Non-conscious modulation of cardiac defense by masked phobic pictures

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Abstract

The present study investigated the modulation of cardiac defense by presenting emotional pictures under both effective and non-effective masking procedures. The aim was to test Öhman’s model of pre-attentive processing of fear. Participants were 48 women volunteers with intense fear of spiders. The stimulus to elicit cardiac defense was a white noise of 105 dB, 500 ms duration and instantaneous risetime. Subjects had two trials of picture–noise presentation—one with a picture of a spider and one with a picture of a flower—, either under an effective masking procedure (30 ms duration) or a non-effective masking procedure (500 ms duration). Order of presentation was counterbalanced. Dependent variables were heart rate and subjective assessment of the noise. Results showed an increased cardiac response in the first trial and a less reduced cardiac response in the second trial when the noise was preceded by the phobic picture under both masking procedures. The response was accompanied by an increase in the subjective unpleasantness of the noise. These results provide support to Öhman’s theoretical model.

Keywords: Cardiac defense; Heart rate; Unconsciousness; Backward masking; Emotional modulation; Fear potentiation

1. Introduction

Studies of emotional modulation of the startle reflex using the picture viewing paradigm (Lang, 1995) have consistently reported larger amplitude to unpleasant and smaller amplitude to pleasant pictures, compared to neutral ones (Bradley, 2000). Lang and colleagues have proposed a motivational priming model to explain this effect: perceptual stimuli that engage the aversive motivational system potentiate...
defensive reflexes whereas perceptual stimuli that engage the appetitive motivational system inhibit defensive reflexes (Lang et al., 1997). The motivational priming model has been extensively investigated with regards to the eyeblink component of the startle reflex and with regards to a large variety of perceptual affective foregrounds: pictures (Bradley, 2000), films (Jansen and Fridja, 1994), sounds (Bradley and Lang, 2000) and odors (Miltner, 1994). Although the same effect is expected with regards to other defensive reflexes, such as cardiac defense, few studies have been reported concerning affective modulation of protective responses different from eyeblink startle (Cook and Turpin, 1997; Sánchez et al., 2002).

Two recent studies have applied Lang’s picture viewing paradigm to examine the emotional modulation of cardiac defense (Sánchez, 2000; Sánchez et al., 2002), a response to intense auditory stimulation characterized by a sequence of heart rate changes, observable within the 80 s after stimulus onset, with two accelerative and two decelerative components in alternating order (Vila et al., 1992; Vila, 2002). The results of both studies confirmed the expected hypothesis from the motivational priming model: The accelerative components of the response were potentiated when participants were visualizing unpleasant (first study) and phobic (second study) pictures, as compared to neutral and pleasant ones. In addition, a profound modification of the pattern of the potentiated response was observed: the first deceleration disappeared and the two accelerations merged showing a single larger acceleration.

The finding of similar affective modulation of two protective responses–eyeblink startle and cardiac defense–gives further support to Lang’ motivational priming model. Moreover, it suggests that the neural circuitry underlying the affective modulation of eyeblink startle–activation of the central nucleus of the amygdala acting on the primary spinal circuit of the startle reflex (Davis, 1997)–acts also on the autonomic network mediating cardiac defense (Lang et al., 2002; Thayer and Siegle, 2002). A further implication concerns the topographic change of the cardiac defense response when the intense acoustic stimulus is preceded by unpleasant or phobic pictures. It has been suggested (Vila et al., 2003) that the disappearance of the deceleration between the two accelerations reflects the transfer of the averesively motivated attentional factors induced by the primed noise to the preceding stimulus, whereas the mixture of the two accelerations into a single one reflects the advancement in time of the preparatory protective actions in order to better match successful adaptation.

The involvement of attentional factors in defense has been emphasized by Öhman and colleagues in their studies on pre-attentional processing of fear (Öhman, 2000). Using the backward masking technique, introduced by Marcel (1983) in his studies of semantic priming, in combination with the skin conductance response, they have conducted a large series of studies to demonstrate that fear relevant stimuli (pictures of snakes, spiders and fear–angry faces), as compared to irrelevant stimuli (pictures of flowers, mushrooms and neutral–happy faces), can be pre-attentively processed (Öhman and Soares, 1993, 1994; Esteves et al., 1994a,b; Wong et al., 1994; Öhman and Soares, 1998; Katkin et al., 2001). Additional support for Öhman’s view comes from electromyographic and neuroimaging studies of unconscious processing of masked facial expressions (Morris et al., 1998; Whalen et al., 1998; Dimberg, 2000; Dimberg et al., 2000). Pre-attentive processing of other types of visual stimuli, such as alcohol pictures, have also been reported (Ingjaldsson et al., 2003a,b). However, in spite of the large series of studies reporting non-conscious processing of fearful stimuli, up to now no positive results have been reported using as the dependent variable specific measures of defense, such as eyeblink startle or cardiac defense. Two preliminary reports on emotional modulation of eyeblink startle by viewing masked phobic pictures failed to show the expected modulation (Merckelbach et al., 1995; Globisch et al., 1996). As regards cardiac defense, no study has yet been reported.

The aim of the present study was to test whether the emotional modulation of cardiac defense, observed under non-masked conditions using the picture viewing paradigm (Sánchez et al., 2002), can also be observed under masked conditions using Öhman’s backward masking procedure. Evidence of such a modulation would provide additional support to both Lang’s motivational priming model and
Öhman’s model of pre-attentive processing of fear. Moreover, since it is held that skin conductance does not differentiate between orienting and defense (Graham, 1979) or, in more general terms, between attention and emotion (Öhman, 1997), we can expect that heart rate data will help to clarify whether the pre-attentive processing of fear stimuli, indexed in Öhman’s studies by the skin conductance response, represents just an orienting response, a defense response or both.

2. Method

2.1. Participants

The participants in the study were 48 women volunteers. Their mean age was 19.83 (standard deviation=1.85, range between 18 and 27 years old). All were students of psychology at the University of Granada, with intense fear of spiders. They were selected from a group of 332 students according to their scores on the Spider Questionnaire (SPQ) (Klorman et al., 1974). This questionnaire is formed by 31 fear of spider related items with scores ranging from 0 to 31. In Klorman et al.’s psychometric study, internal consistency is reported to range from 0.83 to 0.90. They also report discriminant validity and descriptive statistics for the SPQ on a sample of 122 female students from the University of Wisconsin. In their sample, a score of 14 corresponded to percentile 75. All subjects of our study had a score on the SPQ equal to or higher than 15, corresponding to percentile 86 of our student pool. None of the participants had auditory or visual deficits or cardiovascular problems and were not under pharmacological treatment.

2.2. Design

We used a mixed factorial design 2(×2), with the first between group factor consciousness, with two levels (conscious group —non-effective masking and non-conscious group —effective masking), and the second repeated measure factor picture content, with two levels (phobic picture and non-phobic picture). All subjects saw both pictures, counterbalancing the order of presentation within each group, so that half of the subjects viewed the phobic picture first and then the non-phobic one, and the other half viewed the pictures in the opposite order. For the statistical analysis, the order of picture presentation (spider/flower versus flower/spider) was considered, the final design being a 2 (consciousness)×2 (order) (×2 (picture)) design.

2.3. Psychophysiological test

A picture of a spider (number 1220) and a picture of a flower (number 5030), both selected from the International Affective Picture System (Lang et al., 1988) were used as stimuli. The mask, which was always the same, was a picture made from randomly reassembled small cut pieces of both pictures. The acoustic stimulus used to elicit the cardiac defense response was a white noise of 105 dB, 500 ms duration and instantaneous risetime. The sequence of the test, without interruption, was as follows: (a) a 10 min rest period, (b) two runs of picture–noise presentation with an intertrial interval of 120 s and (c) a final rest period of 120 s. Each run was initiated without any warning signal and consisted of a sub-sequence of 6 picture–mask presentations, 1 each second, for 6 s. The duration of the mask was always 100 ms, with the duration of the picture variable depending on the experimental condition (500 ms in the conscious condition and 30 ms in the non-conscious condition). The acoustic stimulus always appeared 3.5 s after the beginning of the run, coinciding with the onset of the fourth mask presentation (see Fig. 1). The pictures were projected on the wall in front of the subject, at distance of 3 m, occupying a rectangle of 143×94 cm. The dependent variable was beat-to-beat heart rate recorded using an electrocardiograph (EKG) and simultaneous digital R–R interval transformation in milliseconds using the VPM software program (Cook, 1997).

2.4. Apparatus

The following equipment was used: (a) a Grass polygraph with a 7P4 preamplifier to record the electrocardiogram at lead II; (b) a Coulbourn audio system (modules S81-02, S84-04, S82-24 and S22-18) to generate the white noise and present it.
binaurally through earphones (Telephonic TDH Model-49), the intensity of the sound being calibrated with a sonometer (Bruek and Kjaer, model 2235) and an artificial ear (Bruek and Kjaer, model 4153); (c) two Kodak Ektapro 9000 slide projectors, with computer shutter control, one to present the picture and the other to present the mask; and (d) an Advantech card (model PCL812PG) with a 12 bit analog-to-digital converter and with digital input–output functions, run by a Pentium 2 computer, to control the experimental session through the VPM program (Cook, 1997).

2.5. Dependent variables

2.5.1. Heart rate

2.5.1.1. Cardiac defense response. Weighted averaged second by second heart rate was measured during the 80 s following onset of each auditory stimulus and was expressed in terms of difference scores with respect to a baseline level (15 s before each trial). To facilitate the statistical analysis, we followed the same procedure used in previous studies: the 80 values were reduced to 10 medians of 10 progressively longer intervals: two of 3 s, two of 5 s, three of 7 s and three of 13 s (Vila et al., 1992).

2.5.1.2. Heart rate response to the pictures. Weighted average heart rate was measured each half second during the 3.5 s preceding the defense noise and expressed as difference scores with respect to the baseline level (15 s before the trial).

2.5.2. Self-report measures

In addition to the SPQ for subject selection, two questionnaires were used: a recognition questionnaire and a questionnaire of subjective reactivity to the noise.

2.5.2.1. Recognition questionnaire. This questionnaire was developed for the present study based on similar procedures used in backward masking studies. It had two parts: a first part included an open-ended question about the identity of the visual stimuli presented and a second part consisting of a forced-choice test. In the forced-choice test, the subject was shown 12 printed pictures from the International Affective Picture System (4 spiders, 4 flowers and 4 snakes), including the two pictures used in the psychophysiological test. The subjects had to indicate the two pictures that were actually presented during the experimental session and to state their confidence in their guess using a scale from 0 to 9 (0 meaning not at all sure and 9 meaning completely sure).

2.5.2.2. Questionnaire of subjective reactivity to the noise. In this questionnaire, subjects rated separately the intensity and unpleasantness of the first noise presented using a scale from 0 to 9, 0 meaning not at all intense or unpleasant and 9 extremely intense or unpleasant.
2.6. Procedure

Each participant attended a single laboratory session divided into three stages:

(1) Pre-experimental stage: The participant received information about the experimental session and signed the informed consent form. They were informed that the purpose of the study was to record physiological data while they were trying to relax during several minutes followed by the presentation of visual pictures and noises. No specific information was given about the content of the pictures nor the intensity of the noises. Then, an interview was conducted in order to ascertain age, visual or auditory deficits, health and pharmacological treatment. After the specific instructions were read, the electrodes and earphones were attached, the physiological recording checked, and the participant left alone in a semi-darkened room. Participants were asked to breathe normally during the test and maintain their eyes open looking at a fixation point located at a distance of 3 m in front of the participant.

(2) Experimental stage: The participant did the psychophysiological test following the sequence described above. During this phase, the experimenter was in the adjoining room controlling the correct functioning of the equipment. The rooms were audiovisually connected via video camera.

(3) Post-experimental stage: After removing the earphones, the participants completed the Recognition Questionnaire and the Subjective Reactivity Questionnaire. Then the electrodes were removed and the participant given the academic credit for her participation.

2.6.1. Statistical analysis

Analysis of variance for repeated measures was applied to the heart rate data with two between group factors (consciousness and order) and two repeated measures factors (picture and time). Consciousness, order and picture had two levels each. Time had 10 levels for the cardiac defense response, corresponding to the 10 median values, and 7 levels for the response to the picture–mask sequence corresponding to the 7 half seconds before the noise. The Greenhouse Geisser correction was applied to the repeated measure factors. Analysis of significant interaction effects were performed following Keppel’s procedure (Keppel, 1991). First, we identified the levels of the interacting factors explaining the significant effects (simple effects analysis). Then, if more than two groups or conditions were involved, multiple pairwise comparisons would be performed using Bonferroni test. The level of significance was set at 0.05 for all analysis. In addition, planned comparisons for cardiac defense—limited to the first five medians, the ones tracing the accelerative component of the response—, as well as effect sizes, would be performed for conscious and non-conscious groups separately. Effect sizes were calculated as the point-biserial correlation, \( r \) (Rosenthal and Rosnow, 1991). Statistical analysis for the self-report measures was a \( 2 \times 2 \) analysis of variance with the two between group factors (consciousness and order).

3. Results

3.1. Recognition of the visual stimuli

The results of the recognition questionnaire showed that all subjects in the non-effective masking group (conscious) correctly recognized the spider and the flower presented during the experimental session. From the 24 participants making up the effective masking group (non-conscious), 5 recognized the spider in the forced choice test (2 in the subgroup order 1 and 3 in the subgroup order 2) and 1 recognized the flower (belonging to the subgroup order 1). These six subjects were excluded from the statistical analyses reported here. The final sample of the study consisted of 42 subjects: 12 in the conscious group order 1, 12 in the conscious group order 2, 9 in the non-conscious group order 1 and 9 in the non-conscious group order 2.

3.2. Heart rate

3.2.1. Cardiac response to the picture–mask sequence before the noise

The \( 2 \times 2(\times 2 \times 7) \) ANOVA showed no significant effect in any of the main factors and interactions. Fig. 2 plots the cardiac response to the picture–mask sequence...
sequence just before the response to the noise as a function of picture, order and consciousness. The Time factor showed an overall heart rate form characterized by an initial acceleration followed by a deceleration. This response pattern is more pronounced in the second run–irrespective of picture content–suggesting a non-significant tendency to show such response after the first noise presentation (see Fig. 2a and b). However, this tendency is only observed in the non-conscious group, both in orders...
1 and 2, suggesting that the faster (non-conscious) picture–mask sequence is probably the relevant factor explaining this response pattern after the first noise presentation (see Fig. 2e and f).

3.2.2. Cardiac defense response
The $2 \times 2(2 \times 10)$ ANOVA showed significant main effects of picture ($F(1,38)=4.19$, $p<0.05$) and time ($F(9,342)=14.47$, $p<0.0001$, $\eta=0.422$) and significant interaction effects of picture\times order ($F(1,38)=9.97$, $p<0.003$) and picture\times order\times time ($F(9,342)=4.82$, $p<0.001$, $\eta=0.474$). No significant main or interaction effects were obtained for consciousness.

The significant main effect of the picture factor indicates that the average heart rate response to the noise was significantly different depending on

![Graphs showing cardiac defense response to intense noise](image-url)

Fig. 3. Cardiac defense response to the intense noise. (a and b) Average of conscious and non-conscious groups in order 1 and order 2; (c and d) conscious group in order 1 and order 2; (e and f) non-conscious group in order 1 and order 2.
whether the picture projected was phobic or non-phobic. The significant main effect of time indicates the presence of a specific pattern of heart rate changes produced by the noise. The significant picture×order and picture×order×time interactions indicate that the different response to the phobic and non-phobic picture was affected by the order of picture presentation and by the time interval of the heart rate changes after the eliciting stimulus. Analysis of the picture×time interaction in order 1 showed a significant picture×time effect in order 1–spider followed by flower–(F(9,180)=5.76, p<0.0011, ε=0.441), not in order 2–flower followed by spider–(F(9,180)=1.33, p>0.270, ε=0.388). Subsequent analysis of the picture×time interaction in order 1 showed significant differences between spider and flower in medians 1 to 7 (all Fs>5.46, ps<0.02). The pattern of the cardiac response in order 1 shows a larger and longer initial acceleration when the noise was preceded by the picture of the spider than when preceded by the picture of the flower (see Fig. 3a). In order 2, no significant differences are observed in the pattern of both responses (see Fig. 3b). Given the rapid reduction of the response with repetition of the stimulus, this later result in order 2 indicates that the rapid reduction tendency was eliminated by presenting in the second run the phobic picture.

As regards the conscious and non-conscious groups, a similar pattern of response is observed, although the magnitude of the changes is larger in the conscious group (see Fig. 3c and e). Planned comparisons showed, in the conscious group order 1, significant differences between the phobic and non-phobic picture in median 2 (F(1,11)=13.90, p<0.004, r=0.75), median 3 (F(1,11)=37.74, p<0.001, r=0.88), median 4 (F(1,11)=8.01, p<0.02, r=0.65) and median 5 (F(1,11)=21.42, p<0.002, r=0.81). In order 2, the conscious group did not show any significant difference. In the non-conscious group order 1, significant differences appeared in median 1 (F(1,8)=10.55, p<0.02, r=0.75) and median 2 (F(1,8)=5.36, p<0.05, r=0.63), no significant difference being found in order 2 in any of the medians.

3.3. Subjective reactivity

A 2×2 analysis of variance with two between group factors, picture and consciousness, was applied to the assessment of the intensity and unpleasantness of the noise. A significant main effect of picture was found for unpleasantness (F(1,38)=4.52, p<0.04), the noise being evaluated as more unpleasant when presented with the phobic picture (mean=8.44, S.D.=0.98) than when presented with the non-phobic picture (mean=7.74, S.D.=1.13). No other main or interaction effect was found for unpleasantness or intensity. The absence of significant effects of consciousness indicates that the conscious and non-conscious groups were not significantly different (conscious group: mean phobic=8.66, S.D.=0.49; mean non-phobic=7.92, S.D.=1.0; non-conscious group: mean phobic=8.22, S.D.=1.39; mean non-phobic=7.55, S.D.=1.33).

4. Discussion

The results of the present study provide evidence for the modulating effect of affective priming on the cardiac defense response: the response was increased in the first trial, and less reduced in the second trial, when the noise was preceded by the phobic picture. Importantly, this effect was present in both effectively masked (non-conscious) and non-effectively masked (conscious) conditions. These results thus provide support and extend Ohman’s model of pre-attentive processing of fear by generalizing to another defensive reflex, the cardiac defense response. Moreover, these results support and extend Lang’s motivational priming model by generalizing to another defensive process. The results suggest that the effect is due to a defensive and not an orienting response.

The backward masking technique was effective in most, but not all, of our subjects. 18 out of the 24 participants in the non-conscious group could not identify any of the visual stimuli in the recognition questionnaire. Those six subjects who showed recognition of any of the stimuli were excluded from the statistical analysis in order to guarantee a proper comparison between the masked and the unmasked conditions. The finding of around 20% of the subjects in the effective masking group being able to report some awareness of the masking procedure replicates...
previous studies (Morris et al., 1998; Whalen et al., 1998).

The analysis of the heart rate response to the picture–mask sequence during the 3.5 s preceding the noise did not show any significant response pattern nor differences between groups (conscious versus non-conscious, order 1 versus order 2) and conditions (phobic versus non-phobic). Therefore, no evidence of differential processing of the phobic picture under either conscious or non-conscious conditions was obtained prior to the noise. It seems that the complex picture–mask sequence, with a picture, a mask and a no picture interval presented each second, together with the reduced number of trials, has prevented the elicitation of a significant heart rate response. There was a tendency to show an initial heart rate acceleration—with peak at 1.5 s—followed by a deceleration—with peak just before the noise onset—but only in the non-conscious group and on the second run, irrespective of picture content. This tendency may be explained as due to conditioning effects (the noise acting as unconditioned stimulus) on the non-conscious stimulus sequence preceding the noise (the shorter picture–mask presentations acting as a more salient conditioned stimulus). However, due to the reduced number of trials, no conclusion can be drawn on the lack of a significant heart rate response during this interval. Studies showing heart rate responses to masked and unmasked pictures (Ingjaldestson et al., 2003a,b) have used a higher number of trials (20) and a longer time interval after picture onset (11 s). Interestingly, evidence of differential processing was only obtained in the masked condition.

As regards cardiac defense, we found a potentiated and modified cardiac response to the noise when primed by viewing non-masked phobic material. The potentiated heart rate response was predicted following Lang’s motivational priming model. According to this model, the presentation of the phobic picture activates the aversive motivational system, which in turn potentiates reflexes that match the system, such as motor startle or cardiac defense. The potentiated heart rate response is consistent with previous findings (Sánchez et al., 2002) and with the neural model underlying fear potentiation of startle proposed by Davis and colleagues (Davis et al., 1999; Lang et al., 2002). They have hypothesized that the same neural circuitry involving the central nucleus of the amygdala applies to other forms of defensive reactions such as freezing, fight, flight, facial expressions of fear and autonomic reactions.

The modified pattern of the cardiac response suggests that the defense reaction to an intense acoustic stimulus is different depending on whether the eliciting stimulus is primed or unprimed. The disappearance of the deceleration and the observation of a single large acceleration suggest a change in the normal defense response to an intense noise alone. Although alternative explanations are possible, it has been speculated that the modified pattern reflects a change in the sequence of attentional and motivational processes involved in natural defense (Pérez et al., 2000; Vila et al., 2003; see also Blanchard and Blanchard, 1989; Fanselow, 1994; Lang et al., 1997, 2002): the initial attentional factors, presumably indexed by the deceleration, seem to disappear under primed conditions, and the subsequent preparatory protective actions, presumably indexed by the second acceleration, seem to advance in time.

Turning now to the masked condition, our data suggest that the above considerations also apply when the subjects are unaware of the preceding fearful stimulus. The same pattern of cardiac changes was observed, although the magnitude of the changes was smaller. The effect size of the significant planned comparisons between the phobic and non-phobic picture was always greater for the conscious than for the non-conscious group. However, in both the conscious and non-conscious groups, the effect sizes were in the medium to large effect size range as described by Cohen (1992). In addition, no significant group differences were found between the conscious and non-conscious groups and all the significant differences found in the form of the response in both groups went in the same direction.

These data support Öhman’s model of pre-attentive processing of fear (Öhman, 1997, 2000). However, in contrast to Öhman’s studies, we have used as the dependent variable the heart rate response instead of the skin conductance response. In this context, the skin conductance response has been generally interpreted as an attentional response (the orienting reflex), in spite of being studied in the context of aversive classical conditioning or in response to phobic stimuli (Esteves et al., 1994b; Öhman and Soares, 1994). The
issue is whether the skin conductance data on masked processing of aversive stimuli is indicative of orienting, defense or both. Our finding of a potentiated heart rate acceleration to the eliciting noise only when the masked phobic stimulus was presented suggests that the nature of the response is indeed defensive. The affective rating of the first noise supports this interpretation: subjects in both conscious and non-conscious groups rated as more unpleasant the noise preceded by the phobic stimulus than the noise preceded by the non-phobic one.

In summary, our results provide evidence that a fear relevant picture, compared to a non-fear relevant picture, potentiates the cardiac defense response under both effective masking conditions and non-effective masking conditions. These results support both Lang’s motivational priming model and Öhman’s model of pre-attentive processing of fear.

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