Ehlers, Margraf, Davies, & Roth, 1988; McNally et al., 1994; Maidenberg, Chen, Craske, Bohn, & Bystritsky, 1996), endorse more panic-related and negative self-attributions (Reinecke et al., 2011), preferentially interpret negative information in an ambiguous manner (e.g. Lundh et al., 1999; Teachman, 2007), and display memory biases to negative information (Lim & Kim, 2005). These attentional biases to negativity and threat seem to be rapid and rather automatic and might be especially evident if the threatening stimulus is being presented subconsciously (Lim et al., 2005). Theoretical models of Panic Disorder, such as cognitive (e.g. Beck & Clark, 1997) and learning models (Bouton et al., 2001), suggest a role of biased-information processing to threat stimuli in general to explain these behaviours, perhaps stemming from an inability of successfully modulating automatic fear responses though conscious processes (Windmann, 2002).

2.1.3. Panic Disorder and Emotion processing: Electrophysiological Results

A few studies have attempted to shed light on the electrophysiological correlates of attentional biases in Panic Disorder patients. Findings on the most widely studied EEG component, the P300 waveform, have been mixed and altogether inconclusive. Some studies have reported shorter P300 latencies compared to controls (e.g. Hanatani et al., 2005), whereas others have found longer latencies (e.g. Thomas et al., 2013, 2014). There are several reports that have described reduced P300 amplitude compared to healthy controls (e.g. Thomas et al., 2013, 2014), and a few that found enlarged amplitudes (e.g. Pauli et al., 1997). A recent meta-analysis combining the published results on the P300 in Panic Disorder patients only found a trend in reduction in P300 amplitude and no other significant abnormalities (Howe, Pinto & DeLuca, 2014). However, major depressive disorder (MDD) is very often comorbid with Panic Disorder, and P300 amplitude reductions are a common finding in MDD (Zhou, Wang & Wang, 2018). Finally, P3 amplitude effects may depend on the type of stimuli- neutral (such as the original oddball task) – showing reductions, or emotional- showing increases-confounding the interpretation of meta-analytical approaches.

Employing emotional stimuli, other ERP components have been shown to be altered in Panic Disorder patients compared to healthy controls in paradigms investigating attentional biases to threat: The occipital P1, with either shorter latencies or augmented amplitude (Thomas et al., 2013), and a later sustained positive slow wave (600-800 ms) at parietal scalp sites, likely an LPP (Pauli et al., 1997). Interestingly, one study (Pauli et al., 2005) found earlier effects (100-200 ms, and 200-400 ms) at frontal electrodes in Panic Disorder patients, but only when stimuli were presented below conscious threshold. Early ERP effects at frontal sites in patients were also found in a memory study using an old/new paradigm with neutral and threat words (Windmann, 2002). Intriguingly, Panic Disorder patients showed the same old/new ERP effect for neutral as to threat words in a time window of 300 to 500 ms at frontal sites; that is, they "responded as if they associated negative implications or consequences with the neutral words" (Windmann, 2002, p. 366). The researchers suggest that this effect might arise from deficits in the inhibition of fear signals stemming from a miscommunication between vmPFC and the amygdala (Windmann, 2002).

2.1.4. Panic Disorder and Emotion processing: fMRI studies

In recent years, several fMRI studies have tried to shed light on the neural underpinnings of emotional processing in Panic Disorder. A recent fMRI study employing the eStroop task with panic-related and neutral words in Panic Disorder patients and healthy controls reported increased activation of the left inferior frontal gyrus in response to panic-related words among Panic Disorder patients (Dresler et al., 2012). Some studies have reported decreased amygdala activation in Panic Disorder patients to negative stimulus materials (Pillay et al., 2006; Demenescu et al., 2006; Ottaviani et al. 2012), whereas other researchers have reported hyperactivity of this brain structure (e.g. Thomas et al., 2001; van den Heuvel et al., 2005). The amygdala is part of a network involved in emotion detection and regulation in healthy participants, besides the brainstem, thalamus, insula, cingulate and prefrontal cortex, which are also part of this circuitry. Increased activation of this network (e.g. Feldker, et al., 2016; Brinkmann et al., 2017) has been associated with a variety of anxiety disorders, including panic (Poletti et al., 2015). However, responses seem to depend on the specific task used, and therefore results of the previous studies vary. In a recent review of available brain imaging studies, Dresler et al. (2013) suggested as a consensus that a hyperactive amygdala seems to

be characteristic of the *state* rather than *trait* aspects of Panic Disorder. In general, it seems that a network that signals fear is abnormally activated in Panic Disorder. The fact that brain activation in cortico-limbic structures of Panic Disorder patients changes following successful treatment with cognitive-behavioural therapy further underlines this idea (Grambal, Hlustik, & Prasko, 2015).

2.1.5. The current study

The current project used the same paradigm as in study 1, consisting of the supraliminal and subliminal eStroop task, to extend the analysis of hypervigilance to threat signals to a group of Panic Disorder patients to find out more about the origin of typical physical anxiety symptoms in this group, and to compare the results to the high AS participants.

Hypotheses

For the behavioural analysis, we hypothesized that specific slowing of reaction time to threat words would only be present as a block effect (threat versus positive blocks) in the supraliminal tasks in the Panic Disorder patients only (e.g. Lundh et al., 1999; Li et al., 2007). We expected to find the same slowing of reaction time to threat material in the subliminal task in the patients, since some studies have reported emotional interference to threat stimuli in subliminal tasks in anxiety patients (e.g. Lundh et al., 1999; Thomas et al., 2013).

The following hypotheses were formulated with regard to the electrophysiological correlates of the eStroop tasks:

In the supraliminal task, based on the findings from Taake et al. (2009) in the high AS participants, we anticipated the following effects. First, under the assumption that trait anxiety and clinical disease state are both characterized by hypervigilance to threat producing an *early* attentional bias, we expected greater amplitude of the early frontal EAP to threat words than neutral words in the threat blocks. Finally, we predicted that the later LPP to threat compared to neutral words- reflecting conscious, deliberate processing of threat information- would be equally present in both Panic Disorder and healthy participants, in line with the emotional literature, and Taake et al. (2007).

More critical, however, were the predictions in the subliminal task. In agreement with the existing literature on hypervigilance to threat in Panic Disorder patients particularly for subliminal stimuli (e.g. Pauli et al., 2005), we predicted that in Panic Disorder participants, threat stimuli below perceptual threshold would elicit frontal positivities - possibly even earlier than the EAP. We did not expect AN380 or LPP effects to be present in the subliminal task due to the lack of conscious awareness or conflict generated by threat stimuli in either Panic Disorder or healthy participants.

2.2. Methods

The study involved two phases: A screening and experimental session. The Simon Fraser University Ethics Board approved all aspects of these experiments. All participants gave their informed and written consent before participating in this study and received a small monetary remuneration for their involvement.

2.2.1. Screening Session

Recruitment and Reimbursement

Seventeen patient participants were recruited through flyers posted at SFU, in Anxiety Clinics in the Lower Mainland, and through ads on Craigslist and local newspapers, such as Georgia Strait and North Shore News. The Anxiety Clinics were contacted for their permission to post the flyers, and potential participants contacted us directly.

Fourteen age and gender matched healthy participants composed the control group. Both SFU members and community members were recruited through flyers posted at SFU or through the method of 'snowballing'. We asked participants to contact people they suggested as third parties to ask whether that third party had any objection to the release of their name for contact and then we contacted them directly.
All participants were screened through the Anxiety Disorders Interview Schedule for DSM-IV. The ADIS-IV is a structured interview designed to assess for current episodes of anxiety disorders and to permit differential diagnoses between anxiety disorders, according to DSM-IV criteria (Dinardo, Moras, & Barlow, 1993). The ADIS-IV has demonstrated reliability and validity. Using this kind of structural interview decreases the information variance and the interviewer variance. It is also a quantifiable assessment,

because each diagnosed disorder is rated on a severity scale and can be used for comparisons between groups. The ADIS-IV was used to confirm the participants' diagnosis, and also to ensure that the control participants were indeed free of any disorder. Assessment of current mood, somatoform, and substance use disorders were also included because of their high rate of comorbidity with anxiety.

Furthermore, the following inclusion and exclusion criteria were exerted:

<u>Inclusion criteria</u>

- Normal or corrected-to-normal vision.

- Age: 19-55 years of age.

- For the experimental group: Patients met DSM-IV criteria for a primary diagnosis of Panic Disorder, with or without Agoraphobia, and with or without other comorbid anxiety disorders, either on psychoactive medication or drug-free.

- For the control group: Age and gender matched healthy individuals, without any current or life-time psychiatric diagnosis.

<u>Exclusion criteria</u>

- Color blindness, as this study involved identification of colored images and participants with color blindness might have had difficulty in such process.

- Language: Participants who did not speak English as a first language, due to the linguistic nature of the task.

- Other primary psychiatric, neurological diagnoses, or medical conditions, current or past, since these would have affected electrical signals in the brain.

The principal investigator was thoroughly and extensively trained on administration and scoring of the ADIS-IV by a practising clinical psychologist specialised in the diagnosis and treatment of anxiety disorders. Training entailed handling the different circumstances that can arise during psychiatric interviews, as well as several meetings on how to conduct the interviews and issues involved in the diagnosis of Panic Disorder.

Finally, the principal investigator conducted a practice interview on the clinical psychologist.

The current study took on average four hours to complete for patient participants (2 hours for the ADIS-IV and 2 hours for the EEG session), and three hours for the controls (1 hour for the ADIS-IV and 2 hours for the EEG session). Participants were reimbursed $20 per hour. In case of withdrawal from the study, they were compensated for the portion they completed. Minimum reimbursement was $20.

2.2.2. Experimental Session

The same procedures that were followed in Study 1 were employed in this study.

Threshold Determination Phase

The same procedures that were followed in Study 1 were employed in this study.

2.2.3. Behavioral Task

The tasks used in this study were the same as in Study 1.

2.2.4. ERP measures

The electroencephalogram (EEG) was recorded from 64 standard 10-20 scalp sites mounted in an elastic cap with active Ag/AgCl electrodes (Biosemi Active Two system, Amsterdam, NL). Six additional channels included four eye movement channels (two at the external canthi and two below each orbit) and two reference electrodes placed on each mastoid bone. EEG and EOG channels were digitized at a sampling rate of 512 Hz. Off-line processing was performed in BESA 5.3 (Brain Electric Source Analysis, Gräfelfing, Germany). Filter settings included a highpass zero-phase filter set at 0.1Hz (12dB/oct), and a low-pass filter of 30 Hz. ERPs were timelocked to stimulus onset and baseline corrected to the mean voltage of the 200 ms pre-stimulus interval. Trials containing blinks and eye movements were removed prior to ERP averaging by applying a semi-automated artifact detection routine based on amplitude at eye movement channels implemented in BESA (thresholds were individually adjusted). A total of four participants (3 patients for the supraliminal analysis, 2 patients for the

subliminal analysis, and 1 control participant for both analyses) were excluded for either excessive blinking or technical difficulties while recording behavioural performance, leaving a total of 14 patients and 13 control participants for the supraliminal analysis, and a total of 15 patients and 13 control participants for the subliminal analysis.

2.2.5. Behavioural Data Analysis

The behavioural data analysis has also been previously described (See Study 1).

2.2.6. ERP Data Analysis

The ERP data analysis procedures were the same as in Study 1.

2.2.7. Statistical Analyses

For the statistical analysis, mixed-design ANOVAs were employed with between factor being group (patients vs. controls), and within factor being emotional block (threat vs. positive), and trial type (emotional vs. neutral). Global analyses were followed by more restricted ANOVAs and t-tests upon significance of interactions, or when a priori hypotheses for group differences were present. For all analyses, the critical p-value was set at 0.05, and degrees of freedom were corrected with the Greenhouse–Geisser epsilon method to correct for sphericity (Greenhouse and Geisser, 1954).

2.3. Results

2.3.1. Clinical and demographic variables

Demographic characteristics of the participant groups are summarized in Table 2-1. The patient and control participants did not differ on age [$t_{(1\ 27)}$= .06, p=.952)], education [$t_{(1\ 23)}$= -1.9, p=.07)], or handedness (all participants were right-handed). However, there was a greater number of women overall (28 female vs 3 male participants, $\chi^2_{(1)}$ = 20.16, p = 0.000), as well as within the patient participant group (16 female vs 1 male participants, $\chi^2_{(1)}$ = 13.24, p = 0.000), and within the control group (12 female vs 2 male participants, $\chi^2_{(1)}$ = 7.14, p = 0.008. Not surprisingly, patient participants reported significantly higher scores on the ASI [$t_{(1\ 27)}$= 5.86, p=.000)], the

47

BDI [$t(1\ 24)= 2.73$, p=.011)], the STAI-S [$t(1\ 27)= 3.52$, p=.001)], and the STAI-T [$t(1\ 25)= 3.76$, p=.001)].

2.3.2. Behavioural Results

Threshold Determination Phase

Table 2-2 depicts separately for panic patients and controls the mean perception thresholds and frequency of which each perception threshold occurred within each group. The patients and controls did not differ on the mean perception threshold [$t(1,26)=-.91$, p=.371]. The most common threshold was 13 ms.

Supraliminal Task Reaction Times

In the mixed design ANOVA, there was a marginally significant main effect of emotional block [$F(1,29)=3.86$ p=.058]. All participants responded on average 14.5 ms slower to threat blocks than to positive blocks. Such slowing in the threat blocks relative to the positive blocks was independent of type of stimulus, being present for both threat and neutral words. In spite of the RT interference being on average larger for the patient than the control group (17 ms vs. 12 ms), the Group X Emotional Block interaction did not approach significance, $F(1,29)=.132$ p=.72 and t tests run within each group failed to reach significance (patient group: t [(1,17)=1.53, p=.145], control group: t [(1,12)=1.59, p=.139]. Table 2-3 and Figure 2-1 illustrate the reaction times.

Subliminal Task reaction times

In the mixed design ANOVA, there was a significant main effect of emotional block [$F(1,29)=6.04$ p=.021]. All participants responded on average 16.06 ms slower to threat blocks than to positive blocks. Such slowing in the threat blocks relative to the positive blocks was independent of type of stimulus, being present for both threat and neutral words. In spite of the three-way interaction not being significant ($F(1,27)=2.62$ p=.117), upon inspection of the means, we proceeded with restricted analyses within group.

In the patient group, the interaction of emotional block x type of trial was significant, $F(1,15) = 5.16$, p =0.39. Patients responded on average 18.1 ms slower to threat words (742.1 ms) than to neutral words (724.0) within the threat blocks [$t(1\ 15)=$

3.01, p=.009)]. No such slowing was present in the positive blocks [t (1 15)=-.369, p =.717]. In contrast, in the low AS group, no significant effect of emotional block was present, F (1, 12) = 4.41, p = .058, and the interaction of emotional block x type of trial was also not significant, F (1, 12) = 0.063, p= 0.806. There was no RT difference as a function of emotional block or trial type. See Table 2-4 and Figure 2-2.

Recognition Task

Percentages of correctly recognized words and false alarms as a function of group and word type are displayed in Table 2-5 for each group. As can be seen, the percentage of words correctly recognized from the subliminal task was very small, and therefore it is very unlikely that the consciously recalled words exerted a great influence onto the behavioural or ERP effects.

2.3.3. ERP Results

Supraliminal task

Within Block Effects

A later latency, posteriorly distributed positive slow wave (LPP, peaking around 400 ms over parietal scalp) displayed greater amplitude for threat than neutral words. Such LPP modulation appeared to be present in the patient group only. For the LPP window (350-550 ms), a posterior parietal ROI was chosen, collapsing sites C1/C2, C3/C4 and CP1/CP2.

Parietal LPP (350-550 ms)

There was a significant interaction of block x type of trial x group [F (1,25)=4.432, p=.045] in the global ANOVA. No main effect or other interaction approached significance (for all p > .10). Within the patient group, there was a significant interaction of block x type of trial [F (1 13)=7.434, p=.044]. Mean amplitude of the LPP over posterior scalp was greater in response to threat than neutral words in the patients (threat words: 7.22± .64 µV; neutral words: 5.89 ± .69 µV) t [(1 13)=2.99, p=.010]. In contrast, in the positive blocks, there was no difference between positive and neutral words (positive words: 6.55± .99 µV; neutral words: 6.67 ± 1.00 µV) t [(1 13$)$ =-.234, p=.819]. These effects are illustrated in Figure 2-3.
Within the control group, there were no significant main effects or interactions (for all p >

.10). LPP amplitude did not differ as a function of emotional trial (threat words: 4.32 ± 1.44 μV; neutral words: 4.68 ±1.19 μV) t [(1 12)=-.53, p=.61].

Subliminal task

Block Effects

First, the evoked response to words in the threat blocks relative to words in the positive blocks revealed greater amplitude of a unilateral positive wave with frontal distribution over the left hemisphere, peaking between 200 and 320 ms (here called the Early Anterior Positivity, the EAP), and a bilateral positive wave with frontocentral distribution, peaking between 350 and 550 ms (here called frontal LPP). Such effects were only observed in the panic patient group. Anterior lateral regions of interest (ROIs) were selected, by averaging together electrode sites for each effect where the voltage difference was maximal. For the EAP, sites AF7/AF8, AF3/AF4, F3/F4 and F5/F6 were used, and for the frontal LPP, the ROI consisted of electrodes F1/F2, FC5/FC6, FC3/FC4, and FC1/FC2.

Secondly, the evoked response to words in the threat blocks relative to words in the positive blocks revealed greater amplitude of a bilateral positive wave with parietal distribution, peaking between 350-550 ms (here called the LPP). Such effect was also only observed in the panic patient group. For this analysis, parietal lateral regions of interest were selected for each hemisphere, by averaging together electrode sites C1/C2, C3/C4, CP3/CP4 and CP1 and CP2.

Early Anterior Positivity (EAP 200-320 ms)

In the mixed design global ANOVA, there were no significant main effects or interactions (for all p > .10).

Frontal LPP (350 – 550 ms)

In the mixed design ANOVA, there was a significant main effect of Emotional block [F (1,26)=7.38, p=.012] and a significant interaction of Emotional block x patient group [F (1,26)=6.47, p=.017]. In the patient group, mean amplitude of the EAP over frontal scalp was greater in response to threat blocks relative to positive blocks (threat blocks: 4.05 ± 1.62 μV; positive blocks: 1.37 ± 2.08 μV) [F (1,14)=11.11, p=.005]. In contrast, in the control group, EAP amplitude did not differ as a function of emotional

block (threat blocks: 1.36 ± 1.07 µV; positive blocks: 1.28 ± 1.09 µV) [$F_{(1,12)}$=.023, p=.883]. These effects are illustrated in Figure 2-4. No other main effects or interactions approached significance (p>.10).

Parietal LPP (350 – 550 ms)

In the mixed design ANOVA, there was a significant main effect of Emotional block [$F_{(1,26)}$=6.08, p=.021]. The interaction of emotional group x block approached significance [$F_{(1,26)}$=3.31 p=.080, and given our apriori hypothesis of a selective parietal enhancement to threat blocks in the patient group, and based on visual inspection of the waveforms, we proceeded with within-group analyses. In the patient group, mean amplitude of the LPP over bilateral parietal scalp was greater in response to threat blocks relative to positive blocks (threat blocks: 6.49 ± 1.52 µV; positive blocks: 4.24 ± 2.09 µV) [$F_{(1,14)}$=6.12, p=.027]. In contrast, in the control group, LPP amplitude did not differ as a function of emotional block (threat blocks: 5.03 ± 1.57 µV; positive blocks: 4.69 ± 1.61 µV) [$F_{(1,12)}$=.691, p=.422]. These effects are illustrated in Figure 2-4. No other main effects or interactions approached significance (p>.10).

2.4. Discussion

Behavioural and electrophysiological correlates of threat-related emotional processing were explored in Panic Disorder patients and healthy controls using a supraliminal and subliminal version of the eStroop task.

Behaviourally, in the supraliminal task, a marginally significant slowing of RT to threat words was present as a block effect (threat versus positive blocks, a 'slow' effect). Such weak emotional interference was present in both groups. Interestingly however, in the subliminal task, only panic patients were selectively slower to respond to threat relative to neutral words within the threat blocks (a 'fast' effect).

Several threat-specific ERP effects was found. In the supraliminal task, the only significant effect was a selectively greater amplitude modulation for the posterior LPP (350-550 ms) for threat relative to neutral words for panic patients only. In the subliminal task, as hypothesized, threat-related ERP modulations for the threat blocks were identified over anterior bilateral frontal scalp, in the form of a *positive* polarity enhancement in response to threat blocks relative to positive blocks, peaking at a time

window between 350 and 550 ms (frontal LPP). This effect was only present in the patient group. Finally, a block effect (Threat versus Positive Blocks) was also found as a modulation of the late positive potential (LPP) over posterior scalp (350 ms- 550 ms), being marginally significant in the patient group only.

Behavioural findings

Panic Disorder and control participants were found to have similar <u>perception thresholds</u> and they were comparable in terms of the percentages of correctly recognized words and false alarms in the subliminal Stroop task, with both <u>Hits and False Alarms</u> at chance rate in both groups.

<u>Reaction times:</u> There was a marginally significant main effect of emotional block in the *supraliminal task* which was independent of type of stimulus, being present for both threat and neutral words. As in Study 1, both groups displayed a similar tendency to respond slower to the threat blocks, with no difference between the two. This result is not in line with several other studies showing a specific attentional bias in Panic Disorder (e.g. Dresler et al., 2012; Mogg et al., 1993; Lundh et al., 1999). Consider, however, that a recent meta-analysis by Bar-Haim et al. (2007) concluded that the only attentional task capable of producing emotional interference in non-anxious healthy controls is a *block* design of the eStroop task, so it is not surprising that the control group in our study behaved similarly to the Panic Disorder group, and both contributed to the observed marginal interference effect.

Subliminal task: There was a specific slowing of reaction time within the threat blocks: Patients responded on average 18.1 ms slower to threat relative to neutral words. No such effect was found in the control group. In light of the fact that the patients did not differ in the number of Hits and False Alarms in the recognition test, and therefore did not consciously distinguish between the threat and neutral words, this finding is quite surprising and was not expected. However, behavioural interference in participants with clinical anxiety in subliminal paradigms has been reported before. Mogg et al. (1993) conducted a subliminal and supraliminal eStroop task in three groups of participants (Generalized Anxiety Disorder patients, Depression patients, and healthy controls), and reported larger interference scores for negative words in the anxiety patients in the subliminal task. The researchers used a mixed trial design, with each block containing all the different categories of words, so it was impossible for the

participants to anticipate the order of stimuli. Furthermore, just like in our design, their study included an awareness check which confirmed that participants' recognition of the stimuli was at chance level. Similarly, Lundh et al. (1999) conducted a subliminal eStroop task in Panic Disorder patients and healthy controls and reported interference in patients for panic-related words, as well as Lim & Kim (2005), who found interference in panic patients for physical threat and negative words in a subliminal Stroop experiment. Hence, our results are consistent with other studies, and support the notion that in clinical anxiety, attentional biases are automatic and do not depend on conscious awareness of the stimuli.

ERP findings

Supraliminal task – posterior LPP. Mean amplitude of the *LPP* (350 ms – 550 ms) over posterior scalp was greater in response to threat than neutral words in the patient group in the *supraliminal Stroop* task. This result is in line with a large body of literature, showing that emotionality and valence of visual stimuli (words and faces) are reflected in modulations of this component. A few studies conducted in Panic Disorder patients are in line with these findings as well. For instance, Pauli et al. (2005) presented threat and neutral words to Panic Disorder and healthy participants, with emotion being task-irrelevant. Threat words elicited greater amplitude LPPs (400-600 ms) than neutral words in the patients. Furthermore, Thomas et al. (2013) conducted an eStroop task in Panic Disorder patients, Obsessive-Compulsive Disorder patients, and healthy controls, and found increased P300 amplitudes to threat versus neutral stimuli in those participants across groups who showed Stroop interference. Unexpectedly, and contrary to our findings in study 1, Taake et al. (2009), and some studies in the current literature that used the eStroop task in healthy participants (e.g. Thomas et al., 2007; Thomas et al. 2013), the control group did not show increased amplitude of the LPP component to either class of stimuli. We speculate that our small sample size (n=13) of the control group may at least contribute to this null finding.

More importantly, our hypothesis regarding a frontal *EAP* in the patient group in both the supraliminal and subliminal task was not confirmed. We had formulated this hypothesis based on the assumption that *trait* anxiety and clinical disease *state* are both characterized by hypervigilance to threat exemplified in an *early* attentional bias in both the supraliminal and subliminal task. A different explanation is now needed.

In first place, lack of EAP modulation in the control group could index a relatively low concern for threat stimuli, e.g., no attentional bias. In fact, our control participants scored low on the ASI (M=10.6), and although unselected for AS status in study 2, they were comparable to the low AS group of study 1 (M=10.3). Recall that in the supraliminal task in study 1, a selective frontal EAP modulation to threat words was present in the high AS group only.

A different explanation is necessary for the lack of EAP modulation for Panic Disorder patients in both tasks. Biological theories of Panic Disorder have speculated that the prefrontal cortex in patients cannot inhibit a neural circuit relevant for anxiety that is associated with amygdala hyperactivity (Ohta et al., 2008). Hence, according to this view, *hypoactive* prefrontal cortical functioning is expected in Panic Disorder patients and might represent the "pathophysiological core of Panic Disorder (Ohta et al., 2009, p.1058), reflecting inadequate top-down control. In fact, several brain imaging studies have detected hypofrontality in Panic Disorder patients (Ohta et al., 2008; Ball, Ramsawh, Campbell-Sills, Paulus & Stein, 2013; Reinecke et al., 2015), supporting the hypothesis that PFC recruitment is broadly impaired in Panic Disorder. On the other hand, it has been reported that high trait anxiety individuals displayed *greater* PFC activation during an emotion regulation task compared to healthy controls (e.g. Ball et al., 2013). The failure in Panic Disorder patients to "engage PFC during emotion regulation may be part of the critical transition from dispositionally high anxiety to an anxiety disorder" (Ball et al., 2013, p. 1475), and could explain the fact that the high AS individuals in study 1 displayed an EAP in both tasks. "Heightened PFC activation in anxious, non-clinical adults could be a compensatory mechanism in individuals with dispositionally high anxiety" (Ball et al., 2013, p. 1483).

There is an alternative explanation regarding the absence of the EAP in the patients. There are some studies that suggest that at the core of Panic Disorder lies a hypoactive amygdala instead of a hyperactive one. For instance, in a study where Panic Disorder patients had to identify fearful faces, the patients' amygdala was significantly less activated in comparison to controls (Pillay et al., 2006). Furthermore, rapid habituation of the amygdala in response to threatening stimuli has been reported (Phelps et al., 2001), and prolonged exposure to phobic stimuli extinguished activity in the amygdala (Frederikson et al., 1995). A possible explanation for these findings might be that chronic hyperarousal in Panic Disorder patients to negative stimuli might diminish

emotional responses, and therefore reduce amygdala responsiveness (Ottaviani et al., 2012). Considering our findings, it is therefore possible that reduced responses in the amygdala in the patients made the recruitment of PFC for top-down control unnecessary, therefore leading to our EAP null finding.

Subliminal task. Mean amplitude over frontal scalp was greater in response to threat compared to positive blocks in the patients only, in a time window of 350-550 ms (frontal LPP). A similar result was obtained in a study where physical threat and neutral words were randomly presented below individual perception threshold to Panic Disorder patients and healthy controls in a mixed-trial paradigm (Pauli et al., 2005). Subjects read each word after a 3-sec delay. A frontal positivity (200-400 ms) distinguished threat from neutral words in both groups, being more pronounced in Panic Disorder patients. The authors concluded that panic patients are characterized by an elaborate processing of disorder relevant stimuli.

The presence of a *parietal LPP (350-550 ms)* in the patients was unexpected and surprising, although it has been reported before (Pauli et al., 1997). This effect might reflect conscious attempts for attempted top-down control, as this component is indicative of conscious evaluation and regulation. The patients might try to adopt conscious strategies to minimize the impact of their processing abnormalities (Windmann et al., 2002), although this notion is speculative. It is also possible that increased emotional arousal signalled by an overactive amygdala not checked by mPFC in Panic Disorder patients may cause cortical circuits typically devoted to deliberating cognitive processing in association areas (the generators of the frontal and parietal LPP) to be abnormally activated, even in response to subliminal presentations of disorder relevant stimuli (similar to Pauli et al., 2005).

2.4.2. Limitations

Notable limitations to the current study include the restricted and small sample size. Only 14 participants with Panic Disorder and 13 controls completed the supraliminal EEG portion of the task with an adequate number of correct responses to be included in further analyses, and there was only a total of 15 patient and 13 control participants for the subliminal analysis. This is obviously a smaller sample size that was originally intended and desired. Due to the nature of Panic Disorder and the resulting

challenges patients must face (e.g. some patients have difficulty leaving their house, or driving over a bridge, etc.), it was difficult to recruit enough participants that were able and willing to make the journey to the SFU campus to be involved in the experiment. Therefore, we might not have had sufficient power to detect subtle group differences and effects, potentially resulting in Type II errors. This problem of sample selection and low power may account for some of the null results in this study.

Furthermore, the results of this study may have limited generalizability, since the panic patient group consisted almost entirely of women. Hence, the results may not translate to male patients.

Another limitation of the study is the fact that the patient group included some participants that were medicated for their anxiety (35%). Again, due to the difficulty of recruiting the panic patients, it was not possible to find an entire medicated or non-medicated sample. Anderer, Saletu, Semlitsch and Pascual-Marqui (2002) reported that use of the sedative Lorazepam can prolong P300 latencies, and that the SSRI antidepressant Citalopram can enhance P300 source strength in a widespread area of pre-frontal cortex, indicating increased cortical activity in this brain region. In contrary, a study by van Laar et al. (2001) found no effect of the SSRI Paroxetine on P3 measures, but instead that the 5-HT2 receptor antagonist nefazodone increased reaction times. Hence, since the patient sample included one patient who reported use of Lorazepam on a PRN basis, three patients medicated with Citalopram, and one medicated with the 5-HT2 receptor antagonist trazadone, it is possible that the use of these drugs induced differential pharmaco-EEG and performance than would not have been observed otherwise in these patients. However, since four patients constituted less than half of the panic patient sample, the principal investigator considers the possibility that these induced pharmaco-EEG effects are responsible for the observed P300 differences between the patients and controls as relatively small.

Finally, even though the principal investigator had been extensively trained on how to conduct the diagnostic interviews using the ADIS-IV, no inter-rater reliability testing was part of this training. However, all patients of the study sample had been diagnosed with Panic Disorder before by either a physician or psychologist, and the diagnosis was then confirmed by the principal investigator, so the chance of having

included a patient with a false diagnosis of Panic Disorder is viewed as being relatively small by the first author.

Lastly, as in Study 1, the two tasks could have been analyzed in one overall ANOVA, by including task (subliminal vs. supraliminal) as a within-subject factor in such analysis. We decided against this design to avoid the difficulty of teasing apart interactions with too many factors. Furthermore, we were using multiple statistical tests without correction for multiple comparisons, which increases the chance of erroneous inferences are to occur.

2.4.3. Significance of Project

Panic Disorder is a serious, chronic condition with a high degree of impairment on the patient's life. Since many of the symptoms of Panic Disorder resemble those of clinical diseases, Panic Disorder patients frequently use medical services and undergo costly diagnostic procedures to rule out conditions such as heart disease (Katon, 1996). This places not only a financial burden on the medical system, but also comprises social and functional impairment of the affected patient. For example, a recent review study demonstrated that 30.1% of patients complaining of chest pain who arrived at the emergency room (ER) were diagnosed with a Panic Disorder, of which 22.4% exhibited Panic Disorder with no coronary artery disease (CAD). On the other hand, Panic Disorder has been identified as an independent risk factor for subsequent acute myocardial infarction and is associated with coronary artery disease (Walters et al., 2008). More research is needed to find out about the mechanisms responsible for physical anxiety to decrease the hazard of coronary heart disease in those that are diagnosed with Panic Disorder, and to reduce unnecessary medical visits.

The studies provided a thorough and well-controlled examination of attentional biases and emotional word processing in controls, participants with high trait anxiety, and in Panic Disorder patients. The subliminal processing distinction is emerging as a critical variable in the affective processing literature (e.g. Pauli et al., 2005), but is not being utilized frequently yet in this group of participants, and therefore deserved investigation. A more specific aim of the project was studying hypervigilance to threat stimuli in Panic Disorder patients. Identifying markers of cognitive distortions in Panic Disorder has important implications both for vulnerability to onset and to recurrence of

acute episodes. By studying cognitive biases, we ultimately would like to help better current therapies for affective disorders, such as Cognitive-Behavioral Therapy (CBT). CBT effectively postpones relapse by addressing these cognitive biases and may be improved by a deeper understanding of the underlying mechanisms. Research on the neurophysiological correlates of cognitive bias in anxiety has only just begun and may help to target patients who would benefit from CBT.

2.5. Study 2 Tables

Table 2-1:   **Sociodemographic Characteristics of the Participants**

Characteristic	Patients (n=17)		Controls (n=14)	
	n	%	n	%
Male	1	5.9	2	14.3
Female	16§	94.1	12§	85.7
Use of medication for anxiety		35.3		0
Handedness (right)	17	100	14	100
	Mean	**SD**	**Mean**	**SD**
Age (years)	30.2	12.5	30.0	10.0
Education (years) [a]	14.3	2.9	16.5	3.0
ASI [**]	32.7	11.0	10.6	9.6
BDI [c][*]	14.3	11.1	5.3	4.7
STAI-S [b][**]	42.5	12.5	28.6	8.7
STAI-T[**]	49.3	14.5	32.3	9.1

[a] 4 participants were excluded due to incomplete questionnaires
[b] 2 participants were excluded due to incomplete questionnaires
[c] 3 participants were excluded due to incomplete questionnaires
§ Within group significance p<.01 (Chi-square test)
[*]   p<.05
[**]   p≤.001

Table 2-2: Perception thresholds of the Participants in the subliminal Stroop task.

Group	Patients (n=15)		Controls (n=13)	
Threshold used (ms)	n		n	
13	11		11	
26	3		2	
39	1		0	
	Mean	**SD**	**Mean**	**SD**
Mean perception threshold (ms)	17.33	8.0	15.0	4.8

Table 2-3: Average reaction time (RT) and Standard Error for each combination of Emotional Block and Word Type in the patient and control group in the supraliminal Stroop task.

		Means in ms (SE)	
Emotional Block	**Word type**	**Patients**	**Controls**
Threat	Threat	760.0 (21.2)	770.1 (21.2)
	Neutral	760.8 (18.3)	776.5 (21.5)
	Threat minus Neutral trial	-8.0 ms	-6.4 ms
Positive	Positive	739.4 (15.5)	762.5 (18.2)
	Neutral	747.0 (17.0)	760.3 (20.2)
	Positive minus Neutral trial	-7.6 ms	2.2 ms
Threat minus Positive Block	(Threat plus Neutral) minus (Positive plus Neutral)	17.2 ms	11.9 ms

Table 2-4: Average reaction time (RT) and Standard Error for each combination of Emotional Block and Word Type in the patient and control group in the subliminal Stroop task.

		Means in ms (SE)	
Emotional Block	**Word type**	**Patients**	**Controls**
Threat	Threat	742.1 (18.0)	768.0 (29.4)
	Neutral	724.0 (17.8)	769.2 (26.7)
	Threat minus Neutral trial	18.1 ms	-1.0 ms
Positive	Positive	718.7 (17.4)	750.8 (28.6)
	Neutral	720.8 (16.5)	749.0 (31.5)
	Positive minus Neutral trial	-2.1 ms	1.8 ms
Threat minus Positive Block	(Threat plus Neutral) minus (Positive plus Neutral)	13.31 ms	18.8 ms

Table 2-5: Percentages of correctly recognized words and false alarms as a function of Group and Word Type in the subliminal Stroop task.

Presence of stimulus	**Word type**	**Patients**	**Controls**
Hits	Threat	2.8%	1.4%
	Neutral	0.5%	0.4%
	Positive	1.3%	1.8%
	Overall	0.4%	0.3%
False Alarms	Threat	1.8%	0.9%
	Neutral	0.5%	0.2%
	Positive	0.6%	0.6%
	Overall	0.3%	0.2%

2.6. Study 2 Figures
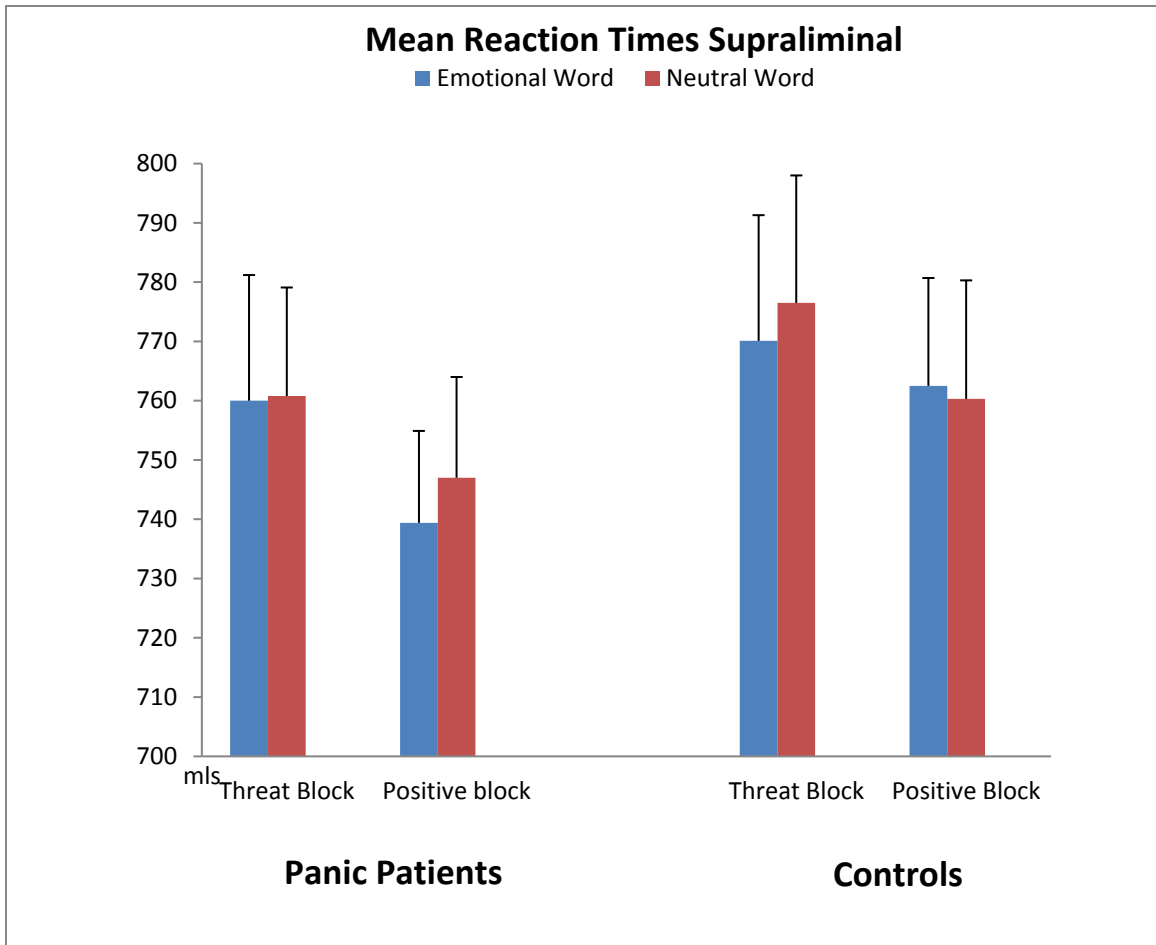
Figure 2-1: **Mean RT as a function of Group, Emotional Block, and Word Type in the supraliminal Stroop task.**

Mean Reaction Times Subliminal
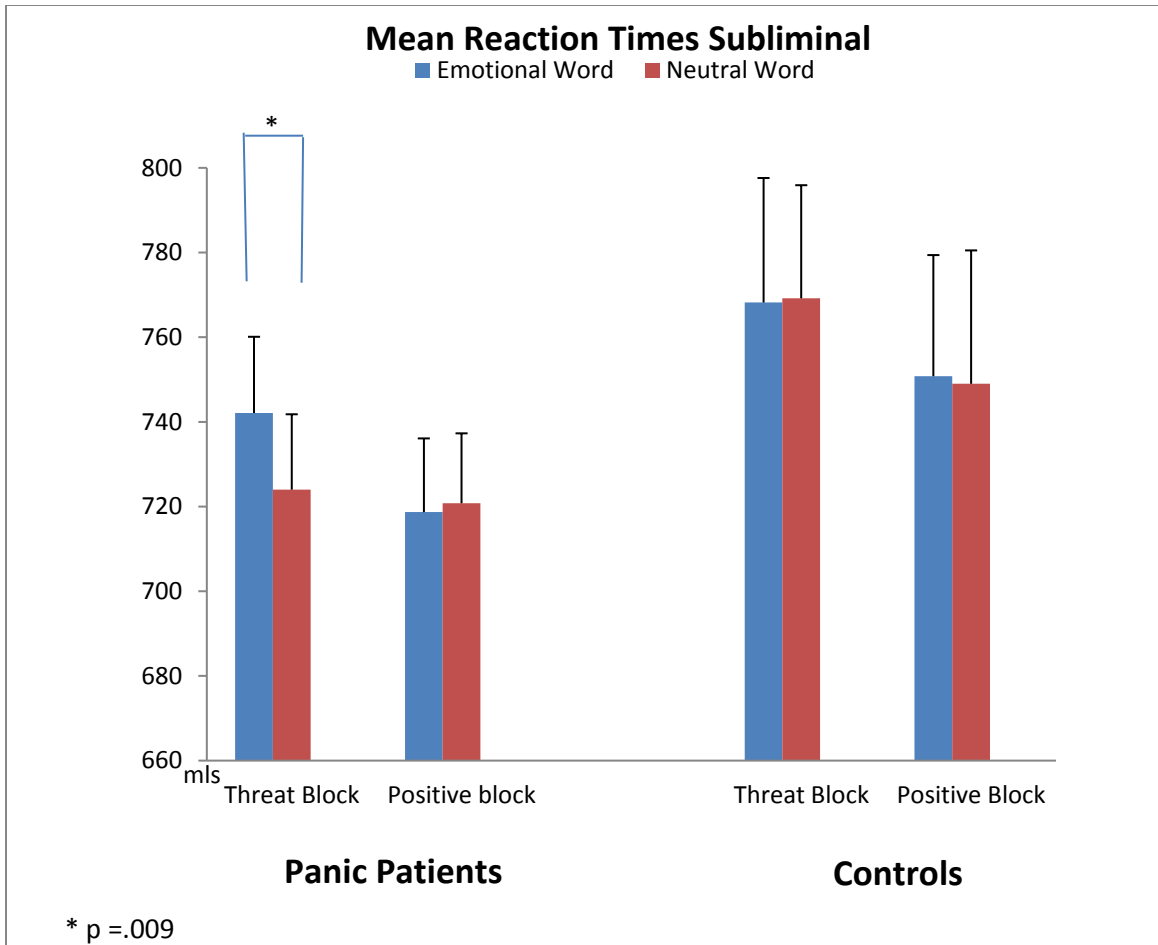
Figure 2-2:   **Mean RT as a function of Group, Emotional Block, and Word Type in the subliminal Stroop task.**
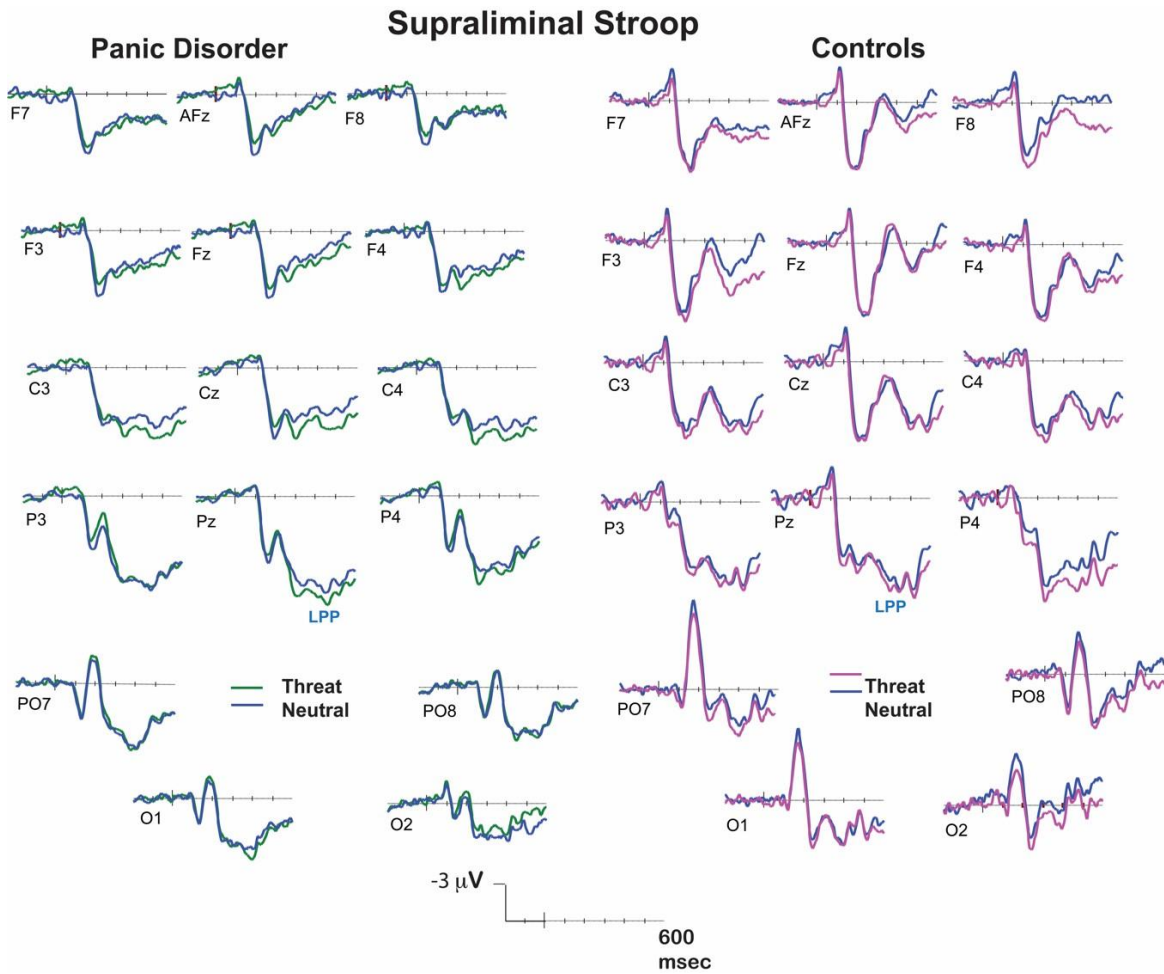
Figure 2-3: **Grandaverage waveforms from the Supraliminal Stroop task for the Threat Blocks at 16 representative scalp sites over frontal, central, parietal, and occipital scalp for the Panic Patient Group and the Control Group.**
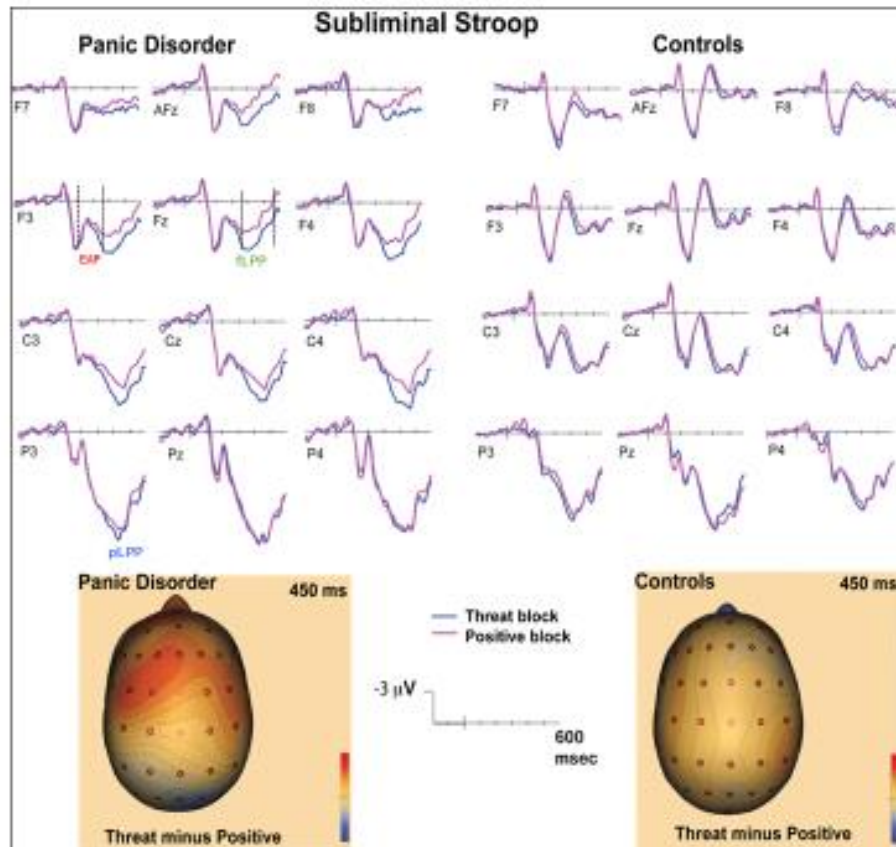
Figure 2-4: **Grandaverage waveforms from the Subliminal Stroop task for the Threat Blocks and Positive Blocks at 12 representative scalp sites over frontal, central, parietal, and occipital scalp for the Panic Patient Group and the Control Group.**

Chapter 3.   General Conclusion

The major goal of this project was to shed light on the question whether the behavioural and brain electrophysiological correlates of threat processing are similar or different in *trait* anxiety and clinical disease *state,* and whether both groups were characterized by hypervigilance to threat exemplified in an *early* attentional bias in both the supraliminal and subliminal task at a behavioral and brain electrophysiology level. A summary of results of Study 1 and Study 2 can be found in Table 3-1.

We paid particular attention to early frontal ERP effects. With the knowledge that early threat detection in the amygdala cannot produce a signal detectable from the scalp – a deep, small and round structure does not have the proper geometry to produce coherent summed electrical fields propagated to the scalp surface- we followed others in the emotion ERP field and interpreted the EAP as reflecting activity of an early cortical salience detection system (in this case responding to threat) through either subcortical (amygdala or thalamus) projections to medial PFC, or fast feedforward cortico-cortical projections from visual cortex to prefrontal cortex (Pourtois et al., 2013). In consideration of the recent construct of automatic emotion regulation in cognitive and social neuroscience (i.e., Braunstein, Gross, & Ochsner, 2017; Ochsner & Gross, 2008), the knowledge of bidirectional projections between ventromedial PFC and amygdala also providing the neural circuitry for fear extinction across species, and the fMRI data showing inverse sign changes in activation in amygdala and ventromedial PFC, we now extend the role of the EAP to include not only early detection of threat information, but also the concomitant *automatic* down-regulation of amygdala activity in response to negative self-relevant stimuli in order to prevent a generalized hyperarousal with a full-fledged emotional response (including physiological arousal and a subjective affective change).

EAP in high trait anxiety

Consistent with our predictions, we replicated our previous finding of significantly greater amplitude early frontal positivity (EAP) to threat words in the supraliminal eStroop task selectively for individuals with high Anxiety Sensitivity. Central to this project was the prediction that the high AS group would exhibit increased and even earlier frontal modulations when emotional words were presented below perceptual threshold in the subliminal eStroop task.

In fact, our hypothesis was upheld. Threat-containing blocks in the high AS group elicited significantly more positive electrical activity over anterior frontal scalp than positive blocks in the EAP time window as well as in a preceding time window (the P150). It is worth noting that participants did not show evidence of differential recognition of threat words, which may perhaps explain why there was no EAP differentiation of threat and neutral words within the threat blocks.

We take these combined early frontal positivity effects as evidence that in high trait anxiety they may index a *hyperactive* or hypersensitive early threat detection system located in medial PFC or insular cortex, which resting state fMRI has revealed to be main hubs of the salience cortical network (Menon, 2011; Seeley et al., 2007). Confirmatory evidence comes from fMRI studies on individuals with dispositional anxiety, who show *increased* activation in ventromedial PFC in an eStroop task in response to negative words. Such effect was interpreted by the authors as a compensatory mechanism, reflecting an attempt to downregulate ongoing emotional experience generated by the amygdala (Engels et al., 2007). Importantly, a similar increased activation in vMPFC or adjacent rostral Anterior Cingulate Cortex has been reported in patients with less severe symptoms, e.g. in GAD patients (Etkin et al., 2009), or depression patients with mild symptoms or in clinical remission (Liotti & Mayberg, 2001). In fact, *increased* rostral ACC activity appeared to predict treatment response in acute depression, while *decreased* rostral ACC/vMPFC activity predicted treatment non-response (Mayberg et al., 1997). There seems to be an inverse correlation between symptom severity and functional impairment of mPFC (Ball et al., 2013).

Behaviour, EAP and LPPs in Panic Disorder

Based on our previous work on behavioural and electrophysiological correlates of attentional bias to physical threat in high Anxiety Sensitivity and knowing that high AS is a recognized susceptibility factor for Panic Disorder, we predicted that the same EAP abnormalities as observed in the former would be also present in patients with the disease state. Contrary to our expectations, we found a different set of results among Panic Disorder patients.

While the impact of threat words (the attentional bias to threat) was evident in this group both behaviourally and in terms of brain electrical responses, the results were in fact quite different from those in the healthy group with high trait physical anxiety. In

Panic Disorder patients, the most sensitive task to elicit behavioural and ERP differences was the subliminal task. Behaviourally, Panic Disorder patients were slower to respond to subliminal threat relative to neutral words in the threat blocks, an effect not seen in high AS individuals. More importantly, there was <u>no</u> evidence of early differential frontal ERP modulation to threat words (EAP and P150) in either the supraliminal or subliminal eStroop task. In contrast to the lack of noticeable EAP effects in the subliminal task, there were significant sustained positivity modulations to threat blocks at a later time epoch (frontal and parietal LPP). LPPs and related P3s are typically associated with effortful, elaborative and conscious processing of emotional material and cognitive decision making (e.g. Hajcak, 2010). Their generators are broadly distributed over cortical association regions of dorsolateral PFC, parietal and temporal cortex, which are known substrates of attention and memory processes (perhaps active during cognitive brooding over conscious concern-specific thoughts or situations). The presence of LPP effects was particularly surprising while processing subliminal threat, and it may be related to the observed behavioural differences. It may suggest that cortical networks devoted to cognitive processes may indeed be occupied in Panic Disorder patients by automatic threat related associations.

We postulate here that in Panic Disorder patients, threat information triggering a hyperresponsive amygdala (not measurable by EEG) would fail to cause sufficient top-down emotion regulating activity from vmPFC, resulting in the absence of a sizeable early EAP. Nonetheless, the emotional arousal generated in the amygdala would cause later effects (starting around 350 ms) through widespread connections to multiple cortical targets resulting in the frontal and parietal LPP effects. In other words, the lack of an EAP in our patient sample in both tasks might reflect a decreased ability to recruit and sufficiently engage mPFC for salience detection and automatic emotion regulation in the disease state. That is, in patients, an amplified bottom-up threat detection system, most likely involving the amygdala, might have exhausted the top-down control system over time, making it unable to exert appropriate control, as it would make the detection of salience more difficult (Ball et al., 2013). In support of these notions, several studies have confirmed an over- responsiveness of limbic circuitry in anxiety disorders (e.g. Etkin & Wagner, 2007), and hypofrontality in Panic Disorder patients (Ohta et al., 2008; Ball, Ramsawh, Campbell-Sills, Paulus & Stein, 2013; Reinecke et al., 2015). Our study cannot directly measure responsiveness of deep midline structures involved in emotion

generation, as ERPs do not reflect activity of the amygdala, hypothalamus, septal nuclei, etc., and therefore we can only measure activity in *cortical* target areas of limbic-cortical projections, which we believe is being reflected in EAP effects.

In summary, the inverse correlation between symptom severity and prefrontal activation supports the hypothesis that PFC hypoactivation could be a component of the transition of dispositionally high anxiety to disordered anxiety (Ball et al., 2013). We propose that presence or absence of the EAP could provide a marker able to quantify such transition. A hyperactive mPFC (and its putative early ERP correlate, the EAP) would be a marker of vulnerability to Panic Disorder (a trait-marker) and early attempts at compensating hyperactive limbic circuitry in the early stages of disease. At later and more severe stages of the disorder, hyperactivity would turn into vMPFC hypoactivity, as shown in several fMRI studies in Panic Disorder (Ohta et al., 2008; Ball, Ramsawh, Campbell-Sills, Paulus & Stein, 2013; Reinecke et al., 2015), and PTSD (Ehlert, Wager, Heinrichs & Heim, 2009).

Remaining questions, recommendations and future studies

The question remains how (and which) high trait anxious individuals transition to the disease state, especially since AS is a core risk factor in the development of Panic Disorder. Actual Panic Disorder symptomatology onset may depend, among other things, on how individuals regulate their emotion (Cisler, Olatunji, Feldner, & Forsyth, 2009). Is a hypoactive PFC a precursor of this transition, or is it linked to the expression of active symptoms? Does vMPFC hypofunction gets worse over time as a function of symptom severity, phase of the illness, or age? Could vmPFC hyper- or hypofunction (and related EAP) provide trait markers of treatment response? For example, Panic Disorder patients who can respond to psychotherapy (CBT or exposure therapy) could have vmPFC and EAP hyperactivity, while those who will require drugs (such as SSRIs) could have vmPFC and EAP hypoactivity. Furthermore, can vmPFC hypofunction (and related reduced EAP) represent a state marker? Would it reduce or normalize after successful treatment of Panic Disorder symptoms with psychotherapy or drugs? In one study, brain activation in cortico-limbic structures of Panic Disorder patients changed following successful treatment with cognitive-behavioural therapy (Grambal, Hlustik, & Prasko, 2015).

Future studies could address these questions by employing larger samples of Panic Disorder patients and using longitudinal designs with multiple sessions over different phases of the disease, or before and after treatment. Answers to these questions will hopefully lead to innovations in more individualized and advanced therapy options for this common and debilitating anxiety disorder.

3.2. Chapter 3 Table

Table 3-1: **Summary of results of Study 1 and Study 2 for Reaction Times and ERP amplitude as a function of task, participant group, time window, and type of effect.**

	Reaction Times	130-200 ms P150	200-320 ms EAP	350-500 ms AN450	350-550 ms Parietal LPP	350-550 ms Frontal LPP
Supraliminal Stroop						
High AS Group			EAP	AN450	Parietal LPP	
Low AS Group					Parietal LPP	
Panic Disorder Patients					Parietal LPP	
Controls Study 2						
Subliminal Stroop						
High AS Group		P150	EAP			Frontal LPP
Low AS Group						
Panic Disorder Patients	Trial effect				Parietal LPP	Frontal LPP
Controls Study 2						
Trial effect :	Specific increase of response time to threat relative to neutral within block.					
Block effect:	Increase of amplitude to threat blocks relative to positive blocks.					
Trial effect:	Specific increase of amplitude to threat relative to neutral within block.					
Block effect:	Decrease of amplitude to threat blocks relative to positive blocks.					

References

Anderer, P., Saletu, B., Semlitsch, H. V., & Pascual-Marqui, R. D. (2002). Perceptual and Cognitive Event-Related Potentials in Neuropsychopharmacology: Methodological aspects and Clinical Applications (Pharmaco-ERP Topography and Tomography). *Methods Find Exp Clin Pharmacol, 24*(*Suppl C),* 121-137.

Asmaro, D., Jaspers-Fayer, F., Sramko, V., Taake, I., Carolan, P., & Liotti, M. (2012). Spatiotemporal dynamics of the hedonic processing of chocolate images in individuals with and without trait chocolate craving. *Appetite, 58*(3), 790-799.

Asmundson, G.J.G., Sandler, L.S., Wilson, K.G., & Walker, J.R., (1992). Selective attention towards physical threat in patients with Panic Disorder. *Journal of Anxiety Disorders 6*, 295–303.

Ball, T. M., Ramsawh, H. J., Campbell-Sills, L., Paulus, M.P., & Stein, M. B. (2013). Prefrontal dysfunction during emotion regulation in generalized anxiety and Panic Disorders. *Psychological Medicine*, *43*, 1475-1486.

Bar-Haim, Y., Lamy, D., Pergamin, L., Bakermans-Kranenburg, M. J., & van Ijzendoorn, M. H. (2007). Threat-related attentional bias in anxious and nonanxious individuals: A meta-analytic study. *Psychological Bulletin, 133*, 1-24.

Barlow, D. H. (1985). Dimensions of anxiety disorders. In A. H. Tuma & J. D. Maser (Eds.). *Anxiety and the anxiety disorders,* 479-500. Hillsdale, NJ: Erlbaum.

Beaver, J. D., Mogg, K., & Bradley, B. P. (2005). Emotional conditioning to masked stimuli and modulation of visuospatial attention. *Emotion, 5*, 67-79.

Beck, A.T., & Clark, D.A. (1997). An information processing model of anxiety: automatic and strategic processes. *Behaviour Research and Therapy, 35*, 49–58.

Beck, A. T., Steer, R. A., & Carbin, M. G. (1988). Psychometric properties of the Beck Depression Inventory: Twenty-five years of evaluation. *Clinical Psychology Review*, *8*, 77-100.

Becker, E. S., Rinck, M., Margraf, J., & Roth, W. T. (2001). The emotional Stroop effect in anxiety disorders: General emotionality or disorder specificity? *Journal of Anxiety Disorders*, *15*(3), 147-159.

Berchicci, M., Spinelli, D., & DiRusso, F. (2016). New insight into old waves. Matching stimulus and response-locked ERPs on the same time-window. *Biological Psychology*, *117*, 202-215.

Berggren, N., & Derakshan, N. (2013). The role of consciousness in attentional control differences in trait anxiety. *Cognition and Emotion*, *27*(5), 923-931.

Blanchette, I. (2006). Snakes, spiders, guns, and syringes: How specific are evolutionary constraints on the detection of threatening stimuli? *Q. J. Exp. Psychol. (Colchester), 59*, 1484-1504.

Bouton, M.E., Mineka, S., & Barlow, D.H. (2001). A modern learning theory perspective on the etiology of Panic Disorder. *Psychological Review, 108*, 4–32.

Bradley, B. P., Mogg, K., Milar, N., Bonham-Carter, C., Fergusson, E., Jenkins, J. & Parr, M. (1997). Attentional biases for emotional faces. *Cognition and Emotion, 11*, (1), 25-42.

Braunstein, L. M., Gross, J. J., & Ochsner, K. N. (2017). Explicit and implicit emotion regulation: a multi-level framework. *Social Cognitive and Affective Neuroscience*, *12*(10), 1545-1557.

Brinkmann, L., Buff, C., Feldker, K., Tupak, S. V., Becker, M. P. I., Herrmann, M.J., & Straube, T. (2017). Distinct phasic and sustained brain responses and connectivity of amygdala and bed nucleus of the stria terminalis during threat anticipation in Panic Disorder. *Psychological Medicine*, *47*, 2675-2688.

Brooks, S. J., Savov, V., Allzen, E., Benedict, C., Fredriksson, R., & Schioth, H. B. (2012). Exposure to subliminal arousing stimuli induces robust activation in the amygdala, hippocampus, anterior cingulate, insular cortex, and primary visual cortex: A systematic meta-analysis of fMRI studies. *NeuroImage, 59*, 2962-2973.

Brown, T.A.,DiNardo, P., & Barlow, D. H. (1994). *Anxiety Disorders Interview Schedule Lifetime Version (ADIS-IV-L).* ISBN: 9780195186734

Bryant, R. A., Kemp, A. H., Felmingham, K.L., Liddell, B., Olivieri, G., Peduto, A., Gordon, E., & Williams, L. M. (2008). Enhanced amygdala and medial prefrontal activation during nonconscious processing of fear in posttraumatic stress disorder. An fMRI study. *Human Brain Mapping, 29*, 517-523.

Carlson, J. M., Beacher, F., Reinke, K. S., Habib, R., Harmon-Jones, E., Mujica-Parodi, L. R., & Hajcak, G. (2012). Nonconscious attention bias to threat is correlated with anterior cingulate cortex gray matter volume: A voxel-based morphometry result and replication. *NeuroImage, 59*, 1713-1718.

Carlson J. M., & Reinke, K. S. (2008). Masked fearful faces modulate the orienting of covert spatial attention. *Emotion, 8,* 522-529.

Carolan, P. L., Jaspers-Fayer, F., Asmaro, D. T., Douglas, K. S. & Liotti, M. (2014). Electrophysiology of blunted emotional bias in psychopathic personality. *Psychophysiology, 51,* 36-41.

Carretie, L. (2014). Exogenous automatic attention to emotional stimuli: A review. *Cognitive Affective Behavioural Neuroscience, 14*, 1228-1258.

Carter, C. S., Maddock, R. J., & Magliozzi, J. (1992). Patterns of Abnormal Processing of Emotional Information in Panic Disorder and Major Depression. *Psychopathology, 25*, 65-70.

Ciscler, J. M., Olatuniji, B. O., Feldner, M. T., & Forsyth, J. P. (2009). Emotion regulation and the anxiety disorders: An integrative overview. *J Psychopathol Behav Assess, 32*, 68-82.

Compton, R. J. (2003). The interface between emotion and attention: a review of evidence from psychology and neuroscience. *Behavioral and cognitive neuroscience reviews, 2*(2), 115-129.

Deacon, B. & Abramowitz, J., 2007. Anxiety sensitivity and its dimensions across the anxiety disorders. *Anxiety Disorders, 20*, 837 – 857.

Demenescu, L. R., Renken, R., Kortekaas, R., van Tol, M. J., Marsman, J. B., van Buchem, M. A., van der Wee, M. J., Veltman, D. J., den Boer, J. A., & Aleman, A. (2001). Neural correlates of perception of emotional facial expressions in out-patients with mild - to - moderate depression and anxiety. A multicentre fMRI study. *Psychological Medicine, 41*, 2253-2264.

Dillon, D. G., Cooper, J.J., Grent-t-Jong, T., Woldorff, M. G. & LaBar, K. S. (2006). Dissociation of event-related potentials indexing arousal and semantic cohesion during emotional word encoding. *Brain and Cognition, 62*, 43-57.

Dinardo, P.A., Moras, K., & Barlow, D. H. (1993). Reliability of DSM-III-R Anxiety Disorder Categories – using the anxiety disorders interview schedule revised (ADIS-R). *Archives of General Psychiatry, 50*(4), 251-256.

Dresler, T., Attar, C. H., Spitzer, C., Loewe, B., Deckert, J., Buechel, C., Ehlis, A. C., & Fallgatter, A. J. (2012). Neural correlates of the Emotional Stroop task in Panic Disorder patients: An event-related fMRI study. J*ournal of Psychiatric Research, 46*, 1627-1634.

Dresler, T., Ehlis, A. C., Attar, Ernst, L. H., Tupak, S. V., Hahn, T., Warrings, B., Markulin, F., Spitzer, C., Loewe, B., Deckert, J. & Fallgatter, A. J. (2012). Reliability of the Emotional Stroop task: An investigation of patients with Panic Disorder. *Journal of Psychiatric Research, 46*, 1243-1248.

Dresler T., Guhn, A., Tupak, S. V., Ehlis, A.-C., Herrmann, M.J., Fallgatter, A. J., Deckert, J., & Domschke, K. (2013). Revise the revised? New dimensions of the neuroanatomical hypothesis of Panic Disorder. *J Neural Transm, 120*, 3-29.

Eimer, M., & Holmes, A. (2002). An ERP study of the time course of emotional face processing. *NeuroReport, 13*(4), 427-431.

Eimer, M., Kiss, M., & Holmes, A. (2008). Links between rapid ERP responses to fearful faces and conscious awareness. *Journal of Neuropsychology, 2*, 165-181.

Ehlers, A., Margraf, J., Davies, S., & Roth, W. T. (1988). Selective processing of threat cues in subjects with panic attacks. *Cognition and Emotion, 2*(3), 201-219.

Ehlert, U., Wagner, D., Heinrichs, M. & Heim, C. (1999). Psychobiological aspects of posttraumatic stress disorder. *Nervenarzt, 70*(9), 773-779.

Engels, A. S., Heller, W., Mohanty, A., Herrington, J. D., Banich, M. T., Webb, A. G., & Miller, G.A. (2007). Specificity of regional brain activity in anxiety types during emotion processing. *Psychophysiology, 44*, 352-363.

Etkin, A., Klemenhagen, K. C., Dudman, J. T., Rogan, M. T., Hen, R., Kandel, E. R., & Hirsch, J. (2004). Individual differences in trait anxiety predict the response of the basolateral amygdala to unconsciously processed fearful faces. *Neuron, 44*, 1043-1055.

Etkin, A., Prater, K. E., Schatzberg, A. F., Menon, V., & Greicius, M. D. (2009). Disrupted amygdala subregion functional connectivity and evidence of a compensatory network in generalized anxiety disorder*. Arch. Gen. Psychiatry, 66*, 1361-1372.

Etkin, A. & Schatzberg, A. F. (2011). Common abnormalities and disorder-specific compensation during implicit regulation of emotional processing in generalized anxiety and major depressive disorders. *American Journal* of Psychiatry, *168*, 968-978.

Etkin, A. & Wagner, T. D. (2007). Functional neuroimaging of anxiety: a meta-analysis of emotional processing in PTSD, social anxiety disorder, and specific phobia. *American Journal of Psychiatry, 164*, 1476-1488.

Eysenck, M. W., Derakshan, N., Santos, R., & Calvo, M. (2007). Anxiety and cognitive performance: The attentional control theory. *Emotion, 7*(2), 336-353.

Fava, L., & Morton, J. (2009). Causal models of Panic Disorder theories. *Clinical Psychology Review, 29*, 623-637.

Feldker, K., Heitmann, C. Y., Neumeister, P., Bruchmann, M., Vibrans, L., Zwitserlood, P., & Straube, T. (2016). Brain responses to disorder-related visual threat in Panic Disorder. *Human Brain Mapping, 37*, 4439-4453.

Feng, C., Becker, B., Huang, W., Wu, X., Eickhoff, S. B., & Chen, T. (2018). Neural substrates of the emotion-word and emotional counting Stroop tasks in healthy and clinical populations: A meta-analysis of functional brain imaging studies. *NeuroImage, 173*, 258-274.

Fischler, I. & Bradley, M. (2006). Event-related potential studies of language and emotion: Words, phrases, and task effects. *Progress in Brain Research, 156*, 185-203.

Foti, D., & Hajcak, G. (2008). Deconstructing reappraisal: Descriptions preceding arousing pictures modulate the subsequent neural response. *Journal of Cognitive Neuroscience, 20*, 977-988.

Frederikson, M., Wik, G., Annas, P., Ericson, K., & Stone-Elander, S. (1995). Functional neuroanatomy of visually elicited simple phobic fear: Additional data and theoretical analysis. *Psychophysiology*, *32*, 43-48.

Fox, E. (2002). Processing emotional facial expressions: The role of anxiety and awareness. *Cogn. Affect. Behav. Neuroscience, 2*, 52-63.

Geiger, M. J., Neufang, S., Stein, D. J., & Domschke, K. (2014). Arousal and the attentional network in Panic Disorder. *Human Psychopharmacology*, *29*, 599-603.

Gibbons, H. (2009). Evaluative priming from subliminal emotional words: Insights from event-related potentials and individual differences related to anxiety. *Consciousness and Cognition*, 18, 383-400.

Grambal, A., Hlustik, P., & Prasko, J. (2015). What fMRI can tell us about Panic Disorder: bridging the gap between neurobiology and psychotherapy. *Neuroendocrinology letters*, *36*(3), 214-225.

Hajcak, G., MacNamara, A., & Olvet, D. M. (2010). Event-related potentials, emotion, and emotion regulation: An integrative review. *Developmental Neuropsychology, 35*(2), 129-155.

Hanatani, T., Sumi, N., Taguchi, S., Fujimoto, O., Nan-Ho, H., & Takeda, M. (2005). Event-related potentials in Panic Disorder and generalized anxiety disorder. *Psychiatry Clin Neurosci, 59*, 83–88

Hansen, R. D. & Hansen, C. H. (1988). Repression of emotionally tagged memories: The architecture of less complex emotions. *Journal of Personality and Social Psychology, 55*(5), 811-818.

Hope, D. A., Rapee, R. M., Heimberg, R. G., & Dombeck, M. J. (1990). Representations of the self in social phobia: Vulnerability to social threat. *Cognitive Therapy and Research*, *14*(2), 177-189.

Howe, A. S., Pinto, A. & DeLuca, V. (2014). Meta-analysis of P300 waveform in Panic Disorder. *Exp. Brain Research, 232*, 3221-3232.

Hunt, C., Keogh, E., & French, C. C. (2006). Anxiety Sensitivity: The role of conscious awareness and selective attentional bias to threat. *Emotion*, *6*(3), 418-428.

Ito, T.A., Larsen, J. T., & Smith, N. K. (1998). Negative information weighs more heavily on the brain: The negativity bias in evaluative categorizations. *Journal of Personality and Social Psychology, 75*(4), 887-900.

Kawaski, H., Adolphs, R., Kaufman, O., Damasio, H., Damasio, A. R., Granner, M., Bakken, H., Hori, T., & Howard, M. A. (2001). Single neuron responses to emotional visual stimuli recorded in human ventral prefrontal cortex. *Nat. Neurosci, 4*, 15-16.

Katon, W. (1996). Panic Disorder: Relationship to high medical utilization, unexplained physical symptoms, and medical costs. *Journal of Clinical Psychiatry*, *57*(10), 11-22.

Keogh, E., Dillon, C., Georgiou, G., & Hunt, C. (2001). Selective attentional biases for physical threat in physical anxiety sensitivity. *Journal of Anxiety Disorders, 15*, 299-315.

Kober, H., Barrett, L. F., Joseph, J., Bliss-Moreau, E., Lindquist, K., & Wager, T. D., (2008). Functional grouping and cortical-subcortical interactions in emotion: A meta-analysis of neuroimaging studies. *NeuroImage, 42,* 998-1031.

Kucera, H., & Francis, N. (1967). Computational analysis of present-day American English index. *Brown University Press*, Providence, RI.

LeDoux, J. W. (1996). *The Emotional Brain: The Mysterious Underpinnings of Emotional Life.* New York: Simon and Schuster.

Li, W., Zinbarg, R. E., & Paller, K. A., (2007). Trait anxiety modulates supraliminal and subliminal threat: Brain potential evidence for early and late processing influences. *Cognitive, Affective, & Behavioural Neuroscience, 7*(1), 25-36.

Liddell, B. J., Brown, K. J., Kemp, A. H., Barton, M. J., Das, P., Peduto, A., Gordon, E., & Williams, L.M. (2005). A direct brainstem-amygdala-cortical alarm system for subliminal signals of threat. *NeuroImage, 24*, 235-243.

Lim, S.-L., & Kim, J.-H. (2005). Cognitive processing of emotional information in depression, panic, and somatoform disorder. *Journal of Abnormal Psychology, 114*(1), 50-61.

Liotti, M., Brannan, S., Egan, G., Shade, R., Madden, L., Abplanapl, Robillard, R., Lancaster, J., Zamarripa, F. E., Fox, P. T., & Denton, D. (2001). Brain responses associated with consciousness of breathlessness (air hunger). *PNAS, 98*(4), 2035-2040.

Liotti, M. & Mayberg, H. (2001). The role of functional neuroimaging in the neuropsychology of depression. *Journal of Clinical and Experimental Neuropsychology*, *23*(1), 121-136.

Lipka, J., Hoffmann, M., Miltner, W. H. R., & Straube, R. (2014). Effects of Cognitive-Behavioural Therapy on brain responses to subliminal and supraliminal threat and their functional significance in specific phobia. *Biological Psychiatry, 76*, 869-877.

Lundh, L. G., Wikstrom, J., Westerlund, J., & Ost, L. G. (1999). Preattentive bias for emotional information in Panic Disorder with Agoraphobia. *Journal of Abnormal Psychology, 108*, 222-232.

Macleod, C. & Hagan, R. (1992). Individual differences in the selective processing of threatening information, and emotional responses to a stressful life event. *Behav. Res. Ther.*, *30*(2), 153-161.

MacLeod, C., Mathews, A., & Tata, P. (1986). Attentional bias in emotional disorder. *Journal of Abnormal Psychology, 95*, 15-20.

Maidenberg, E., Chen, E., Craske, M., Bohn, P., & Bystritsky, A. (1996). Specificity of attentional bias in Panic Disorder and Social Phobia. *Journal of Anxiety Disorders, 10*(6), 529-541.

Maller, R. G., & Reiss, S. (1992). Anxiety Sensitivity in 1984 and Panic Attacks in 1987. *Journal of Anxiety Disorders*, *6*, 241-247.

Mayberg, H. S., Brannan, S. K., Mahurin, R. K., Jerabek, P.A., Brickman, J.S., Tekell, J. L., Silva, J. A., McGinnis, S., Glass, T.G., Martin, C. C., & Fox, T. P. (1997). Cingulate function in depression: A potential predictor of treatment response. *Neuroreport*, *8*(4), 1057-1061.

McNally, R.J., Amir, N., Louro, C.E., Lukach, B.M., Riemann, B.C., & Calamari, J.E. (1994). Cognitive processing of idiographic emotional information in Panic Disorder. *Behaviour Research and Therapy, 32*, 119–122.

McNally, R.J. (2002). Anxiety sensitivity and Panic Disorder. *Biological Psychiatry 52*(10), 938–946.

Meneguzzo, P., Tsakiris, M., Schioth, H. B., Stein, D. J., & Brooks, S. J. (2014). Subliminal versus supraliminal stimuli activate neural responses in anterior cingulate cortex, fusiform gyrus, and insula: A meta-analysis of fMRI studies. *BMC Psychology, 2*, 52.

Menon, V. (2011). Large scale brain networks and psychopathology: a unifying triple network model. *Trends in Cognitive Sciences*, *15*(10), 483-506.

Mogg, K., & Bradley, B. P. (1998). A cognitive-motivational view analysis of anxiety. *Behaviour Research and Therapy, 36*, 809-848.

Mogg, K., Bradley, B. P., Williams, R., & Mathews, A. (1993). Subliminal processing of emotional information in Anxiety and Depression. *Journal of Abnormal Psychology*, *102*(2), 304-311.

Mogg, K., Kentish, J., & Bradley, B. P. (1992). Effects of anxiety and awareness on colour-identification latencies for emotional words. *Behav. Res. Ther.*, *31*(6), 559-567.

Mogg, K., Mathews, G., & Eysenck, M. (1992). Attentional bias in clinical anxiety states. *Cognition and Emotion, 6*, 149-159.

Monk, C.S., Telzer, E. H., Mogg, K., Bradley, B. P., Mai, X., Louro, H. M., Chen, G., McClure-Tone, E. B., Ernst, M., & Pine, D. S. (2008). Amygdala and ventrolateral prefrontal cortex activation to masked angry faces in children and adolescents with generalized anxiety disorder. *Arch. Gen. Psychiatry, 65*, 568-576.

Mueller, E. M., Hofmann, S. G., Santesso, D. L., Meuret, A. E., Bitran, S. & Pizzagalli, D. A., (2009). Electrophysiological evidence of attentional biases in social anxiety disorder. *Psychological Medicine, 39*(7), 1141-1152.

Naragon-Gainey, K. (2010). Meta-Analysis of the Relations of Anxiety Sensitivity to the Depressive and Anxiety Disorders. *Psychological Bulletin, 136*(1), 128-150.

Naumann, E., Bartussek, D., Diedrich, O., & Laufer, M. (1992). Assessing cognitive and affective information processing functions of the brain by means of the late positive complex of the evet-related potential. *Journal of Psychophysiology, 6,* 285-298.

Northoff, G., Richter, A., Gessner, M., Schlagenhauf, F., Fell, J., Baumgart, F., Kaulisch, T., Koetter, R., Stephan, K. E., Leschinger, A., Hagner, T., Bargel, B., Witzel, T., Hinrichs, H., Bogerts, B., Scheich, H., & Heinze, H. J. (2000). Functional dissociation between and lateral prefrontal cortical spatiotemporal activation in negative and positive emotions: a combined fMRI/MEG study. *Cereb. Cortex, 10,* 93-107.

Ochsner, K. N., & Gross, J.J. (2008). Cognitive emotion regulation: Insights from social cognitive and affective neuroscience. *Current directions in Psychological Science, 17*(2), 153-158.

Oehmann, A., Flykt, A., & Esteves, F. (2001). Emotion drives attention: Detecting the snake in the grass. *Journal of Experimental Psychology: General, 130*, 466-478.

Ohta, H., Yamagata, B., Tomioka, H., Takahashi, T., Yano, M., Nakagome, K., & Mimura, M. (2008). Hypofrontality in Panic Disorder and Major Depressive Disorder assessed by multi-channel near – infrared spectroscopy. *Depression and Anxiety, 25,* 1053-1059.

Olatunji, B.O., & Wolitzky-Taylor, K. B. (2009). Anxiety Sensitivity and the Anxiety Disorders: A Meta-Analytic Review and Synthesis. *Psychological Bulletin, 135*(6), 974-999.

Ottaviani, C., Cevolani, D., Nucifora, V., Borlimi, R., Agati, R., Leonardi, M., De Plato, G., & Brighetti, G. (2012). Amygdala responses to masked and low spatial frequency fearful faces: A preliminary fMRI study in Panic Disorder. *Psychiatry Research: Neuroimaging, 203,* 159-165.

Pauli, P., Amrhein, C., Muehlberger, A., Dengler, W., & Wiedemann, G. (2005). Electrocortical evidence for an early abnormal processing of panic-related words in Panic Disorder patients. *International Journal of Psychophysiology*, *57*, 33-41.

Pauli, P., Dengler, W., Wiedemann, G., Montoya, P., Birbaumer, N., Flor, H.& Buchkremer, G. (1997). Behavioral and neurophysiological evidence for altered processing of anxiety-related words in Panic Disorder. *Journal of Abnormal Psychology, 106*, 213-220.

Perez-Benitez, C. I., Shea, M. T., Raffa, S., Rende, R., Dyck, I. R., Ramsawh, H. J., Edelen, M. O., & Keller, M. B. (2009). Anxiety Sensitivity as a Predictor of the Clinical Course of Panic Disorder: A 1-year follow-up study. *Depression and Anxiety, 26*, 335-342.

Pergamin-Hight, L., Naim, R., Bakersmans-Kranenburg, M. J., van Ijzendoorn, M. H., & Bar-Haim, Y. (2015). Content specificity of attention bias to threat in anxiety disorders: A meta-analysis. *Clinical Psychology Review*, *35*, 10-18.

Phaf, R. H., & Kan, K.-J., 2007. The automaticity of emotional Stroop: A meta-analysis. *Journal of Behavior Therapy and Experimental Psychiatry*, *38*, 184-199.

Phelps, E. A., O'Connor, K. J., Gatenby, J. C., Gore, J. C., Grillon, C., & Davis, M. (2001). Activation of the left amygdala to a cognitive representation of fear. *Nature Neuroscience*, *4*, 437-441.

Pillay, S. S., Gruber, S. A., Rogowska, J., Simpson, N., & Yurgelun-Todd, D. A. (2006). fMRI of fearful facial affect recognition in Panic Disorder: the cingulate gyrus – amygdala connection. *Journal of Affective Disorders*, *94*, 173-181.

Poletti, S., Radaelli, D., Cucchi, M., Ricci, L., Vai, B., Smeraldi, E., & Benedetti, F. (2015). Neural correlates of anxiety sensitivity in Panic Disorder: A functional magnetic resonance imaging study. *Psychiatry Research: Neuroimaging*, *233,* 95-101.

Pourtois, G., Schettino, A., & Vuilleumier, P. (2013). Brain mechanisms for emotional influences on perception and attention: What is magic and what is not. *Biological Psychology, 2013*, 92, 492-512.

Pourtois, G., Grandjean, D., Sander, D., & Vuilleumier, P. (2004). Electrophysiological correlates of rapid spatial orienting towards fearful faces. *Cereb. Cortex, 14*, 619-633.

Reinecke, A., Cooper, M., Favaron, E., Massey-Chase, R., & Harmer, C. (2011). Attentional bias in untreated Panic Disorder. *Psychiatry Research, 185*, 387-393.

Reinecke, A., Filippini, N., Berna, C., Western, D. G., Hanson, B., Cooper, M. J., Taggart, P., & Harmer, C. J. (2015). Effective emotion regulation strategies improve fMRI and ECG markers of psychopathology in Panic Disorder. Implications for psychological treatment action. *Translational Psychiatry*, 1-10.

Reiss, S., & McNally, R. J. (1985). Expectancy model of fear. In S. Reiss & R. R. Bootzin (Eds).*Theoretical issues in behavior therapy*. New York: Academic Press.

Reiss, S., Peterson, R. A., Gursky, D. M., & McNally, R. J. (1986). Anxiety sensitivity, anxiety frequency, and the prediction of fearfulness. *Behaviour Research and Therapy*, *24*, 1-8.

Sass, S. M., Heller, W., Stewart, J. L., Silton, R. L., Edgar, J. C., Fisher, J. E., & Miller, G. A. (2010). Time course of attentional bias in anxiety: Emotion and gender specificity. *Psychophysiology*, *47*, 247-259.

Schacht, A., & Sommer, W. (2009). Time course and task dependence of emotion effects in word processing. *Cognitive, Affective, & Behavioural Neuroscienc*e, *9*(1), 28-43.

Schmidt, N. B. & Lerew, D. R. (1997). The role of anxiety sensitivity in the pathogenesis of panic: Prospective evaluation of spontaneous panic attacks during acute stress. *Journal of Abnormal Psychology, 106*(3), 355-364.

Schupp, H. T., Cuthbert, B. N., Bradley, M. M., Hillman, C. H., Hamm, A. L. O. & Lang, P. J. (2004). Brain process in emotional perception: Motivated attention. *Cognition and Emotion*, *18*, 593-611.

Seeley, W. W., Menon, V., Schatzberg, A. F., Keller, J., Glover, G. H., Kenna, H., Reiss, A. L., & Greicius, M. D. (2007). Dissociable intrinsic connectivity networks for salience processing and executive control. *The Journal of Neuroscience, 27*(9), 2349-2356.

Silverman W.K., & Albano A.M. (1996). The anxiety disorders interview schedule for DSM-IV - Child and parent versions. San Antonio: Psychological Corporation.

Simpson, J. R., Oenguer, D., Akbudak, E., Conturo, T. E., Ollinger, J. M., Snyder, A. Z., Gusnard, D. A., & Raichle, M. E. (2000). The emotional modulation of cognitive processing: an fMRI study. *J. Cogn. Neuroscience*, *12*, 157-170.

Smith, N. K., Cacioppo, J. T., Larsen, J. T., & Chartrand, T. L. (2003). May I have your attention, please: Electrocortical response to positive and negative stimuli. *Neuropsychologia, 41*, 171-183

Spielberger, C. D., Gorsuch, R. L., & Lushene, R. E. (1983). STAI manual for the State-Trait Anxiety Inventory. *Consulting Psychologists Press*, California, Palo Alto.

Taake, I., Jaspers-Fayer, F., & Liotti, M. (2008). Early frontal responses elicited by physical threat words in an Emotional Stroop Task: Modulation by anxiety sensitivity. *Biological Psychology, 81*, 48-57.

Taylor, M. J., Batty, M., & Itier, R. J. (2004). The faces of development: A review of early face processing over childhood. *J. Cogn. Neuroscience, 16*, 1426-1442.

Teachman, B.A., Smith-Janik, S.B., & Saporito, J. (2007). Information processing biases and Panic Disorder: Relationships among cognitive and symptom measures. *Behaviour Research and Therapy, 45*, 1791–1811.

Thomas, K. M., Drevets, D. C., Dahl, R. E., Ryan, N. D., Birmaher, B., Eccard, C. H., Axelson, D., Whalen, P. J., & Casey, B. J. (2001). Amygdala response to fearful faces in anxious and depressed children. *Archives of General Psychiatry*, *58*, 1057-1063.

Thomas, S.J., Johnstone, S.J., & Gonsalvez, C.J. (2007). Event-related potentials during an emotional Stroop task. *International Journal of Psychophysiology, 63*, 221-231.

Thomas, S.J., Gonsalvez, C.J., & Johnstone, S.J. (2013). Neural time course of threat-related attentional bias and interference in panic and obsessive-compulsive disorders. *Biol Psychol 94*, 116–129.

Thomas, S.J., Gonsalvez, C.J., & Johnstone, S.J. (2014). How specific are inhibitory deficits to obsessive-compulsive disorder? A neurophysiological comparison with Panic Disorder. *Clin Neurophysiol, 125*, 463–475.

Van den Heuvel, O. A., Veltman, D. J., Groenewegen, H. J., Witter, M. P., Merkelbach, J., Cath, D. C., van Balkom, H. J. L. M., van Oppen, P., & van Dyck, R. (2005). Disorder-specific neuroanatomical correlates of attentional bias in Obsessive-compulsive disorder, Panic Disorder, and hypochondriasis. *Arch Gen Psychiatry*, *62*, 922-933.

Van den Hout, M., Tenney, N., Huygens, K., Merckelbach, H., & Kindt, M. (1995). Responding to subliminal threat cues is related to trait anxiety and emotional vulnerability: A successful replication of Macleod and Hagan (1992). *Behav. Res. Ther.*, *33*(4), 451-454.

Van Hoff, J. C., Dietz, K. C., Sharma, D. & Bowman, H. (2008). Neural correlates of intrusion of emotion words in a modified Stroop task. *International Journal of Psychophysiology*, *67*(1), 23-34.

Van Laar, M.W., Volkerts, E. R., Verbaten, M. N., Trooster, S., van Megen, H. J., & Kenemans, J. L. (2002). Differential effects of amitriptyline, nefazodone, and paroxetine on performance and brain indices of visual selective attention and working memory. *Psychopharmacology, 162,* 351-363.

Walters, K., Rait, G., Petersen, I., Williams, R., & Nazareth, I. (2008). Panic Disorder and risk of onset coronary heart disease, acute myocardial infarction, and cardiac mortality: Cohort study using the general practice research database. *European Heart Journal, 29,* 2981-298.

West, R., & Alain, C. (1999). Event-related neural activity associated with the Stroop task. *Cognitive Brain Research*, *8*(2), 157-164.

West, R. (2003). Neural correlates of cognitive control and conflict detection in the Stroop and digit-location tasks. *Neuropsychologia*, *41*(8), 1122-1135.

Williams, J. M. G., Mathews, A., & MacLeod, C. (1996). The emotional Stroop task and psychopathology. *Psychological Bulletin, 120*(1), 3-24.

Williams, J. M. G., Watts, F. N., MacLeod, C., & Matthews, A. (1997). *Cognitive Psychology and Emotional Disorders.* John Wiley, Chichester.

Williams, L. M. & Gordon, E. (2007a). Dynamic organization of the emotional brain: Responsivity, stability, and instability. *Neuroscientist, 13*, 349 – 372.

Williams, L. M., Palmer, D., Liddell, B. J., Song, L., & Gordon. E. (2006). The when and where or perceiving signals of threat versus non-threat. *NeuroImage, 31*, 458-467.

Williams, L. M., Kemp, A., Felmingham, K., Liddell, B., Palmer, D., & Bryant, R. A. (2007b). Neural biases to covert and overt signals of fear: Dissociation by trait anxiety and depression. *Journal of Cognitive Neuroscience*, *19*(10), 1595 – 1608.

Windmann, S., Sakhavat, Z., & Kutas, M. (2002). Electrophysiological evidence reveals affective evaluation deficits early in stimulus processing in patients with Panic Disorder. *J. Abnorm. Psychology, 111*,357-369.

Zhou, L.N., Wang, G.H. & Wang, H. L. (2018). Abnormalities of P300 before and after antidepressant treatment in depression: an ERP-sLORETA study. *Neuroreport*, *29*(3), 160-168.

Appendix A.

Word stimuli used in the supraliminal and subliminal Stroop tasks

Supraliminal Stroop	Threat	Positive	Neutral
1	Asthma	Humor	Helmet
2	Collapse	Fairness	Township
3	Stress	Energy	Season
4	Panic	Honey	Slope
5	Sweat	Glory	Clock
6	Anxiety	Comfort	Journal
7	Dizzy	Super	Tenth
8	Suffering	Desirable	Automatic
9	Seizure	Ecstasy	Cyclist
10	Stroke	Flower	String
11	Ulcer	Shine	Olive
12	Worry	Smile	Grass
Subliminal Stroop	**Threat**	**Positive**	**Neutral**
1	Tumor	Fortune	Feather
2	Bleed	Happy	Brochure
3	Toxic	Laugh	Washer
4	Fever	Fantastic	Record
5	Decay	Success	Fountain
6	Virus	Positive	Garage
7	Choke	Elated	Happy
8	Shaky	Pleased	Tower
9	Fatal	Content	Shelf
10	Cancer	Peaceful	Graph
11	Paralysis	Victory	Brick
12	Disease	Reward	Label

Appendix B.

Demographics and Medical Questionnaire

- What is your age? _____

- What is your gender? _____

- What is your major (if known)? _____

- Years of post-secondary education? _____

- GPA? _____

- Were you born in Canada? Yes / No
- If 'No' what is your country of origin? _____

- Is English your first language? Yes / No

- If 'No' for how many years have you spoken English fluently? _____

- What is your dominant hand (the hand that your write with)? Left / Right

- Are you wearing glasses or contacts? Yes / No

- Is your vision normal or corrected to normal if wearing glasses/contacts? Yes / No

- Are you color-blind? Yes / No

- Have you been diagnosed by a mental health professional and/or treated for any of the following:

- Depression Yes No

- Anxiety Yes No

- Attention-Deficit Disorder Yes No

- Thought Disorder Yes No

- Other (specify): _____

- Have you ever seen a neurologist or been to an emergency room for:

- Loss of motor or sensory function Yes No

- Loss of consciousness Yes No

- Head concussion Yes No

- Sleep disorder Yes No

- Migraines Yes No

- CT scan, MRI scan or Electroencephalogram Yes No

- Have you been told you have a learning disorder or disability, such as dyslexia (i.e. a reading disorder)? Yes / No

  ➢ If Yes, please explain:_____

- Do you have a serious medical condition? Yes / No

  ➢ If Yes, please explain:_____

- Are you currently taking any prescription medication? Yes / No

  ➢ If Yes, please explain:_____

- Do you use non-prescription drugs (optional) Yes/No

  ➢ If Yes, please explain
  (optional):_____

- Which/how many alcoholic beverages do you typically have in a week: _____

- How many hours do you typically sleep? _____

- How many hours did you sleep last night? _____

Appendix C.

Test Battery Measures: Descriptions

Anxiety Disorders Interview Schedule for DSM-IV

The ADIS-IV is a structured interview designed to assess for current episodes of anxiety disorders and to permit differential diagnoses between anxiety disorders, according to DSM-IV criteria (Brown, DiNardo & Barlow, 1994; Silverman & Albano, 1996). The ADIS-IV has demonstrated reliability and validity; for example, the interrater reliability for agoraphobia with Panic Disorder was estimated as .85 (Barlow, 1985). Using this kind of structural interview decreases the information variance and the interviewer variance. It is also a quantifiable assessment, because each diagnosed disorder is rated on a severity scale and can be used for comparisons between groups. The ADIS-IV also contains screening questions for psychotic and conversion symptoms and family psychiatric history. It also entails a very detailed section to determine the patient's medical and psychiatric treatment history.

The ADIS-IV was used to confirm the participants' diagnoses, and to ensure that the control participants were indeed free of any disorder. Current mood, somatoform and substance use disorders were also included because of their high rate of comorbidity with anxiety, and the symptoms of these disorders are often very similar to those of anxiety disorders.

Anxiety Sensitivity Index

The ASI was invented by Reiss et al. (1984), designed to measure the fear of behaviors or sensations associated with the experience of anxiety, and to fulfil the need for a psychometrically sound measure of anxiety sensitivity. It consists of 16 statements and takes less than 5 minutes to finish. Each item evaluates concerns about the possible negative consequences of anxiety symptoms. Participants labelled how accurate the statement described how they felt in general. Each item is rated on a 5-point Likert-type scale ranging from 0 (*not at all*) to 4 (*very much*). The ASI has demonstrated adequate internal consistency, and test re-test reliability was found to be .71 across three years (Reiss et al., 1994), confirming Reiss' and McNally's hypothesis that anxiety sensitivity is

a personality trait (Reiss & McNally, 1995), and that a high score on the ASI is a powerful and unique predictor who will have panic attacks in the future. For example, Reiss et al. (1992) found that participants with high ASI scores are five times more likely to have panic attacks within the next years compared to people with low ASI scores.

State-Trait Anxiety Inventory

The STAI was invented by Spielberger et al. (1983), designed to measure state and trait anxiety. It consists of two sets of 20 statements for both state and trait anxiety. Participants labelled how accurate the statements described how they felt at the moment or in general. It took less than 5 minutes to finish. Whereas the state anxiety questionnaire asks subjects to rate how they feel at the moment of testing, the trait anxiety scale asks subjects to rate how they feel in general. Internal reliability, test-retest reliability, and convergent and discriminant validity have been demonstrated for the state and trait scales in both younger and older adults (Spielberger et al., 1983).

Beck Depression Inventory

The BDI was invented by Beck et al. (1988), designed to measure symptoms of depression. It consists of 21 groups of statements describing feelings for the last two weeks. Participants picked one statement that best indicated their feelings from every group. The statements can be rated from 0 to 3 in terms of intensity. The BDI takes 5 to 10 minutes to finish, and the result is being calculated by adding up the score given to each of the 21 items. Cut-off scores for patients are the following: "None or minimal depression is < 10; mild to moderate depression is 10 to 18; moderate to severe depression is 19 to 29; and severe depression is 30 to 63" (Beck et al., 1988, p. 79). The BDI measures state depressive symptomology for the day it was completed by inquiring about second-order depression symptoms, reflecting negative attitudes, performance difficulties and somatic complaints. It has high internal consistency in psychiatric and non-psychiatric samples (mean estimate 0.87), and high concurrent validity. There is also evidence that BDI symptoms can discriminate medical patients, non-medical patients and healthy individuals, as well as psychiatric and non-psychiatric patients (Beck et al., 1988).