



The effects of contextual threat and anxiety on affective startle modulation



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ABSTRACT

The startle reflex is attenuated and potentiated when participants are viewing pleasant and unpleasant images, respectively. Research demonstrates that threatening contexts also potentiate startle, but it remains unclear how a threatening context might impact startle modulation to emotional images, especially as a function of trait anxiety. The present study measured startle reactivity while 43 participants viewed pleasant, unpleasant, and neutral images across conditions of threat-of-shock and safety (i.e., no shock). Compared to neutral images, startle was potentiated during unpleasant images and attenuated during pleasant images. Threat-of-shock potentiated startle during all picture types, suggesting that threat-of-shock broadly sensitized the defensive system but did not change affective modulation of startle. Lastly, higher levels of trait anxiety were associated with less startle potentiation during unpleasant images across both conditions—a finding in line with previous research demonstrating deficient threat mobilization in response to unpleasant stimuli among highly anxious individuals.

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1. Introduction

The defensive startle response is a cross-species reflex elicited by abrupt and intense sensory stimuli. In humans, the startle response is most notably evidenced by rapid eye closure, and this eye blink reflex is modulated by the motivational state of the individual (Grillon & Baas, 2003; Landis & Hunt, 1939; Lang, Bradley, & Cuthbert, 1997). Specifically, the startle reflex is potentiated when an individual's aversive motivational system is primed, and attenuated when the appetitive motivational system is primed (Lang et al., 1997). A large literature has established that viewing arousing unpleasant pictures (e.g., scenes of human threat, animal attack, or mutilation) significantly potentiates the startle reflex, whereas viewing arousing pleasant pictures (e.g., happy babies, smiling faces, or erotica) attenuates the reflex (see Lang, 1995; Lang et al., 1997).

Affective modulation of startle has similarly been demonstrated when participants view expressive faces. For instance, viewing angry faces has been shown to potentiate startle (Dunning, Auriemmo, Castille, & Hajcak, 2010; Hess, Sabourin, & Kleck, 2007; Springer, Rosas, McGetrick, & Bowers, 2007); however, other types of aversive faces (e.g., fearful expressions) do not reliably potentiate

the startle reflex (Anokhin & Golosheykin, 2010; Dunsmoor, Mitroff, & LaBar, 2009; Grillon & Charney, 2011; Springer et al., 2007). Recent work has suggested that aversive faces may not engage the defensive motivational system to the same degree as highly unpleasant scenes. For instance, when viewing facial expressions and pictures from the International Affective Picture System (IAPS; Lang, Bradley, & Cuthbert, 2005), participants rated expressive faces lower on dimensions of arousal and valence compared to IAPS (Britton, Taylor, Sudheimer, & Liberzon, 2006). Moreover, in a direct comparison of IAPS and emotional faces, Wangelin, Bradley, Kastner, and Lang (2012) found that facial expressions did not activate affective physiological responses (i.e., startle response, skin conductance, and event related potentials) as strongly as emotional scenes.

In addition to unpleasant pictures, the startle reflex is also potentiated in the presence of a variety of other unpleasant, arousing stimuli. For instance, conditioned stimuli that predict an electric shock can prime the defensive motivational system, leading to potentiated startle response (Brown, Kalish, & Farber, 1951; Davis, Falls, Campeau, & Kim, 1993; Davis, 2006; Grillon & Baas, 2003; Grillon & Davis, 1997). Startle magnitude can track the generalization (Hajcak et al., 2009; Lissek et al., 2008, 2010) and extinction (Alvarez, Johnson, & Grillon, 2007; Orr et al., 2000; Vansteenwegen, Crombez, Baeyens, & Eelen, 1998) of conditioned stimuli as well. Further, cues that predict other forms of aversive states, like difficulty in respiration, have also led to potentiation of the startle reflex (Lang et al., 2011).

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In addition to foreground stimuli, threatening contexts also engage the defensive motivational system. For example, threat-of-shock compared to safe (i.e., no-shock) experimental conditions reliably potentiate the startle reflex (Bradley, Moulder, & Lang, 2005; Bradley, Silakowski, & Lang, 2008; Grillon, Ameli, Woods, Merikangas, & Davis, 1991); startle was higher in amplitude during conditions when participants anticipated an electric shock compared to when shocks were not anticipated, and these effects were evident prior to the actual delivery of a single shock (Grillon et al., 1991). In a fear conditioning paradigm using virtual reality environments, Grillon et al. (2006) had participants explore three different contexts: a room where no shocks would occur, a room with predictable shocks, and a room with unpredictable shocks. Anxiety ratings were highest in the room with unpredictable shocks—and the baseline startle reflex was largest in the context with unpredictable aversive shocks compared to the other two rooms (Grillon, Baas, Cornwell, & Johnson, 2006). Other forms of contextual threat, such as the threat of aversive abdominal stimulation (Hubbard et al., 2011; Naliboff et al., 2008) or a strenuous hyperventilation challenge (Melzig, Holtz, Michalowski, & Hamm, 2011), also potentiate the startle reflex.

Thus, the startle reflex is potentiated when participants view unpleasant visual stimuli, stimuli that predict punishment or aversive states, and when participants are in threatening contexts. However, much less work has been done examining how unpleasant visual stimuli and threatening contexts might interact to impact the defensive startle reflex, although existing research suggests several possibilities. One possibility is that contextual threat may selectively sensitize defensive responding during aversive stimuli, but have little or no effect on defensive responding during pleasantly valenced stimuli. In support of this possibility, Grillon and Charney (2011) had participants view fearful and neutral faces during alternating phases of threat-of-shock or safety (happy faces were also compared to neutral faces, but in a separate testing session). Fearful faces potentiated startle during the threat-of-shock condition, but not during a safety condition. These data suggest that a threatening context may uniquely sensitize defensive mobilization to threatening foreground stimuli; indeed, it would seem adaptive for defense mechanisms to respond selectively to potentially harmful or aversive stimuli under conditions of heightened threat (Grillon & Charney, 2011).

A second possibility is that contextual threat may sensitize defensive responding, regardless of foreground stimulus valence. For example, participants in a non-stressful control group demonstrated greater amygdala reactivity to angry and fearful faces compared to happy faces (van Marle, Hermans, Qin, & Fernández, 2009); however, for participants undergoing an acute stress induction, amygdala response was equally enhanced for all affective faces (angry, fearful, and happy; van Marle et al., 2009). Other lines of work also support the notion that threat-of-shock may indiscriminately sensitize the processing of all sensory stimuli, as evidenced by increased amygdala and insula activity (Cornwell et al., 2007) and increased brainstem auditory evoked potentials (Baas, Milstein, Donlevy, & Grillon, 2006). Taken together, these data suggest that a stressful or threatening context may sensitize defensive reactivity to all stimuli, at a loss of stimulus specificity.

The primary aim of the present study was to examine how a threatening context would impact the well-established pattern of startle modulation to arousing affective pictures. To this end, we measured startle reactivity while participants viewed pleasant, unpleasant, and neutral images across conditions of threat-of-shock and safety. We expect that during the safety condition, startle response will be attenuated during pleasant pictures and potentiated during unpleasant pictures. Considering the previously reviewed literature, two competing hypotheses emerge for defensive reactivity during the threat-of-shock condition: (1)

if threatening contexts activate defensive reactivity uniquely to aversive stimuli, then startle response should be increasingly potentiated only during the unpleasant pictures; and (2) if threatening contexts sensitize the processing of stimuli indiscriminately, then startle response during all picture types should be potentiated, but the pattern of affective modulation should remain intact.

Lastly, it would be fruitful to examine whether individual differences in anxiety might impact affective startle modulation in the presence and absence of threat-of-shock. Some evidence exists for associations between enhanced startle potentiation and individual differences in anxiety-related traits, such as increased levels of fearfulness, behavioral inhibition, and harm avoidance (as reviewed in Grillon & Baas, 2003). Yet other studies find no associations between startle modulation and characteristics of anxiety, including anxious apprehension and defensiveness (Nitschke et al., 2002) and negative affectivity (Cook, Davis, Hawk, Spence, & Gautier, 1992). The relation between startle and trait anxiety (e.g., scores on the trait scale of the STAI) in particular remains unclear in existing literature. For instance, in a threat-of-shock study involving healthy participants, Grillon, Ameli, Foot, and Davis (1993) found no association between fear-potentiated startle response and trait anxiety. In a social threat paradigm, startle magnitude in anticipation of giving a speech was positively correlated with trait social anxiety, but not general trait anxiety (Cornwell, Johnson, Berardi, & Grillon, 2006). Among clinical samples, startle was not related to trait anxiety in a threat-of-shock paradigm among PTSD patients (Grillon, Morgan, Davis, & Southwick, 1998). However, when focused on a subgroup of PTSD patients (i.e., those having experienced multiple compared to single traumas), McTeague et al. (2010) found that reduced startle reactivity to aversive imagery was concomitant with greater trait anxiety and depressive comorbidity. Moreover, among a large sample of patients with a variety of anxiety disorders (specific phobia, social phobia, panic disorder, and generalized anxiety disorder), lower levels of trait anxiety were found to predict larger startle reactivity during fearful imagery (Lang & McTeague, 2009). Given these findings, more work is needed to clarify the relation between defensive startle reactivity and trait anxiety. Hence, a secondary and exploratory aim of this study is to examine how individual differences in trait anxiety may relate to affective startle modulation under contexts of threat-of-shock and safety.

2. Methods

2.1. Participants

Fifty-two undergraduate students participated in the present paradigm. Of those, nine were excluded from analysis due to poor quality physiological recordings (excessive EMG artifacts and/or startle non-responders), leaving 43 participants (30 female and 13 male) with a mean age of 20.02 ($SD=2.55$) in the present study. All participants gave written informed consent and received course credit for their participation. This research was approved by the Stony Brook University Institutional Review Board.

2.2. Stimuli

Fifty-four pictures from the International Affective Picture System (IAPS; Lang, Bradley, et al., 2005) were selected: 18 unpleasant images that depicted threat (e.g., knife attacks), 18 pleasant images that depicted erotica (e.g., nude couples), and 18 neutral images that included people (e.g., working at a computer desk).¹ Normative valence ratings significantly differed between all categories of picture content: pleasant ($M=6.57$, $SD=0.43$) and neutral ($M=5.45$, $SD=0.36$; $t(34)=8.50$, $p<0.001$, $d=2.82$), unpleasant ($M=2.85$, $SD=0.64$) and neutral ($t(34)=-15.02$, $p<0.001$,

¹ The numbers of the IAPS pictures used were the following: pleasant (4604, 4647, 4651, 4652, 4658, 4659, 4660, 4664, 4668, 4669, 4670, 4677, 4680, 4687, 4693, 4697, 4800, 4810), unpleasant (1050, 1120, 1205, 1300, 1304, 1932, 6230, 6231, 6243, 6244, 6250, 6300, 6350, 6370, 6550, 6563, 6570, 9425), and neutral (2026, 2036, 2038, 2191, 2214, 2374, 2384, 2575, 2580, 5390, 7033, 7041, 7081, 7140, 7500, 7504, 7513, 7546).

$d=5.01$), and pleasant and unpleasant ($t(34)=-20.45$, $p<0.001$, $d=6.82$). Additionally, pleasant ($M=6.53$, $SD=0.33$) and unpleasant ($M=6.56$, $SD=0.49$) pictures were reliably higher on normative arousal ratings than neutral ($M=3.44$, $SD=0.49$) pictures ($t(34)=22.30$, $p<0.001$, $d=7.40$ and $t(34)=19.03$, $p<0.001$, $d=6.37$, respectively), but did not differ from each other ($t(34)=0.18$, $p>0.80$, $d=.07$).

All the pictures were presented on a Pentium class computer, using Presentation software (Neurobehavioral Systems, Inc., Albany, CA, USA) for the presentation and timing of all stimuli. Each picture was displayed in color and occupied the entirety of a 19-in. (48.26 cm) monitor. At a viewing distance of approximately 24 in. (60.96 cm), each picture occupied approximately 40° of visual angle horizontally and vertically.

The acoustic startle probe was a 50 ms burst of white noise that was set to a volume of 105 dB and was delivered through headphones using a tone generator (Contact Precision Instruments, Cambridge, MA, USA). Electrical shocks were delivered to the participant's left tricep using an electrical stimulator (Contact Precision Instruments) that produced 60 Hz constant AC stimulation between 0 and 5 mA for 500 ms. All stimuli and psychophysiological responses were presented and recorded using PYSYLAB hardware and PYSYLAB 8 software (Contact Precision Instruments).

2.3. Procedure

After obtaining informed consent, participants were given detailed task instructions. Each participant completed two conditions of the experiment: receiving unpredictable shocks was possible in one condition (threat-of-shock) but not the other (safety)—the order of conditions was counterbalanced across participants. All participants were instructed that they would be viewing a series of pictures on the computer screen. For the threat-of-shock condition only, the experimenter worked with the participant to set the level of shock intensity, which was determined on an individual basis. Specifically, participants initially received a mild shock, which was gradually raised based on participant feedback. Participants were asked to choose a level of shock that felt uncomfortable but manageable and within their tolerance for pain. Participants were told that the computer would randomly administer between 0 and 9 shocks during the task, and they would not know when or exactly how many shocks would be delivered. During the actual experiment, all participants received exactly 2 shocks—one following trial 2 and one following trial 17. In the safety condition, participants were told that they would not receive electric shocks. Given data that demonstrates an increase in startle response simply by having electrodes attached (Grillon & Ameli, 1998), electrodes were always removed for the no-shock condition.

The experiment began with a four trial startle habituation phase used to elicit initial extreme startle responses. In both the threat-of-shock and safety conditions, participants were presented with 27 trials of randomly ordered pictures (9 unpleasant, 9 pleasant, and 9 neutral scenes). In order to avoid possible habituation effects from viewing the same pictures twice, different picture sets were used in the threat-of-shock and safety conditions (these picture sets were counterbalanced across participants). During each trial, a picture remained on the screen for 6 s and a fixation mark (+) was presented between each picture for 10–11 s. Startle probes were presented during 6 of the 9 trials for each picture category, and occurred randomly between 3 and 5 s following picture onset. In addition, 6 inter-trial interval (ITI) startle probes occurred randomly between 3 and 7 s following stimulus offset in order to reduce probe predictability.

After each condition of the experiment was completed, participants answered a 1-item self-report rating of distress ("how anxious/distressed did you feel during this task?") using a 4-point Likert-type scale that ranged from "Not at all" (0) to "Very" (3). In addition, participants completed the 20-item trait scale of the State-Trait Anxiety Inventory (STAI; Spielberger, 1983) at the end of the experiment. The STAI is a widely used measure of anxiety in adults; the trait scale demonstrates strong concurrent validity with other anxiety scales (correlations ranging from .73 to .85; Spielberger, Reheiser, Ritterband, Sydeman, & Unger, 1995) and demonstrates strong test-retest reliability as well (correlations ranging from .69 to .89; Spielberger, 1983).

2.4. Data recording, reduction, and analysis

Startle responses were recorded from EMG activity using a PYSYLAB Stand Alone Monitor Unit (SAM) and BioAmplifier (Contact Precision Instruments). Two 4 mm Ag–AgCl electrodes were positioned approximately 25 mm apart over the orbicularis oculi muscle beneath the left eye, and an isolated ground was positioned in the middle of the forehead. EMG activity was sampled at 1000 Hz, and band-pass filtered between 30 and 500 Hz. Startle EMG was rectified in a 200 ms window beginning 50 ms before the startle probe and smoothed using a 6-point running average. Startle amplitude was quantified as the maximum response in a 100 ms post-probe window relative to the average activity in the 50 ms pre-probe baseline period. For participants to be included in the analyses, at least 50% of startle-probe trials had to be free of artifacts and/or non-responses; non-response trials were entered as 0 and were included in the present analyses. Startle magnitude for each subject was then converted to T scores to reduce between-subject variability unrelated to variables of interest. Specifically, all startle responses were deviated from the overall startle mean (across both shock and safety conditions) for each participant.

All measures were statistically analyzed using IBM SPSS Statistics 19.0. Startle data was examined by conducting a 2 (Condition Type: threat-of-shock vs safety) \times 4 (Stimulus Type: pleasant, unpleasant, neutral, ITI) repeated-measures ANOVA with

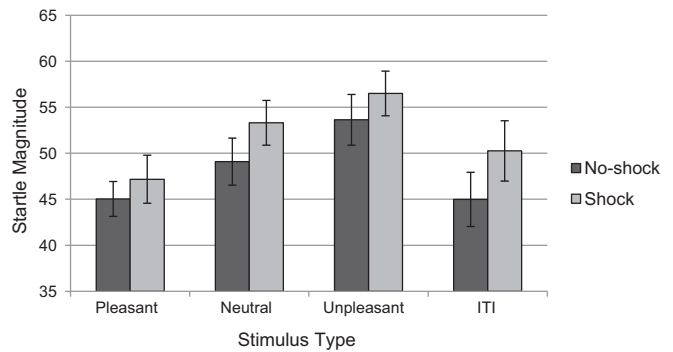


Fig. 1. Average magnitude of startle responses (T scored) elicited while participants viewed pleasant, neutral, and unpleasant pictures during shock and no-shock phases. Startle magnitude elicited by probes during inter-trial intervals (ITI) is also presented (far right). Error bars are 95% confidence intervals.

Greenhouse–Geisser correction applied.² Significant omnibus effects were examined using post hoc paired-samples t -tests with Bonferroni's correction applied for multiple comparisons. To examine emotional modulation of the startle response, we created difference scores by subtracting neutral from both pleasant and unpleasant picture startle magnitude. These difference scores were then correlated with scores on the trait scale of the STAI (Spielberger, 1983) and self-reported distress in order to examine associations between anxiety and emotional modulation of the startle response. This was done separately for the shock and safety conditions.

3. Results

Consistent with the impression from Fig. 1, startle magnitude was larger during the threat-of-shock ($M=51.81$, $SD=5.36$) than safety ($M=48.19$, $SD=5.36$) condition ($F(1,42)=4.92$, $p<0.05$, $\eta_p^2=.11$). Additionally, startle magnitude varied as a function of picture type ($F(3,126)=18.51$, $p<0.001$, $\eta_p^2=.31$); however, the interaction between condition and stimulus type did not reach significance ($F(3,126)=0.92$, $p>0.05$, $\eta_p^2=.02$).³ When collapsing across conditions, startle responses were significantly potentiated during unpleasant ($M=55.07$, $SD=4.90$) compared to neutral ($M=51.20$, $SD=4.97$; $t(42)=2.22$, $p<0.05$, $d=.78$) pictures, and were significantly attenuated during pleasant ($M=46.11$, $SD=5.26$) compared to neutral pictures ($t(42)=-3.93$, $p<0.001$, $d=.99$). Further, startle responses during unpleasant pictures were potentiated compared to pleasant pictures ($t(42)=-7.07$, $p<0.001$, $d=1.76$). Lastly, although participants rated the threat-of-shock ($M=1.49$, $SD=0.94$) compared to safety ($M=1.26$, $SD=0.93$) condition as

² Given the counterbalanced study design, we initially analyzed data using a 2 (Condition Order: shock first vs safety first) \times 2 (Condition Type: threat-of-shock vs safety) \times 4 (Stimulus Type: pleasant, unpleasant, neutral, and ITI) mixed ANOVA to determine whether condition order affected startle data. Results revealed an interaction between condition order and condition type ($F(1,41)=111.04$, $p<0.001$, $\eta_p^2=.73$), such that overall startle magnitude was larger for whichever condition came first (all $t_s(41)>|10.53|$, $p<0.001$), and this difference was greater among participants whose first condition was shock compared to safety ($t(41)=8.28$, $p<0.001$). This is likely the result of startle habituation over time. Importantly, condition order did not interact with any other variable. For all subsequent analyses, we collapsed across condition order.

³ Recent work by Vytal, Cornwell, Arkin, and Grillon (2012) suggests that increased cognitive load can reduce anxiety. Related to our study, it is possible that higher cognitive load from processing affective pictures could take resources away from the anticipatory anxiety elicited by threat of shock—this would be evidenced by larger startle potentiation during shock conditions for ITI stimuli compared to picture stimuli. To examine this possibility, we conducted a 2 (Shock vs No-shock) \times 2 (ITI vs Pictures) repeated-measures ANOVA. Results revealed main effects for each variable but no interaction emerged. Specifically, startle response was larger in shock compared to no-shock conditions ($F(1,42)=5.11$, $p<0.05$, $\eta_p^2=.11$) as well as larger during pictures compared to ITIs ($F(1,42)=6.95$, $p<0.05$, $\eta_p^2=.14$). Further, startle potentiation during the threat-of-shock context did not interact with picture type/ITI ($F(1,42)=1.31$, $p>0.25$, $\eta_p^2=.03$).

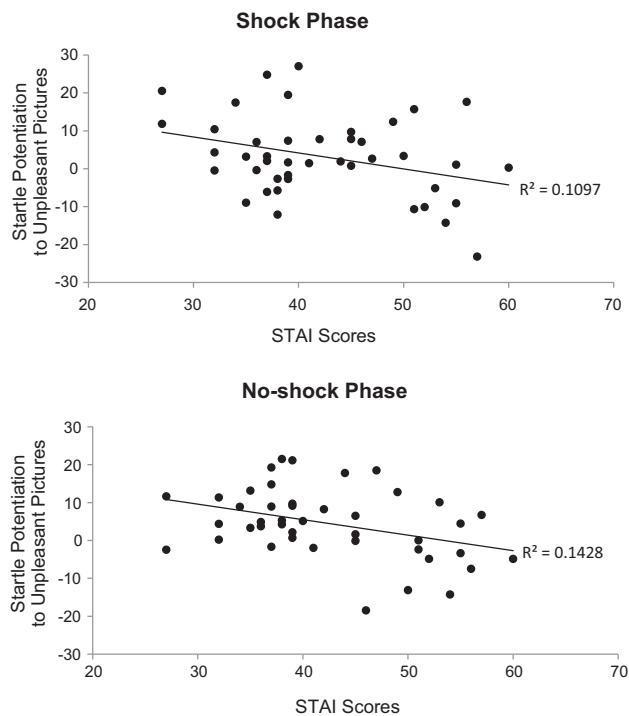


Fig. 2. Scatterplots depicting the correlations between startle potentiation to unpleasant (compared to neutral) pictures and scores on the STAI during the shock (top; $r = -.33$, $p < 0.05$) and no-shock phases (bottom; $r = -.38$, $p < 0.05$).

slightly more distressing, this difference was only marginally significant ($t(42) = 1.81$, $p > 0.07$, $d = .25$).

Participants' average score on the trait scale of the STAI was 42.35 ($SD = 8.41$) with a minimum score of 27 and maximum of 60. Correlational analyses revealed that higher levels of trait anxiety were associated with smaller startle potentiation during unpleasant compared to neutral pictures in both the threat-of-shock ($r = -.33$, $p < 0.05$) and safety conditions ($r = -.38$, $p < 0.05$; see Fig. 2). However, trait anxiety was not associated with startle attenuation during pleasant pictures, or ITI startle amplitude, in either experimental condition (all r s $< |.14|$, p s > 0.35). When collapsing across all stimuli types (pleasant, unpleasant, neutral, and ITI), startle magnitude during the threat-of-shock ($r = -.12$, $p > 0.40$) and safety ($r = .12$, $p > 0.40$) conditions was not significantly related to trait anxiety. Lastly, self-reported ratings of distress during both conditions of the experiment were not correlated with startle magnitudes or trait anxiety (all r s $< |.17|$, p s > 0.05).

4. Discussion

The present study sought to examine how a threatening compared to safe context might impact affective modulation of the startle reflex. In line with previous research, we found the expected linear pattern of affective startle modulation (i.e., unpleasant $>$ neutral $>$ pleasant) across both conditions of the experiment. In addition, our results further supported the notion that threat-of-shock significantly potentiates the startle reflex. Indeed, across all picture types (including ITI startle probes), startle magnitudes were larger in the shock compared to safety condition. However, threat-of-shock did not selectively sensitize the defensive startle reflex during aversive content—the degree to which startle was potentiated in the threatening context was consistent across unpleasant, neutral, and pleasant pictures. Therefore, the present results appear to be in line with studies

that find threatening contexts broadly sensitize the processing of sensory stimuli (Baas et al., 2006; Cornwell et al., 2007).⁴

Grillon and Charney (2011) found an interaction between threatening context and affective modulation of the startle reflex during emotional faces; specifically, viewing fearful compared to neutral faces was associated with an increased startle reflex, but only within a threat-of-shock condition. One methodological difference in the present study is that we used highly arousing emotional scenes from the IAPS rather than faces. Other research confirms that facial stimuli may not activate startle to the same degree as more arousing affective pictures (Dunning et al., 2010; Hess et al., 2007; Springer et al., 2007; Wangelin et al., 2012); indeed, pictures with highly arousing content, such as erotica or human threat, are associated with the most pronounced modulation of the startle reflex (Bradley, Codispoti, & Lang, 2006). Collectively, these data suggest that a threatening context may alter defensive mobilization in response to aversive faces, whereas contextual threat may additively determine startle potentiation for more arousing stimuli. Future studies could address this possibility by examining the startle reflex during both IAPS and facial stimuli in threatening and safe contexts.

When examining individual differences in trait anxiety, we found that participants with higher levels of anxiety displayed less startle potentiation while viewing unpleasant pictures. This correlation was apparent in both the threat-of-shock and safety conditions, demonstrating that the relationship between high anxiety and less startle potentiation was evident with or without a manipulation of contextual threat. These results are in line with research that finds deficient threat mobilization among patients with high levels of diffuse and long-lasting anxiety (Cook, Melamed, Cuthbert, McNeil, & Lang, 1988; Cuthbert et al., 2003; Lang & McTeague, 2009; Lang, McTeague, Cuthbert, 2005, 2007; McNeil, Vrana, Melamed, Cuthbert, & Lang, 1993; McTeague et al., 2009, 2010). Indeed, when Lang and McTeague (2009) subdivided a large sample of patients into groups of low and high trait anxiety (assessed using the STAI), reduced startle reactivity during aversive imagery was found for the high anxious group. Preliminary work by McTeague and Lang (2012) suggests that attenuated startle response among those with chronic, pervasive anxiety could be due to amygdala hypoactivation. Along these lines, the present study also used the STAI and thus extends the work of Lang and McTeague (2009) to a non-clinical sample using affective modulation of startle in the context of IAPS pictures (rather than narrative imagery). Likewise, future research may benefit from using the current paradigm in clinical anxiety samples.

It should be noted that the average STAI score of our sample ($M = 42.35$, $SD = 8.41$) was higher than comparable non-clinical samples (e.g., $M = 31.3$, $SD = 6.4$; Grillon & Charney, 2011) or matched control groups (e.g., $M = 30.8$, $SD = 8.57$; McTeague et al., 2009; McTeague, Lang, Laplante, & Bradley, 2011), but was lower than average scores reported in patients with generalized social phobia ($M = 53.35$, $SD = 6.13$; McTeague et al., 2009) or panic disorder with agoraphobia ($M = 55.26$, $SD = 10.11$; McTeague et al., 2011). In the present study, STAI data was collected at the end of the experimental session; although a trait measure should not be affected by recent experiences, it is possible that this methodological detail could have contributed to higher trait anxiety scores. Further, the

⁴ Other preliminary data, that has not been peer-reviewed, also supports the present findings. Costa, Bradley, and Lang (2008) examined startle response to unpleasant and neutral pictures (pleasant pictures were not included) under conditions of threat-of-shock and safety. Threat-of-shock potentiated startle response, and unpleasant compared to neutral pictures elicited potentiated startle as well. Moreover, no interaction emerged, such that affective modulation of startle (unpleasant $>$ neutral) remained intact across both threat-of-shock and safety conditions (Costa et al., 2008).

higher STAI scores in our sample relative to studies finding no relation between trait anxiety and startle (Grillon & Charney, 2011; Grillon et al., 1993) may further explain the emergence of reduced startle reactivity during unpleasant pictures. Regardless, given that deficient threat mobilization has previously been reported only in highly anxious clinical samples, cautious interpretation of the present results and future replication is warranted.

Another possibility is that anxiety is characterized by an attentional pattern of initial vigilance for threat (Bradley, Mogg, Falla, & Hamilton, 1998; Broadbent & Broadbent, 1988; MacNamara & Hajcak, 2010; Mogg et al., 2000), followed by avoidance of threat (Holmes, Nielsen, & Green, 2008; Koster, Crombez, Verschuere, Van Damme, & Wiersema, 2006; Mogg, Bradley, Miles, & Dixon, 2004; Muhlberger et al., 2009). In the present task, startle probes were presented between 3 and 5 s following picture onset; although we did not specifically measure attentional engagement, it is possible that more anxious individuals were engaged in greater cognitive avoidance of threatening content by this point (a possibility that could only be determined by future research). Also, given that no correlation emerged between trait anxiety and overall startle magnitude in the threat-of-shock condition, it is possible that participants may have found it easier to avoid processing the arousing pictures rather than to avoid thinking about the shock.

Another direction for future research would be to examine the possible influence of depression on this pattern of results, especially considering the high comorbidity among depression and anxiety (Brown & Barlow, 2002; Brown, Campbell, Lehman, Grisham, & Mancill, 2001). For example, when dividing a sample of anxiety patients into low and high levels of depression, smaller startle responses during aversive imagery were found for the high compared to low depression group (Lang & McTeague, 2009). In regard to the present paradigm, future studies should aim to tease apart symptoms of depression and anxiety on defensive reactivity during conditions of threat versus safety, and examine these effects as a function of a broader construct, such as negative affectivity (see McTeague et al., 2009, 2010).

Taken together, threat-of-shock in the present study potentiated startle response regardless of the foreground affective stimulus, suggesting that increased states of anxiety broadly sensitize the defensive startle reflex, but do not impact affective modulation of startle. These results are consistent with work parsing cue-related and contextual effects on startle; potentiated startle is elicited not only by cue-conditioned fear via the central nucleus of the amygdala, but also by generalized contextual anxiety via the bed nucleus of the stria terminalis (Lang, Bradley, & Cuthbert, 1998; Lee & Davis, 1997; Walker & Davis, 1997). Additionally, we found that individuals who scored higher on measures of trait anxiety exhibited less startle potentiation during unpleasant compared to neutral pictures. This association was evident in both the threat-of-shock and safety conditions; combined with previous research, this suggests that deficient threat mobilization to unpleasant stimuli among highly anxious participants can occur regardless of an aversive or neutral context.

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