Atypical modulation of startle in women in face of aversive bodily sensations

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**Abstract**

Eye blink startle magnitude is assumed to be higher in threatening contexts. A scarce amount of studies suggest that this does not hold true when startle is measured during perceived threats to homeostatic integrity. The present study was set up to describe the startle response pattern to a selection of interoceptive stimuli. Female subjects (N = 36) were exposed once to 90 s of continued (1) cold pain, (2) inhalation of a gas mixture of 7.5% CO₂, and (3) breathing against an inspiratory and expiratory resistive load. Each stimulus was preceded and followed by a 90 second period of rest, respectively labeled baseline and recovery. Even after correcting eye blink startle responses for habituation, a decreased startle amplitude was evident during these stimuli. Results suggest that startle amplitude during aversive stimulation is inversely correlated with perceived fearfulness for women, although further studies are necessary to corroborate this interpretation.

1. Introduction

Interoception, the perception of the state of the body, serves to maintain homeostasis and is closely linked to the experience of emotions (Craig, 2002). Interoceptive fear is the apprehension of bodily sensations (Shear et al., 1997) and can manifest itself following the perception of non-painful interoceptive sensations and to excessive physical symptom reports.

Functional disorders, anxiety disorders, and pain related disorders, affect a significantly large part of the population. In all of these disorders interoceptive fears play a key role, implying that the advancement of both clinical and fundamental knowledge on interoceptive fear is of utmost importance. A body of literature as well as a number of laboratory studies imply that the etiology and maintenance of such disorders is due to associative learning processes (Acheson et al., 2007; Bouton et al., 2001; De Peuter et al., 2011; Mayer, 2000; Meulders et al., 2011; Pappens et al., 2013). Because of interoceptive fear conditioning, originally benign sensations can elicit fear responses, when in the past these benign sensations have preceded an aversive interoceptive sensation.

Although interoceptive fear conditioning has a strong pedigree in the understanding of the aforementioned disorders, relatively little research has elaborated on the basic fear response topography to interoceptive stimulations used in the laboratory. Therefore, the major aim of the current study was to document unconditioned fear responding to such interoceptive stimulations. We made a selection of stimuli frequently used in experimental paradigms on pain (e.g., Helsen et al., 2011) and dyspnea (Acheson et al., 2007; Pappens et al., 2011), namely cold pain, inhalation of CO₂-enriched air, and loaded breathing. We selected these particular stimuli because a limited body of literature on startle in response to these stimuli is already available, although as yet no design has presented these three stimuli in comparable manners within subjects. In this initial study, we limited ourselves to women: we justify this choice given that psychosomatic complaints and disorders
have a higher prevalence among women (Kroenke and Spitzer, 1998; Şar, 2010).

The potention – i.e. the relative increase in magnitude – of the eye blink component of startle is a well-validated and widely accepted measure of fear responding. An important question relating to the aim of the current study is whether the eye blink component of startle can provide a reliable indication of fear during aversive interoceptive stimulation. The startle reflex is modulated by the motivational system (Lang et al., 2000), and shows an increased amplitude when experiencing fear (Globisch et al., 1999; Hamm et al., 1997) or something which is otherwise unpleasant (Vrana et al., 1988). Affective modulation of the startle reflex magnitude results from activation of a variety of structures in which the amygdala plays a pivotal role. This modulatory effect of the motivational neurocircuitry on the eye blink motor reflex is described in more detail in the literature (e.g., Davis, 2006; Lang et al., 1998; Misslin, 2003).

Regarding photontion of startle following manipulations that induce fear or unpleasantness is a robust finding, it has predominantly been tested using visual and auditory stimuli. In contrast, the few studies on startle in response to aversive interoceptive stimulation present a more complicated and as yet inconclusive picture of findings.

With regard to thermal pain stimulation, findings are somewhat equivocal. For phasic heat pain, it appears that stimulation of short duration evokes startle potentiation (Crombez et al., 1997), whereas stimulation of a longer duration does not (Horn et al., 2012a, 2012b). For cold pain, there is an overall reduction when averaging startle amplitudes delivered at different times during a prolonged stimulation (Tavernor et al., 2006), whereas such reduction may not be evident at individual time points (De Peuter et al., 2009). Lovallo (1975) describes that pain in response to the cold pressor test (CPT) does not keep rising progressively as time of immersion increases, a finding which may explain why startle probes at particular time intervals are not reduced.

Regarding dyspnea, findings from several studies conducted in our research group strongly suggest that dyspnea induced by the inhalation of CO₂-enriched air is associated with an inhibition of the startle reflex (De Peuter et al., 2009; Pappens et al., 2012; Van Diest et al., 2009b).

Paradoxically, when dyspnea is induced by loaded breathing – a mechanical stimulus creating respiratory resistance – startle potentiation is evident when the stimulus is light (near perceptual threshold level), but absent when a respiratory load of higher (moderate) intensity is administered (Pappens et al., 2010). This is paradoxical, because self-report measures as well as skin conductance indicated that the higher load was more aversive and arousing than the light load.

Possible mechanisms for these findings have been suggested by their respective authors, and will be reviewed in the Discussion section. Regardless of the mechanism responsible for the apparently atypical startle pattern found in earlier studies documenting startle responding to the CPT, inhalation of CO₂-enriched air, and loaded breathing, it seems that startle within one type of stimulus is inversely correlated with unpleasantness (Pappens et al., 2010). The following parsimonious conclusions could be made: (a) these types of aversive interoceptive stimulation are associated with a reduction in startle rather than potentiation. (b) As dyspneic stimuli become more aversive as time progresses, it could be expected that startle responsivity decreases overall as the duration of dyspneic interoceptive stimulation increases. However, (c) startle in response to painful peripheral hypothermic stimulation may be an exception in that pain fluctuates over the course of time, and accordingly, startle may not necessarily decrease linearly over time.

To test these hypotheses, in the current study we subjected these earlier findings to a novel experimental paradigm, allowing for a within-subject comparison of unconditioned defensive responding to these three types of sustained, aversive interoceptive stimulation.

The primary aim of this study was to shed light on the startle response over time to three types of stimulation. Eye blink startle responses were studied during 90 s periods of cold pain, inhalation of CO₂-enriched air, and loaded breathing. In contrast to the studies of Pappens et al. (2010, 2011), which applied loads for only one inspiration, the continued stimulation allowed for testing our assumption that startle declines linearly during the course of dyspneic stimulation. Since we did not expect potentiation but rather a reduction in startle, it was important to make sure any reduction in startle wouldn’t be due to habituation. Therefore it was important to have a design which would allow us to statistically correct for habituation-bound decrease in startle. For this reason, startles were measured during a baseline phase prior to the stimulus phase, and during a recovery phase following the stimulus phase, so that a best fit line could be calculated which would filter out the effects of habituation. Another new element in the current experiment was that respiratory loads were applied both during inspiration and expiration, so that startle eliciting probes would always be administered during actual stimulation.

To test the general conclusions we made earlier, we respectively expected to observe:

(a) A reduced startle blink magnitude during aversive interoceptive stimulation, as compared to prior and following an aversive interoceptive stimulus. Given our design, this would correspond to a reduction of startle during stimulus phase as compared to baseline and recovery phase.

(b) For both dyspneic stimuli, we hypothesized a progressive reduction of the startle magnitude during the stimulus phase, as unpleasant dyspneic stimuli have been shown earlier to be associated with reduced startle responding, and as these stimuli are thought to become progressively more unpleasant as time since the onset increases.

(c) For the CPT, we expected a quadratic response pattern during the stimulus phase, given that the overall average of multiple startle responses is associated with a reduction in amplitude (Tavernor et al., 2000) while no such reduction has been evident during the 30 to 60 second period following stimulus onset (De Peuter et al., 2009), the latter which is perhaps due to the fluctuations in pain sensations during cold stimulation.

In line with earlier findings, it was expected that all stimuli would be scored as unpleasant rather than pleasant, that these stimuli would induce some self-perceived arousal as opposed to complete calm, leading to sub-maximal levels of feelings of dominance, and to induce some fear.

2. Materials and methods

2.1. Participants

Thirty-six female psychology freshmen (mean age: 19 y/old) participated in return for course credit. Exclusion criteria were pregnancy, presence or history of cardiovascular disease, pain-related conditions, or respiratory disease. Participants were randomly assigned to one of six orders of stimulus presentation — stimulus presentation orders were counterbalanced. The study protocol was approved by the Ethics Committee of the Department of Psychology in accordance with the Declaration of Helsinki (World Medical Association, 1997); prior to participation, all subjects read and signed an informed consent with information about the sensations that could possibly follow from exposure to the stimuli, a guarantee about anonymity, and that participation was voluntary and could be terminated at any point in time without loss of the promised course credit.

2.2. Stimuli and apparatuses

2.2.1. Cold pressor

The cold pressor test (CPT) was used as a cold pain (CP) stimulus. The CPT consisted of a Plexiglas water basin (Julabo®, Seelbach,
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