Startle reflex as a physiological measure of emotion regulation

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Abstract

In the present study, the startle blink reflex is used as a measure of emotion regulation to affective picture stimuli. Based on the biphasic theory of emotion, it is hypothesized that the startle response will be largest in magnitude in the presence of negative emotional stimuli (Vrana, Spence, & Lang, 1988). It is also hypothesized that when attempting to decrease emotion, participants will show smaller blink magnitudes to negative images, and larger blink magnitudes when attempting to decrease emotion to positive images due to the aversive nature of the startle reflex. The present study highlights the difficulty of finding emotion regulation to positive images with the startle blink paradigm. Participants were 6 female undergraduate students who viewed negative, positive and neutral affective picture stimuli, and attempted to either maintain or suppress their emotional responses to the images. Emotion modulation of the startle response was recorded before regulation instruction onset. Significant differences were found in blink magnitude for emotion modulation between positive and negative images, but neutral images were not significantly different from either. For negative images, blink magnitudes during emotion suppression were significantly smaller than when maintaining emotion. No significant regulation differences were found for the positive images.

Keywords: *psychophysiology, emotion regulation, emotion modulation, startle blink*

Emotion regulation can be thought of as consciously or unconsciously altering one's behavioural, cognitive, and/or physiological emotional response tendencies towards a goal (Gross, 1998). The ability to regulate emotions is important for both mental and physical well-being, and social interaction (Kim & Hamann, 2007). Dysfunction in emotion regulatory abilities has been thought to play a role in many forms of psychopathology, such as anxiety, depression (Davidson, 2000; Jackson, Larson, & Davidson, 2000), and eating disorders (Clyne, Latner, Gleaves, & Blampied, 2010; Harrison, Sullivan, Tchanturia, & Treasure, 2010; Whiteside, Chen, Neighbors, Hunter, Lo, & Larimer, 2007). Understanding possible mechanisms associated with emotion regulation difficulties are essential to addressing these problems in clinical populations. Although emotion regulation deficits are a core feature of many forms of

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psychopathology, physiological evidence of these emotion regulatory deficits is severely lacking. A large body of research that has been used to correlate psychopathology and emotion regulation has been self-report, (Aldao & Nolen-Hoeksema, 2010; Whiteside et al., 2007) and the ability of individuals to accurately assess their own emotion regulatory abilities has not been confirmed. Little research exists that overtly examines emotion regulation and its possible deficits in an experimental setting using psychophysiological measures of emotion regulation.

The startle blink paradigm is an advantageous physiological tool as it has the ability to differentiate between positive and negative emotional states. This paradigm involves administering a loud blast of white noise, which acts to startle the participant causing their eyes to twitch reflexively. This startle reflex is measured using electromyographic (EMG) sensors placed on the *obicularis oculi* muscle below the left eye. The startle reflex triggers the defensive aversive response of the biphasic appetitive-aversive dimension of emotion; this automatic response is enhanced when the emotional context matches the reflex (Vrana et al., 1988). Funayama, Grillon, Davis, & Phelps (2001) found that the startle response is mediated by the right medial temporal lobe, specifically the amygdala, which plays a key role in aversive or negative emotional states, supporting the theory that the startle response is an aversive reflex. Therefore, the startle response is expected to be potentiated, or larger in magnitude, when it occurs in the presence of aversive emotional states (Lang, Bradley, & Cuthbert, 1990; Vrana et al., 1988). Along the same reasoning, one would expect that in the presence of appetitive or positive emotional states the startle reflex would be attenuated, or smaller in magnitude, due to a mismatch between the reflex and the emotional context (Lang et al., 1990; Vrana et al., 1988). Indeed, research has shown that the startle blink reflex is an effective measure of emotion modulation, with the startle reflex being potentiated in the presence of negative emotions, attenuated in the presence of positive emotions, and moderate in size when there is a lack of emotion (Bradley, Cuthbert, & Lang, 1991; Vrana et al., 1988). An increase in startle magnitude from positive, neutral, to negative emotional contexts has been found when emotions are elicited using visual stimuli (Bradley et al., 1991; Vrana et al., 1988), olfactory stimuli (Miltner, Matjak, Braun, Diekmann, & Brody, 1994), narrative stimuli, such as remembering events (Cook, Hawk, Davis, & Stevenson, 1991), and threat of shock paradigms (Greenwald, Bradley, Cuthbert, & Lang, 1998; Lissek et al., 2007).

Research showing that the startle blink response is an effective measure of emotion regulation to affective stimuli is less conclusive. The conditions common to emotion regulation research are: "maintain", which instructs participants to focus on their emotional response to the stimuli without changing it, "suppress", which involves decreasing emotional response, and "enhance", which involves increasing emotional response to a stimuli (Eippert et al., 2007; Jackson et al., 2000; Lissek et al., 2007; Ray, McRae, Ochsner, & Gross, 2010). Jackson et al. (2000) found both emotion modulation and subsequent regulation to negative pictures compared to neutral. Replication studies by Lissek et al. (2007), which included a threat of shock paradigm, and Lee, Shackman, Jackson, & Davidson (2009) had similar findings of modulation and regulation to aversive contexts compared to neutral.

In concordance with previous research, Eippert et al. (2007) and Ray et al. (2010) found larger blink magnitudes for the negative images from the enhance condition compared to maintain and suppress conditions. However, both studies failed to find significant startle blink differences for the suppress condition compared to the maintain condition. One possible reason for this finding is that the timing of the probes may have been too early to accurately detect suppression, occurring 2 s after the regulation cue compared to the common 3 s delay (Eippert et al., 2007). In further support of this argument, fMRI results showed suppressed amygdala activity during the suppress condition compared to the maintain and enhance conditions, but the suppressed activation occurred after the startle probe measured regulation. Another possible explanation for both studies failing to detect significant regulation of the startle response for the suppress compared to the maintain cue is that both focused only on reappraisal as an emotion regulation technique, as opposed to subjects choosing their own regulation strategies. Perhaps cognitive reappraisal, a cognitive strategy that involves re-evaluating the meaning of an emotional stimulus (Gross, 1998), is not the most effective negative emotion suppression strategy for all individuals.

For all of the emotion regulation studies mentioned thus far, positive picture stimuli was not included. Jackson et al. (2000) contends that the failure to include positive images was due to a difficultly finding attenuation of the startle response to positive emotion compared to neutral during a pilot study. Difficulty finding the expected modulation to positive affective stimuli compared to neutral stimuli is not uncommon (Dillon & La Bar, 2005; Driscoll, Tranel, & Anderson, 2008). Studies that have

included positive emotion regulation have hypothesized that arousal, and not valence, influences the startle response (Dillon & La Bar, 2005; Driscoll et al., 2008). Dillon & La Bar (2005) compared regulation to positive, neutral, and negative stimuli and found that the enhance cue during positive and negative picture presentation elicited larger blink magnitudes than the maintain and suppress cues, this finding runs counter to what would be expected from the aversive matching hypothesis, where enhancing positive emotion should elicit the smallest blink magnitudes (Bradley et al., 1993; Lang et al., 1990; Vrana et al., 1988). The authors contended that this finding suggests that the startle blink response is arousal dependent, meaning that the level of arousal will alter the startle magnitude, regardless of emotional valence. Therefore, increasing arousal will always result in larger startle responses, whether the emotion is positive or negative in valence (Dillon & La Bar, 2005). However, significant differences in blink magnitude for positive compared to neutral stimuli was not found, even though the positive images were rated as significantly more arousing then the neutral images. Therefore these findings are not sufficient to conclude that the startle response is solely impacted by arousal, clearly valence plays some role. Also, unlike Jackson et al. (2000) and others (Lee et al., 2009; Lissek et al., 2007) no significant difference was found between suppress and maintain conditions. Possible explanations for these contradictory findings may be that Dillon & La Bar (2005) did not utilize a within-groups design, which is common to these studies, because of large individual differences in blink magnitudes, combined with the problems associated with using a small sample size.

Driscoll et al. (2008) also looked at the regulation of positive and negative emotion, and found that during the enhance condition, regardless of valence, there were larger blink magnitudes than during the suppress condition. The authors concluded that these findings suggest that arousal is the main factor that controls the startle response. However, across regulation conditions the startle response for negative images was significantly larger than for positive images, even though both were matched on arousal. Being matched on arousal and maintaining differences in startle blink magnitude suggests that the startle response is also influenced by valence. It should also be noted that Driscoll et al. (2008) used a small sample size in their analysis, and consequently their findings should be interpreted with caution.

Clearly, previous research utilizing physiological measures to assess emotion regulation yields inconsistent and confusing findings, and the exact relationship between the startle response and positive emotional cues is not agreed upon or well understood. Possible reasons for a failure to find startle response differences between neutral and positive stimuli may be due to a difficulty eliciting positive emotion in a lab setting (Jackson et al., 2000), and subjective differences in interpreting stimuli as positive. Research shows that arousal also influences the startle response (Dillon & La Bar, 2005; Driscoll et al., 2008; Lang et al., 1990). To effectively measure emotional valence, arousal must be balanced for positive and negative stimuli; however, some research studies fail to take into account the participant's subjective ratings of arousal or valence to the stimuli (Larson et al., 2000; Lee et al., 2009). Problems with current emotion regulation research using the startle blink paradigm are that a majority

of the research fails to include positive affective stimuli (Jackson et al., 2000; Lee et al., 2009; Lissek et al, 2007), while others employ different methodologies or have very few participants (Dillon & La Bar, 2005; Driscoll et al., 2009).

The purpose of this study is to replicate and extend the findings of previous research on emotion regulation using the startle blink paradigm (Jackson et al., 2000), with the inclusion of positive affective stimuli. To overcome the limitations of previous research a balanced design will be used, matching the frequency of picture type and regulation cues. Probe and trial times that are consistent with previous research of emotion modulation and regulation will also be used (Eippert et al., 2007; Jackson et al., 2000).

Emotion modulation is of high importance in this study as it is an established finding in startle paradigm research (Bradley et al., 1991; Sanchez-Navarro et al., 2008; Vrana et al., 1988); failing to obtain the expected linear relationship of valence may suggest problems in the paradigm that limit the ability to interpret the emotion regulation findings. Subjective ratings of valence and arousal to the images will also be measured in conjunction with normative ratings in order to assure that arousal between positive and negative images are matched, and therefore not responsible for differences in regulation.

In this study the independent variable is the combined picture types and regulation cues, which result in 6 distinct conditions. The dependent variable is the blink magnitude elicited by the startle paradigm. Based on this information two main hypotheses were generated. The first hypothesis is that the startle response will show attenuation during the suppress

negative condition compared to the maintain negative condition. The second hypothesis is that the startle response will show potentiation during the suppress positive condition compared to the maintain positive condition. These hypotheses are based on the concept that the startle response is a defensive response to threat, activating the aversive domain of the appetitive-aversive dimension of emotion (Vrana et al., 1988). Therefore, when a negative emotional state is suppressed, the magnitude of the startle reflex will be decreased, and when a positive emotional state is suppressed the startle reflex will increase in magnitude.

Method

Participants
All procedures were approved by the University of British Columbia Behavioural Research Ethics Board, and all participants provided informed consent before participation. Participants were eight female undergraduate students from the University of British Columbia. This study included only females as it was part of a pilot test for a larger study involving emotion regulation and disordered eating behaviours. As disordered eating is much more common among females, males were excluded. Three participants were recruited through the University's Human Subject Pool for 1.5 course credits; the remaining five participants were volunteers from the University's Clinical and Cognitive Neuroscience Lab. Participants ranged in age from 20 to 23 years ($M = 22$, $SD = 1.06$), and all participants had a minimum of 10 years of English fluency. Two participants were excluded from analysis due to a failure to stay awake and alert during picture presentation.

Stimuli
All images were selected from the IAPS based on normative female arousal and valence ratings. Pictures were selected to create three discrete picture types: negative (low valence, high arousal), positive (high valence, high arousal), and neutral (medium valence, low arousal). Thirty-two of each picture type (positive, negative and neutral) were included in the picture set, 16 per block. The overall normative female ratings for valence were negative $M = 2.16$, $SD = 0.61$, positive $M = 7.56$, $SD = 0.61$, and neutral $M = 5.0$, $SD = 0.23$. Paired t-tests revealed significant differences for valence between negative and positive images $t(62) = -35.16$, $p< 0.001$, negative and neutral images $t(62) = -24.5$, $p< 0.001$, and positive and neutral images $t(62) = -22.06$, $p< 0.001$. The overall female normative arousal ratings were negative $M = 6.47$, $SD = 0.74$, positive $M = 6.02$, $SD = 0.76$, and neutral $M = 2.94$, $SD = 0.5$. Paired t-tests revealed significant differences for arousal between negative and neutral images $t(62) = 22.51$, $p< 0.001$, positive and neutral images $t(62) = -19.12$, $p< 0.001$, and positive and negative images $t(62) = 2.42$, $p< 0.05$. However, follow-up t-tests between negative and positive images for all probed conditions revealed no significant differences in arousal between the positive and negative conditions. Each block was organized in quasi-random order, ensuring that the pictures were counterbalanced for regulation instruction, order of presentation, and time of startle probe. No more than three picture types (same valence, regulation cue, or probe time) occurred in a row.

Procedure
Each participant came in for a testing session that lasted approximately 1.5 hrs. Details

were then provided about the nature of the task, and each participant provided informed consent. EMG sensors were then applied to the *obicularis oculi* muscle of the left eye. EMG sensors at the *obicularis oculi* site are used to measure the size of the startle response, as this muscle is an important component of the startle reflex. Electroencephalographic (EEG) sensors were also applied at this time as part of another study; the EEG data will not be included in the analysis.

After receiving both visual and verbal instructions and engaging in a practice session, participants viewed digitized colour images from the International Affective Picture Set (IAPS; Lang et al., 1999). Each picture was presented for 8 s. A total of 96 pictures were presented in 2 blocks of 48 pictures each. At 4 s post-stimulus onset the regulation cue, either a white equal sign or a red minus sign, appeared on the screen briefly. A white equal sign instructed participants to maintain their emotional response to the picture. A red minus sign instructed participants to decrease the intensity of the emotion they were experiencing in response to the picture. Both regulation instructions were presented for all picture types (positive, negative, and neutral) an equal number of times to maintain consistency across trials. After 8 s a black screen replaced the picture for 5 s. The word "RELAX" then appeared on the screen for 5 s, at this time participants were instructed to stop decreasing or maintaining their emotional response to the previous picture and to get ready for the next picture. During picture presentation an acoustic startle probe (a 95 dB, 50 ms burst of white noise generated by the Audacity 1.3 Beta Unicode Software) was presented to both ears at either 3 s (probe A) or 7 s (probe B) after picture onset. Inter-trial probes were

presented 4 times during the trial to decrease the predictability of the probes. There were 6 conditions in total, combining each picture type and regulation cue, which each occurred 8 times during the course of the experiment. No probe was presented for 24 of the trials. After the task was completed the EEG and EMG sensors were removed and the participants filled out an image rating for each picture on arousal and valence using the Self Assessment Manikin (SAM) valence and arousal scales, with 9 rating high (arousal or valence) and 1 rating low, this is the same scale that was used to measure the normative ratings of the IAPS (Bradley & Lang, 1994). Pictures were presented in the same order that they had originally appeared and picture-viewing time was recorded to assess interest. Participants were also asked to fill-out a strategy questionnaire in which they described the strategies they used to regulate their emotions for each picture type (decrease and maintain for positive, negative and neutral images). After the strategy questionnaire was complete all participants were debriefed on the nature of the study.

Emotion regulation instructions
Participants were left free to decide on how to regulate their emotions effectively. No strategies were provided on how to regulate emotions. However, to separate emotion regulation from inattention, participants were instructed to focus on the picture, and told not to produce thoughts or images that were unrelated to the picture or emotion they were experiencing. For example, in the sample trial participants are shown a picture of a striking snake. Participants are then told that if they are presented with a red minus sign and are required to decrease their emotion, fear is the example given, they should accomplish this by decreasing the

intensity of the fear they are experiencing, and not by thinking of something unrelated to the picture or by trying to produce a different emotion. A similar example is provided for a positive picture in the maintain regulation instruction. All participants are also given a 9 trial, 3 of each picture type, practice session and question period before the actual experiment begins to ensure that they understood the regulation instructions.

EMG data collection and analysis

To measure the startle reflex two Ag-AgCl 4 mm electrodes were placed on the *obicularis oculi* muscle below the left eyelid. The ground electrode was placed at the AFz site. Preparation of the sensors involved briskly brushing the skin with medical gauze, cleaning the area with rubbing alcohol, gently exfoliating with saline gel, and brushing the skin once more with gauze. EMG sensors were filled with Quickgel. Impedance levels of the electrodes below 20 kΩ were accepted (Larson et al., 2000). EMG data was obtained with a Brain Products Inc, QuickAmp 72 System. Brain Vision Recorder was used to record the data and Brain Vision Analyzer was used to process and score the startle responses. Data was sampled at 1000 Hz. Signals were digitally filtered offline with high and low pass frequency filters at 30 Hz and 500 Hz (48 dB/octave roll-off) with a 60 Hz Notch filter. Startle EMG was rectified and smoothed with a 20 ms moving window average. The data was baseline corrected, subtracting the average value of the 50 ms baseline period (before the probe onset) from all time points in the segment. The peak startle magnitude was determined within a window of time extending from the time of probe onset to 120 ms. Trials were excluded if the blink began prior to 15 ms following the probe, if there was excess noise in the baseline period (>10 μV), if there was a visible artifact in the segment, or if the magnitude was greater than 3 standard deviations above the subject's individual mean amplitude. Trials with no discernable startle response were given a magnitude of zero and included in the analysis (Larson et al., 2005). Startle responses were standardized using within subject z-score conversions to normalize data, and to reduce the influence of between subjects variability unrelated to psychological processes (Blumenthal, Cuthbert, Filion, Hackley, Lipp, & Van Boxtel, 2005).

Test-retest reliability of the emotion modulated startle response may not be stable over time; a study by Larson et al. (2000) found low reliability at a second assessment. However, replication of this study that includes subjective ratings of arousal and valence to stimuli are needed to help understand why the reliability of the emotion modulated startle appears to be low. A study by Lee et al. (2009) found low test-retest reliability of the emotion regulated startle response, with decreased sensitivity to regulation, compared to a corrugator measure. The startle paradigm measures emotion over a span of milliseconds, which may account for its variability across assessments, whereas corrugator activity is measured over a larger span of time and is therefore likely to be more consistent. To be confident in the ability of the startle paradigm to measure emotion modulation and regulation over time more research needs to be done testing reliability, and the factors that may influence it.
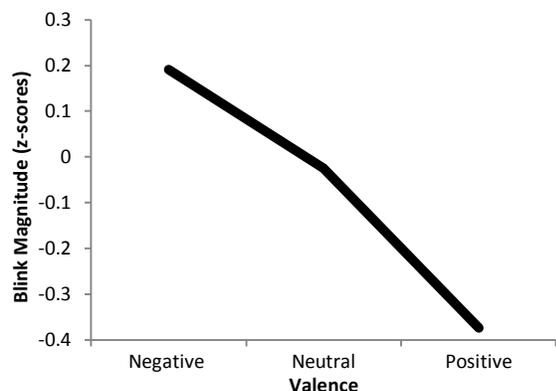
Figure 1. Differences in blink magnitude by affective picture type. Measured 3 s after picture onset.

Results

Image Ratings

A one-way repeated measures ANOVA was conducted to evaluate the participant's subjective ratings of picture valence. A significant effect for valence was revealed, $F(2,10) = 52.28$, $p < 0.001$. Follow-up paired t-tests revealed a significant difference of valence between negative and positive, $t(5) = -7.41$, $p = 0.001$, neutral and negative, $t(5) = 8.48$, $p < 0.001$, and neutral and positive, $t(5) = -5.42$, $p < 0.05$. Therefore, participants rated each picture type as significantly different from each other on emotional valence. A one-way repeated measures ANOVA was conducted to evaluate the participant's subjective ratings of arousal. A significant effect for arousal was revealed, $F(2,10) = 34.37$, $p < 0.001$. Follow-up paired t-tests revealed a significant difference for arousal between neutral and negative $t(5) = -9.44$, $p < 0.001$, and neutral-positive $t(5) = -6.08$, $p < .005$, while the difference between negative and positive, $t(5) = 2.14$, ns were non-significant. In other words, participants rated the positive and negative pictures as high in emotional arousal, while the neutral images were rated as low in emotional arousal. Accordingly, the participant's subjective ratings of valence and arousal for the images matched the normative ratings.

Emotion Modulation

A one-way repeated measures ANOVA was conducted to examine the predicted emotion modulation effects on the startle response to probe A, which was presented 3 s after picture onset, before the regulation cue. The effect of valence was non-significant, $F(2,8) = 3.05$, ns, startle blink magnitudes for each picture type were not significantly different. The hypothesized linear trend of negative>neutral>positive was examined using a linear trend contrast on valence and revealed a significant linear trend, $F(1,4) = 8.39$, $p < 0.05$, blink magnitudes did follow the expected pattern of largest in magnitude to negative images and smallest in magnitude to positive images, supporting the argument that the startle response is an aversive reflex. Follow-up paired t-tests revealed a significant difference between negative and positive, $t(4) = 2.90$, $p < 0.05$ while the difference between negative and neutral, $t(4) = 0.83$, ns and neutral and positive, $t(4) = 1.48$, ns were non-significant (see Figure 1). Therefore, blink magnitudes to negative pictures were significantly larger than to positive pictures. However, no significant differences in blink magnitudes were found between neutral and positive or neutral and negative images.

Emotion Regulation

A 3 (image valence) x 2 (regulation condition) repeated measures ANOVA was conducted for the startle response to probe B, presented at 7 s following picture onset. This revealed a significant main effect for valence, $F(2,8) = 7.08$, $p < 0.05$, blink magnitudes were significantly different across picture types. A main effect for regulation approached significance, $F(1,4) =$

5.08, *p* = 0.087, meaning that differences in blink magnitude between the regulation conditions showed a trend, but were not substantial enough to show significant differences. The interaction between valence and regulation was non-significant, *F*(2,8)=2.74, *ns*. Therefore, blink magnitudes were not significantly affected by the relationship between picture type and regulation cue. The linear contrast for valence was significant with negative>positive>neutral, *F*(1,4) = -8.69, *p*< 0.05. Concretely, a consistent linear trend was found for blink magnitudes and picture type at both probe times, before and after the regulation cue, highlighting the stability of this effect. Paired t-tests were conducted to test the hypothesized regulation effects for the negative and positive images (see Figure 2). A significant difference was found between negative maintain and negative decrease, *t*(4) = 5.01, *p*< 0.05, with blink magnitudes being significantly smaller in the decrease condition than the maintain condition. The difference between positive maintain and positive decrease were non-significant, *t*(4) = 1.06*, ns*. Blink magnitudes were not significantly different in size between the decrease and maintain conditions. As expected there was no significant difference in blink magnitudes between neutral maintain and neutral suppress conditions. Therefore, significant differences in blink magnitude were found only during regulation to the negative images.

Discussion

The initial emotional response to the pictures, before any regulation cues were given, as measured by probe A, was an important characteristic of this study. To help assess that the paradigm was working
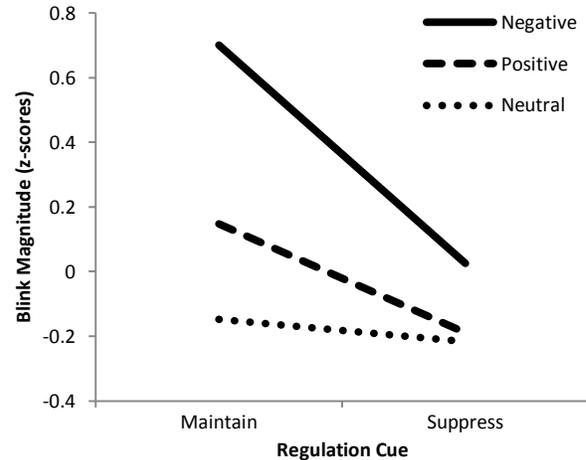

Figure 2. Changes in blink magnitude relative to the affective picture type and regulation cue condition. Measured 7 s after picture onset.

successfully, and that the pictures were eliciting the expected response, we expected to replicate the findings of other emotion modulation research (Bradley et al., 1991; Sanchez-Navarro, Martinez-Selva, Torrente, & Roman, 2008; Vrana et al., 1988). The startle magnitude was significantly larger for negative compared to positive images. However, responses to the neutral images were not significantly different from either the positive or negative images. Failing to find differences in response magnitude for neutral and positive images has been a problem in other emotion regulation studies (Dillon & La Bar, 2005; Driscoll et al., 2008). This finding may indicate that the positive images were not interpreted as positively as expected by the participants.

The lack of significant differences between negative pictures to neutral pictures could be due to a lack of habituation to the startle probe at the beginning of the paradigm. The first probed images in both trials were neutral, and responses to these images were typically much larger than for the rest of the neutral pictures in the task. Therefore, a few large startle responses in the neutral condition

could be responsible for driving the mean up. Leaving out the first trial in the paradigm has been used in previous research as it has been found to be larger than all other responses, and may be useful in future research to avoid habituation effects (Larson et al., 2000). Another possible reason significant differences were not found for positive and negative images to neutral is because of the small sample size in this study. Due to the small sample size, the power to detect smaller significant differences was low.

Emotion regulation to the picture stimuli was also measured by the startle response. The first hypothesis stated that in the suppress negative condition the startle response would be attenuated compared to the maintain negative condition. This hypothesis was supported, during the suppress negative condition the startle response was significantly smaller than during the maintain negative condition. The startle blink paradigm successfully measured voluntary changes in emotional response to a regulation cue for the negative images. The second hypothesis stated that for the positive images, startle blink magnitude would be potentiated during the suppress positive condition compared to the maintain positive condition. As others have found, this hypothesis was not confirmed; the startle response was smaller during the suppress condition than the maintain condition for positive images, however, this difference was not significant (Dillon & La Bar, 2005; Driscoll et al., 2008). In this study the startle blink paradigm was not a sensitive measure of emotion regulation to positive affective stimuli. No significant differences were expected for startle blink magnitudes between the suppress neutral and the maintain neutral conditions, and indeed no differences were found.

Current research has not found the expected emotion regulated startle response to positive images (Dillon & La Bar, 2005; Driscoll et al., 2008). Jackson et al. (2000) has proposed that this may be due to a difficulty in eliciting positive emotion in a laboratory setting. Genuine positive emotion may be harder to elicit in participants than negative emotion, as the participants are sitting in a dark room, unaware of what they will see next, with loud noises occasionally startling them. Also, to ensure that arousal is matched in both negative and positive images, erotic images are often used because other positive images are often rated low on arousal. Although the startle response is supposed to be smallest when viewing erotic images (Lang et al., 1990), it is possible that individuals may vary on their responses to the erotic images in the lab setting, depending on their comfort level.

Another explanation for the failure to find the expected regulation to positive images comes from Dillon and La Bar (2005) who suggested that the emotion regulated startle response is arousal-dependent. Therefore, stimuli that are arousing, regardless of valence, should show an attenuated startle response when suppressed compared to maintained, in opposition to the aversive matching hypothesis. However, both the positive and negative stimuli in the task were matched on arousal for normative and subjective ratings, but positive images failed to show any significant differences between the suppress and maintain conditions, while the negative images did show significant differences between condition. Therefore, arousal does not appear to fully explain why positive emotion regulation has not followed the pattern expected by the biphasic (appetitive-aversive) theory of emotion.

An important limitation of this study, and others, that have attempted to measure positive and negative emotion regulated startle is the small size of the sample (Dillon & La Bar, 2005; Driscoll et al., 2008). The magnitude of the emotion modulated and regulated startle varies widely across individuals (Lang et al., 1990), therefore, a larger sample size is needed before making any strong conclusions about the results. The small sample size reported in this and other research may play a large role in the failure to gather significant or consistent findings of emotion regulation to positive stimuli.

This study only used female undergraduate subjects, as have others (Eippert et al., 2007; Ray et al., 2010), which limits the generalizability of the results. Females were used exclusively in this study as they were participants in part of a larger study involving behaviours most common to women. Although previous research that has included both sexes has found no significant differences of gender (Jackson et al., 2000) it would be beneficial to include both sexes in a study of the emotion regulated startle response with positive and negative stimuli, to be able to further generalize the findings and to test for any possible differences in emotion regulation between gender. How these results relate to other populations, for example, non-educated individuals, older adults or clinical populations is unclear. It would be beneficial if these findings could be replicated with more varied samples.

In the introduction it was suggested that the emotion regulated startle response could potentially be a useful measure of emotion regulation abilities/deficits in clinical samples. Emotion dysregulation, or the inability to regulate one's emotions in a healthy or adaptive way has been linked to destructive behaviours such as binge eating and drinking, gambling, and self-injury (Klonsky, 2008). As most research regarding emotion regulation is self-report it is critical to address if these emotion regulation deficits are also present at the physiological level, in order to better develop and assess treatment strategies that improve emotion regulation cognitively, behaviourally, and physically. However, due to inconsistencies in the research and insufficient data to support the expected hypotheses, more research needs to be done using the startle paradigm on positive and negative emotion regulation, with large sample sizes, to properly understand the mechanisms at work that influence the startle response before it can be used to provide reliable information about clinical samples.

Future studies on the emotion regulated startle response should utilize a larger sample size, balanced paradigms for picture valence, and matched subjective and normative arousal for positive and negative stimuli in order to successfully extend upon current knowledge on the nature of the startle response to emotion regulation. It may also be beneficial to look for more effective ways to elicit positive emotion in the lab setting, beyond the IAPS picture set. Moreover, fear conditioning has been shown to modulate the startle response in the lab (Greenwald et al., 2007; Lissek et al., 2007); perhaps associating a stimulus with a positive experience or reward could also be used to modulate the startle response, eliciting a more genuine positive emotion in the lab. In conclusion, the startle blink paradigm is a promising physiological measure of emotion regulation, however more research needs to be conducted with larger samples before it can be used to assess dysregulation in clinical populations.

Declaration of Conflicting Interests
The author declared they have no conflicts of interests with respect to their authorship or the publication of this article.

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