

## BRIEF REPORT

# Modulation of the initial light reflex during affective picture viewing

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### Abstract

An initial reflexive constriction of the pupil to stimulation—the light reflex—is primarily modulated by brightness, but is attenuated when participants are under threat of shock (i.e., fear-inhibited light reflex). The present study assessed whether the light reflex is similarly attenuated when viewing emotional pictures. Pupil diameter was recorded while participants viewed erotic, violent, and neutral scenes that were matched in brightness; scrambled versions identical in brightness were also presented as an additional control. Compared to viewing neutral scenes, the light reflex was reliably modulated by hedonic content, with significant attenuation both when viewing unpleasant as well as pleasant pictures. No differences in the light reflex were found among scrambled versions. Thus, emotional modulation of the initial light reflex is not confined to a context of fear and is not indicative of brightness differences when viewing pictures of natural scenes.

**Descriptors:** Emotion, Pupil diameter, Light reflex, Pictures

The initial light reflex is an early occurring, parasympathetically mediated constriction of the pupil that occurs primarily in response to changes in brightness, but is also modulated during aversive anticipation. When a cue signals threat of shock, the reflexive constriction to a flash of light presented during the anticipatory period is attenuated compared to safe periods, eliciting a “fear-inhibited light reflex” (Bitsios, Szabadi, & Bradshaw, 1996, 2004). In a previous study (Bradley, Miccoli, Escrig, & Lang, 2008), we found greater *late* pupil dilation when viewing emotionally arousing compared to neutral pictures, and follow-up analyses suggested that specific highly arousing contents, whether unpleasant (e.g., violence) or pleasant (e.g., erotica), may have modulated the amplitude of the initial light reflex. Differences in brightness, however, could have mediated this effect. Thus, in the current study, we examined this issue more closely by presenting highly arousing pictures of erotica and violence that were matched exactly in brightness to each other, as well as to a set of neutral pictures. If the initial light reflex is generally attenuated during emotional picture viewing, it would suggest that modulation of this early pupil reaction is not restricted solely to aversive stimulation.

Pictures were presented in grayscale, and brightness was exactly matched for erotic, violent, and neutral content. As an

additional control for assessing effects of brightness on the initial light reflex, each picture was presented in both an intact and scrambled version. In the scrambled version, pixels were randomly shuffled such that brightness was identical to the intact version but no content (semantic or emotional) remained. If emotion modulates the initial light reflex, we expected that viewing emotional scenes—whether erotic or violent—would elicit a smaller light reflex compared to when viewing neutral scenes, but that the light reflex for scrambled pictures, identical in brightness to the intact versions, would not vary as a function of original picture content.

### Method

#### Participants

Twenty-seven 18- to 21-year-old University of Florida General Psychology students (13 male, 82% Caucasian) signed a consent form and participated for course credit.

#### Materials and Design

Stimuli were 36 pictures<sup>1</sup> selected from the International Affective Picture System (IAPS: Lang, Bradley, & Cuthbert, 2008), consisting of 12 erotic (mean pleasure/arousal = 6.6, 6.4), 12 neutral (mean pleasure/arousal = 5.2, 3.6), and 12 violent (mean pleasure/

Preliminary results from this research study were presented at the 51st annual meeting of the Society for Psychophysiological Research in Boston, MA, in 2011. This work was supported in part by NIMH grants MH098078 and MH094386 to Peter J. Lang.

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1. The IAPS pictures (Lang et al., 2008) used in this study were: Erotic: 4597, 4643, 4647, 4658, 4668, 4676, 4687, 4689, 4690, 4693, 4697, 4698; Neutral: 2026, 2101, 2191, 2211, 2308, 2359, 2372, 2411, 2441, 2513, 2595, 2850; Violence: 3030, 3059, 3064, 3068, 3103, 3131, 3150, 3225, 3530, 6560, 9163, 9433.

arousal = 1.9, 6.4) pictures. All pictures portrayed people, were landscape in orientation, and displayed in 256-bit grayscale. Contrast and figure-ground ratings were matched across the three contents. The 36 pictures were divided into 12 sets of 3 pictures, with each set containing one erotic, one neutral, and one violent picture of identical brightness. Brightness naturally varied from low to high across the 12 sets. A scrambled version of each picture was then created by randomly rearranging pixels to produce a same brightness version of the original intact picture. The final set of stimuli consisted of 36 intact and 36 scrambled pictures, for a total of 72 pictures.

Each picture was displayed for a 6-s free-viewing period, followed by a varying intertrial interval of 9–12 s that presented a fixation cross on a gray screen. Picture presentation was counter-balanced such that an intact and scrambled version of each hedonic content (e.g., erotic, neutral, violence) was presented within a block of six trials. Across participants, three presentation orders were constructed, which counterbalanced whether a specific picture was presented early, middle, or late in the series.

### Apparatus

Pictures were presented using an IBM-compatible computer running Presentation software (Neurobehavioral Systems, San Francisco, CA). Pictures were displayed on a 19-inch monitor located in the experiment room, at a distance of 30 inches (76.2 cm) from where the participant was seated, subtending  $8.9 \times 9.1$  degrees of visual angle.

Pupil diameter was continuously sampled at 60 Hz from 2 s before picture onset to 250 ms before the end of the intertrial interval using an ASL model D6 desk-mounted remote eye tracker (Applied Science Laboratories, Bedford, MA). This system consists of a video camera and an infrared light source, which is focused on the participant's right eye. Face recognition is used to track head movements and keep the pupil in focus. The recording video camera was located in front of the participant, situated just below the stimulus presentation monitor.

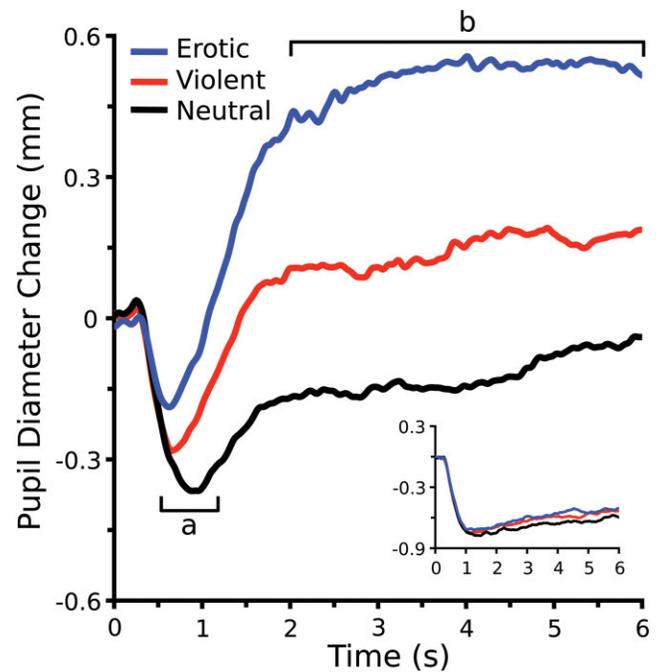
### Procedure

After arriving at the laboratory, each participant signed a consent form and was seated in an upright chair in a small, sound-attenuated, dimly lit room. A calibration procedure was conducted in which the participant was instructed to sequentially look at nine dots that appeared one at a time on the screen. Several scrambled images of varying brightness were presented in order to ensure reliable pupil discrimination at different levels of picture brightness. The participant was then instructed to view each picture for the entire time that it was on the screen and to look at the fixation cross at all other times.

### Data Reduction

Pupil diameter was converted offline from arbitrary units to millimeters, and linear interpolation was used to estimate pupil size for samples in which the pupil was occluded due to blinking. For each trial, pupil diameter during a 1-s baseline prior to picture viewing was subtracted from each of the following pupil samples. Five participants were removed from the final analysis due to unsuccessful pupil discrimination on more than 15% of trials.

Based on the average waveform, the initial light reflex was calculated as the mean pupil change from 0.5 to 1.3 s after picture



**Figure 1.** Change (mm) in pupil diameter from a 1-s baseline preceding picture onset when viewing erotic, neutral, or violent scenes. The initial light reflex (a) was averaged in a window from 0.5 to 1.3 s following picture onset and late pupil diameter (b) was averaged in a window from 2 to 6 s post picture onset. Inset: For scrambled pictures, pupil diameter did not differ as a function of original picture content.

onset (Figure 1a). Trials in which pupil discrimination was less than 50% accurate in this time window were counted as missing (3.2% of total trials), resulting in an average of 11.3 to 12.0 trials remaining for each condition.<sup>2</sup> Late pupil diameter was calculated as the mean pupil diameter change from 2 to 6 s following picture onset (Figure 1b) and included the same trials as used in the analysis of the initial light reflex.

Data were analyzed using a Content (3: erotic, neutral, violent)  $\times$  Version (2: intact, scrambled) repeated measures analysis of variance for each of the dependent variables (initial light reflex, late pupil diameter). Greenhouse-Geisser was used to correct degrees of freedom for sphericity. Means and standard deviations for all comparisons are displayed in Table 1.

## Results

### Light Reflex

Figure 1 illustrates the change in pupil diameter during free viewing of natural scenes, which indicates that the initial light reflex was clearly attenuated when viewing intact erotic and violent compared to neutral scenes. Supporting this were significant main effects of content,  $F(2,20) = 23.5$ ,  $p < .001$ , version,

2. Because some trials were not included in the analysis, it is possible that brightness could vary among trials (pictures) retained in the final analysis. To confirm that these findings do not reflect subtle differences in brightness resulting from missing trials, a secondary analysis was conducted using only those trials in which all data were available for all conditions at each brightness level. Results were identical to those reported here.

**Table 1.** Means (Standard Errors) of the Change in Pupil Diameter (mm) for All Conditions

Hedonic content	Intact pictures		Scrambled versions	
	Initial light reflex	Late pupil diameter	Initial light reflex	Late pupil diameter
Erotic	-.07 (.04)	.52 (.07)	-.61 (.04)	-.57 (.07)
Neutral	-.32 (.04)	-.12 (.06)	-.64 (.05)	-.67 (.06)
Violent	-.20 (.06)	.15 (.08)	-.63 (.05)	-.60 (.05)

Note. Initial light reflex (.5 to 1.3 s); late pupil diameter (2 to 6 s).

$F(1,21) = 179.9$ ,  $p < .001$ , and their interaction,  $F(2,20) = 13.4$ ,  $p < .001$ . Follow-up analyses indicated a significant effect of hedonic content when viewing intact,  $F(2,20) = 35.1$ ,  $p < .001$ , but not scrambled pictures. For intact pictures, viewing either erotic or violent scenes elicited significantly less early pupil constriction compared to neutral scenes,  $F(1,21) = 107.7$ ,  $p < .001$ , and  $F(1,21) = 14.5$ ,  $p < .005$ , respectively, and erotic scenes prompted less constriction than violent scenes,  $F(1,21) = 15.4$ ,  $p < .001$ .

Early modulation of the initial light reflex by emotional content was not due to differences in brightness, since the light reflex did not differ among scrambled versions of the same pictures, which were identical in brightness to the original intact scenes ( $F < 1$ ; see inset, Figure 1). Overall, pupil constriction was larger when viewing scrambled compared to intact pictures (all  $F_s(1,21) > 58$ ,  $p < .001$ ).

**Picture analysis.** To further determine whether the initial light reflex is reliably modulated by emotion during picture viewing, a hierarchical regression was conducted using each intact picture as the unit of analysis, first assessing effects of brightness and then effects of emotionality. As expected, light reflex amplitude was highly influenced by brightness,  $F(1,34) = 106.6$ ,  $p < .001$ ,  $R^2 = .75$ , with pictures higher in brightness prompting larger light reflexes than lower brightness pictures. After removing effects due to brightness, however, ratings of emotionality (i.e., arousal; Lang et al., 2008) continued to account for significant variance in the amplitude of the initial light reflex,  $F(2,33) = 11.3$ ,  $p < .001$ ,  $R^2 = .06$ .

### Late Pupil Diameter

Pupil diameter later in the viewing interval was also modulated by hedonic content, with larger changes when participants viewed emotional compared to neutral scenes (Figure 1b). Significant main effects were obtained for content,  $F(2,20) = 55.3$ ,  $p < .001$ , version,  $F(1,21) = 169.1$ ,  $p < .001$ , and their interaction,  $F(2,20) = 28.7$ ,  $p < .001$ . Follow-up analyses indicated a significant effect of picture content for intact pictures only,  $F(2,20) = 72.0$ ,  $p < .001$ , in which viewing pictures of erotica or violence prompted significantly larger increases in late pupil diameter compared to neutral scenes,  $F(1,21) = 200.6$ ,  $p < .001$ , and  $F(1,21) = 19.1$ ,  $p < .001$ , respectively, and erotic scenes elicited larger changes than violent scenes,  $F(1,21) = 49.3$ ,  $p < .001$ . Overall, late pupil diameter was smaller when participants viewed scrambled compared to intact versions (all  $F_s(1,21) > 70$ ,  $p < .001$ ), but did not differ among scrambled versions of each content.

### Discussion

During picture viewing, emotional modulation of pupil diameter is apparent as early as the initial light reflex. Although this reflex is

strongly affected by brightness, initial pupil constriction was nonetheless significantly attenuated for emotional compared to neutral images, in both the subject and picture analyses. When the same pictures were scrambled to produce versions that were identical in brightness to the intact pictures but lacking any content, there was no difference in the amplitude of the initial light reflex or late pupil diameter as a function of original picture content, confirming that the differences found when viewing intact pictures did not simply reflect subtle differences in brightness.

Previous studies have reported that simply anticipating an aversive event, such as the possible presentation of an electric shock, attenuates initial pupil constriction (i.e., fear-inhibited light reflex; Bitsios et al., 1996, 2004). In these studies, early pupil constriction was attenuated under threat compared to safety, and, importantly, did not show the same attenuation when participants anticipated the presentation of a weak acoustic stimulus. On the other hand, effects of highly pleasant stimulation on early constriction were not tested, and here we found greatly attenuated light reflexes when pictures of erotica were presented. At the least, the current data suggest that modulation of the initial light reflex is not solely confined to a context of fear during free viewing of pictures of natural scenes.

Although changes in pupil diameter are determined by both sympathetic and parasympathetic influences on the sphincter and dilator muscles of the iris (Steinhauer, Siegle, Condray, & Pless, 2004), initial pupil constriction is thought to be mediated by parasympathetic projections from the Edinger-Westphal nucleus to the sphincter muscle. Consistent with this, administration of dapiprazole (a peripheral sympathetic blockade) does not alter the fear-inhibited light reflex (Giakoumaki, Hourdaki, Grinakis, Theou, & Bitsios, 2005).

One interpretation is that variations in pupil diameter during affective picture viewing might be related to locus coeruleus activity. In animals, phasic firing of neurons in the locus coeruleus is associated with increases in pupil size, and both locus coeruleus activity and pupil diameter increase when rare or unexpected stimuli occur (Rajkowski, Kubiak, & Aston-Jones, 1994), consistent with modulation by motivationally or behaviorally relevant stimuli. Because the locus coeruleus has inhibitory connections to the Edinger-Westphal nucleus and excitatory projections to preganglionic sympathetic neurons, it could attenuate the parasympathetically mediated light reflex as well as contribute to later sympathetically mediated changes in pupil diameter (Szabadi, 2012). Future studies utilizing additional pharmacological blockades of appropriate receptors in the dilator and sphincter muscles (e.g., Steinhauer et al., 2004) would be useful in further understanding how the pupil is modulated by emotion.

Modulation of pupil diameter was largest when participants viewed erotic scenes, which is consistent with studies finding the largest late positive potentials (Schupp et al., 2004) and electrodermal reactions (e.g., Lang, Bradley, & Cuthbert, 1997)

when young adults view pictures of erotica, compared to other contents rated as emotionally arousing. An additional finding is that scrambled pictures prompted a larger light reflex than intact pictures. This is probably due to the higher spatial frequency of scrambled versions, which can increase pupil constriction (Link et al., 2006).

### Summary

Viewing emotionally arousing pictures prompted modulation of pupil diameter beginning with the initial light reflex and persisting throughout the viewing interval, suggesting both rapid and sus-

tained effects of emotional arousal on pupil size during free viewing of affective pictures. These findings cannot be explained by subtle differences in picture brightness, as scrambled versions of the same pictures, identical in brightness to the intact versions, did not elicit reliable differences in pupil diameter. Future studies are needed to determine the relative contribution of activity in the sympathetic and parasympathetic branches of the autonomic nervous system on pupil diameter during emotional picture viewing. Most broadly, these data suggest that modulation of the initial light reflex is not confined to a context of fear, and is not indicative of a failure to control brightness when presenting pictures of natural scenes.

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(RECEIVED November 22, 2013; ACCEPTED April 8, 2014)