



Startle modulation by affective faces

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ABSTRACT

Startle reflex modulation by affective pictures is a well-established effect in human emotion research. However, much less is known about startle modulation by affective faces, despite the growing evidence that facial expressions robustly activate emotion-related brain circuits. In this study, acoustic startle probes were administered to 37 young adult participants (20 women) during the viewing of slides from the Pictures of Facial Affect set including neutral, happy, angry, and fearful faces. The effect of expression valence (happy, neutral, and negative) on startle magnitude was highly significant ($p < .001$). Startle reflex was strongly potentiated by negative expressions (fearful and angry), however, no attenuation by happy faces was observed. A significant valence by gender interaction suggests stronger startle potentiation effects in females. These results demonstrate that affective facial expressions can produce significant modulation of the startle reflex.

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1. Introduction

Startle reflex modulation by an affective foreground is a well-established experimental phenomenon in human emotion research. Acoustic startle reflex is an automatic, obligatory defensive response triggered by abrupt and loud noise. Studies of animals (Davis, 1989) and humans (Grillon, 2002) have shown that the startle reflex is potentiated in the presence of a conditioned fear stimulus. Converging evidence suggests that the intensity of the startle reflex depends on the ongoing motivational and affective state of the subject, such that the reflex is facilitated by aversive/defensive motivational states and attenuated by appetitive states (Grillon and Baas, 2003; Lang et al., 1998; Vrana et al., 1988).

The neurobiological substrates of startle modulation by emotion have been extensively studied in animals using fear-potentiated startle paradigms (reviewed in Koch and Schnitzler, 1997). These studies showed that the primary acoustic startle pathway in the brain stem is modulated through direct projections by the secondary pathway, in which the amygdala plays the central role. Thus, the degree of startle reflex modulation by various stimuli can serve as an objective measure of the extent to which particular stimuli activate (or suppress) the neural circuitry underlying the two basic motivational systems, aversive and appetitive, and two emotional states, pleasant and unpleasant (Koch and Schnitzler, 1997; Lang et al., 1998).

Human startle experiments have largely relied on affective pictures as stimulus material for emotion induction, particularly images from the International Affective Picture System (IAPS) (Lang et al., 1999). The IAPS pictures have been shown to produce a robust modulation of the startle reflex, such that reflex magnitude was the largest during unpleasant, intermediate during neutral, and smallest during pleasant pictures (Cuthbert et al., 1996; Lang et al., 1998; Vrana et al., 1988). This valence effect has been replicated using other kinds of stimuli such as emotional sounds (Bradley and Lang, 2000) as well as conditioned stimuli in fear conditioning paradigms (Grillon, 2002). Individual differences in affective modulation of startle have been associated with personality characteristics and psychopathology (Corr et al., 1996; Grillon and Baas, 2003; Vaidyanathan et al., 2009). In contrast to the extant literature on startle modulation by affective pictures, affective facial expressions have been little used in startle experiments, although several important features of facial images such as structural homogeneity and uniform perceptual complexity over a range of emotional expression, as well as low novelty can provide a more rigorous control for picture characteristics that are unrelated to emotional content.

The neural mechanisms of face perception are being increasingly understood (reviewed in Haxby et al., 2002; Posamentier and Abdi, 2003). Studies of amygdala lesions (Adolphs et al., 2005, 1995) and direct electrical stimulation of the amygdala suggest that this structure plays an important role in the processing of facial emotional expressions by humans and non-human primates. Neuroimaging studies have demonstrated a robust activation of the amygdala by emotional facial expressions, especially by fearful faces (Breiter et al., 1996; Morris et al., 1996). Two functional neuroimaging studies directly compared brain activation patterns

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induced by facial expressions and affective pictures from the IAPS. Hariri et al. (2002) found that fearful and angry facial expressions produce a significantly stronger response in the amygdala compared to the IAPS pictures. Moreover, facial stimuli also produced a greater autonomic (skin conductance) response than affective pictures in the same study. Another study found that both affective faces and IAPS pictures recruit similar brain regions but also noted a greater activation of some regions by affective faces (Britton et al., 2006).

It should be noted, however, that the relation between threatening facial expressions and amygdala activation is not universal: amygdala activation has also been reported for other facial expressions and by faces in general (Breiter et al., 1996), and reduced amygdala responses were observed in tasks requiring explicit emotion recognition, in contrast to increased amygdala activation in tasks involving implicit processing of facial expressions (Critchley et al., 2000). These exceptions notwithstanding, available evidence indicates that affective facial expressions, particularly fearful expressions produce a robust activation of the amygdala during passive viewing (i.e., in the absence of explicit processing demands).

The few studies that have examined the effects of the valence of facial expressions on startle modulation have reported mixed results. In a study by Balaban (1995), a startle probe was administered while 5-month-old infants were shown photographic slides of unfamiliar adult faces with happy, neutral, and angry expressions. There was a linear relationship between startle response magnitude and slide valence: the response was augmented during exposure to the angry faces and was reduced during exposure to the happy faces relative to neutral faces. However, a study of 4–8-year-old children did not find differences in startle responses during viewing of angry and neutral faces (Waters et al., 2008). The few studies that used adult samples have also provided mixed findings. One abstract (Alpers and Adolph, 2006) reported no effect of expression valence (i.e., angry and happy vs. neutral) on startle modulation. In another study, pictures of smiling and crying infants failed to produce startle modulation in young adults (Spangler et al., 2001). A recent study by Hess et al. (2007) in which happy, neutral, and angry faces were administered to a group of young adults, also produced mixed results. No main effect of facial expression was found, but there was a significant interaction between expression and the actor's sex. However, possible effects of viewer's gender were not reported.

We are aware of only one published startle reflex study in which both angry and fearful faces were administered (Springer et al., 2007). In one experiment, the authors found a significant effect of facial expression on startle magnitude, where angry but not fearful faces produced an increased eyeblink response compared to all other expressions. A replication experiment using the same paradigm with different facial material failed to show a significant main effect of facial expression on startle, but in pairwise comparisons, angry faces still showed significant differences from other expressions. It should be noted, however, that the startle stimuli were administered on every trial and thus were fully predictable to the subjects, which is not typical for startle modulation studies (Springer et al., 2007).

Taken together, the available evidence does not seem to support the notion that affective facial expression can modulate the startle response in adults as consistently as affective pictures. However, it is important to note that only one study included faces with fearful expression.

The goal of this study was to examine startle reflex modulation by affective faces in a community-based sample of young adults. We hypothesized that affective facial expression would influence the startle magnitude in the direction predicted by the theory of motivation and emotion proposed by Lang et al. (1993), i.e., that the

startle response will be potentiated by expressions with negative emotional valence (fearful and angry) and attenuated by positively valenced (happy) faces. In addition, we intended to examine possible effects of the observer's sex on startle modulation by affective faces.

2. Methods

2.1. Participants

Thirty-nine individuals including 18 men (18–22 years; M age \pm SD: 19.4 ± 1.2 years) and 21 woman (18–21 years; M age \pm SD: 19.0 ± 1.3 years) participated in the study. Participants were recruited through state birth records as part of a larger population-based epidemiological study of twins and families and were included in the present study after screening for exclusion criteria. The criteria included a history of head trauma with loss of consciousness for more than 5 min, known history of epilepsy, currently taking a psychoactive medication, as well as hearing, visual and other physical and mental impairments that could prevent the participants from understanding and following the experimental instructions. Apart from these exclusion criteria, participants were not selected, and the sample is thus well representative of the general population. The study was approved by Washington University Institutional Review Board, and the subjects provided written informed consent.

2.2. Stimuli and procedure

The participants were administered photographs of faces from Ekman's and Friesen's Pictures of Facial Affect set (Ekman, 1976) depicting basic emotional expressions. The procedure was kept very close to procedures commonly used in previous studies employing affective pictures as stimulus material. Each slide was presented on a computer monitor for 6 s, with 12–24 s (average 15 s) intervals between pictures. A total of 55 images were presented, including 18 happy faces, 19 neutral faces, and 18 faces with negative emotional expression (9 angry and 9 fearful). Faces with positive, neutral, and negative emotional expression were presented in a fixed pseudorandom order. Each image consisted of a black-and-white face oval on a black background; the dimensions were 20 cm \times 14 cm, i.e., close to a life-size. The monitor was placed at 110 cm in front of the subject's face; thus the visual angular dimensions of the image were $10.42^\circ \times 7.29^\circ$. A fixation cross was presented in the center of the screen during the inter-picture intervals. Auditory stimuli were administered through calibrated foam insert earphones (Etymotic Research). A 70 dB white noise background was present throughout the experiment. The startle stimuli were 105 dB, 40 ms white noise bursts with near instantaneous rise time. Startle stimuli were administered during two-thirds of the pictures at 3, 4, or 5 s after the picture onset. In addition, 10 startle stimuli were presented during inter-picture intervals (blank screen with a fixation cross). As in previous studies using affective pictures, this presentation schedule was used to minimize the predictability of the startle stimuli.

The first startle stimulus was presented during a neutral face picture and was not scored. Of the remaining 36 startle stimuli presented during the viewing of faces, 12 were administered during neutral faces (5 male and 7 female), 12 during happy faces (6 male and 6 female), and 12 during emotionally negative faces (6 angry and 6 fearful, including 8 male and 4 female faces). The average serial position of the positive, neutral, and negative expressions was 25.7, 23.7, and 22.2 (differences were non-significant: $F(2,34) = .31$, $p = .74$; all pairwise comparisons: $p > .8$).

2.3. Startle EMG recording and quantification

Electromyographic (EMG) activity was recorded from two miniature Ag/AgCl electrodes placed 1 cm apart over the orbicularis oculi muscle beneath the left eye. The EMG data were digitized online with 1000 Hz sampling rate using a Synamps amplifier and were analyzed off-line using Scan 4.3 software (Compumedics-Neuroscan). The EMG recordings were visually inspected, and trials were removed from the analysis if the startle stimulus overlapped with spontaneous eye blinks or there was excessive baseline EMG activity in the startle channel. This resulted in the exclusion of 11.8% of trials on the average. Quantification of the startle response magnitude included bandpass filtering (10–200 Hz), signal rectification, smoothing over 5 adjacent data points with 3 consecutive passes, baseline correction using a 70 ms baseline (from 50 ms before to 20 ms after the stimulus onset) and detection of the peak value in the time window 20–120 ms after the stimulus onset. Startle blink magnitude in individual trials was measured as the peak magnitude relative to the baseline. Two participants were excluded due to noisy baseline EMG recording and/or lack of distinct responses to the startle stimuli (less than $1 \mu\text{V}$ above baseline).

Individual startle trials were sorted into three emotional valence categories according to the foreground picture (happy, neutral, or negative emotional expression) with equal number of trials ($n = 12$) in each category. Angry and fearful expressions were collapsed into a single "negative expression" category. Next, startle responses in individual trials were averaged separately within each facial expression category. Kolmogorov–Smirnov tests indicated that distributions of the average startle response magnitudes from each expression category did not depart significantly from normality.

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