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Research Report
Repetitive picture processing: Autonomic and cortical correlates
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ABSTRACT

Emotionally arousing pictures elicit larger late positive potentials (LPPs) than neutral pictures during passive viewing. Moreover, these cortical responses do not rely on voluntary evaluation of the hedonic content and are relatively unaffected by task demands. In this study, we examined modulation of the late positive potential as it varies with stimulus repetition. Three pictures (pleasant, neutral, unpleasant) were presented up to 60 times each. Although the amplitude of the late positive potential during picture viewing declined with stimulus repetition, affective modulation remained intact. On the other hand, autonomic responses (skin conductance and heart rate change) habituated rapidly with stimulus repetition. These findings suggest that while stimulus detection and categorization, reflected in the LPP, is mandatory, autonomic modulation reflects initial orienting responses that habituate rapidly.

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1. Introduction

Emotional responses are mediated by brain circuits that have evolved to organize behavior for the purpose of survival. Several studies have shown that affective pictures are effective cues in activating emotional response and suggest that affective responses serve different functions and reflect the motivational system that is engaged (i.e., defensive or appetitive) and its intensity of activation (Bradley, 2000; Lang et al., 1997). For instance, skin conductance responses are larger when viewing emotionally arousing (pleasant or unpleasant) pictures, compared to neutral pictures, covarying with the intensity parameter, whereas heart rate change varies with hedonic valence (unpleasant or pleasant), with

more deceleratory heart rate responses elicited when viewing unpleasant, relative to pleasant pictures. Moreover, these responses, for the most part, habituate with repeated presentation of the same affective pictures, suggesting they index differences in initial orienting to motivationally relevant stimuli (Bradley et al., 1993).

Event-related potential studies have consistently found that emotionally arousing (pleasant and unpleasant) pictures elicit a larger late positive potential (LPP) than neutral pictures in a window from 400–800 ms after picture onset (Cacioppo et al., 1994; Cuthbert et al., 2000; Schupp et al., 2000). The magnitude of the late positive potential clearly varies with emotional arousal, with the largest LPPs elicited when viewing the most arousing emotional contents (Schupp et al., 2004).

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This effect has been theoretically linked to the concept of motivated attention, which proposes that motivationally significant stimuli are selectively processed because they naturally engage attentional resources (Bradley, 2000; Lang et al., 1997).

Moreover, these cortical responses do not appear to rely on voluntary evaluation of the hedonic content and are relatively unaffected by task demands. For instance, Cuthbert et al. (1995) compared the magnitude of the LPP elicited during passive viewing to when an evaluative rating task was required and found similar modulation, suggesting that passive viewing elicits similar cortical processing as an explicit evaluative task. Consistent with this reasoning, several behavioral studies have found involuntary semantic processing of affective stimuli (McKenna and Sharma, 2004; Pratto and John, 1991; Stenberg et al., 1998), as well as that affectively congruent stimuli prime both pronunciation and lexical decisions, in the absence of an explicitly evaluative context (Bargh et al., 1996; Giner-Sorolla et al., 1999; Hermans et al., 2001; Wentura, 1998).

In the current study, we examined modulation of the late positive potential as it varies with stimulus repetition, by presenting pleasant, neutral, and unpleasant pictures up to 60 times each. Past studies have found a decrement in the overall amplitude of a number of event-related potentials (ERPs) to repeatedly presented neutral stimuli (Friedman et al., 1998; Ravden and Polich, 1998). For instance, several

studies (Bruin et al., 2000) have found that the N1 and P3 components decrease in amplitude as a function of stimulus repetition, with the decrease in the N1 amplitude faster than that of the P3. One theoretical interpretation of the habituation of the P3 component (Ravden and Polich, 1998) is that it reflects differences in initial orienting activity, drawing on Sokolov's theory that a mismatch between internal and external neural representations prompts orienting (Donchin and Coles, 1988; Sokolov, 1963). Habituation of P3-like potentials is more evident in passive paradigms, compared with active discrimination oddball tasks (Polich and McIsaac, 1994), and in the current study, pictures were passively viewed to optimize the possibility of observing habituation of the event-related potential.

In addition to measuring the late positive potential, we also measured skin conductance and heart rate responses, as well as reports of pleasure and arousal. We expected that the peripheral responses would habituate rapidly, evidencing differences due to the affective content of the stimuli only early in the habituation phase, as found previously (Bradley et al., 1993). Although several previous ERPs studies investigated the effect of picture content on the LPP, none have yet examined the consequence of picture repetition on the LPP. Thus, the major question is whether the late positive potential will decrease with repetition and whether, in particular, affective modulation changes with repetitive processing.

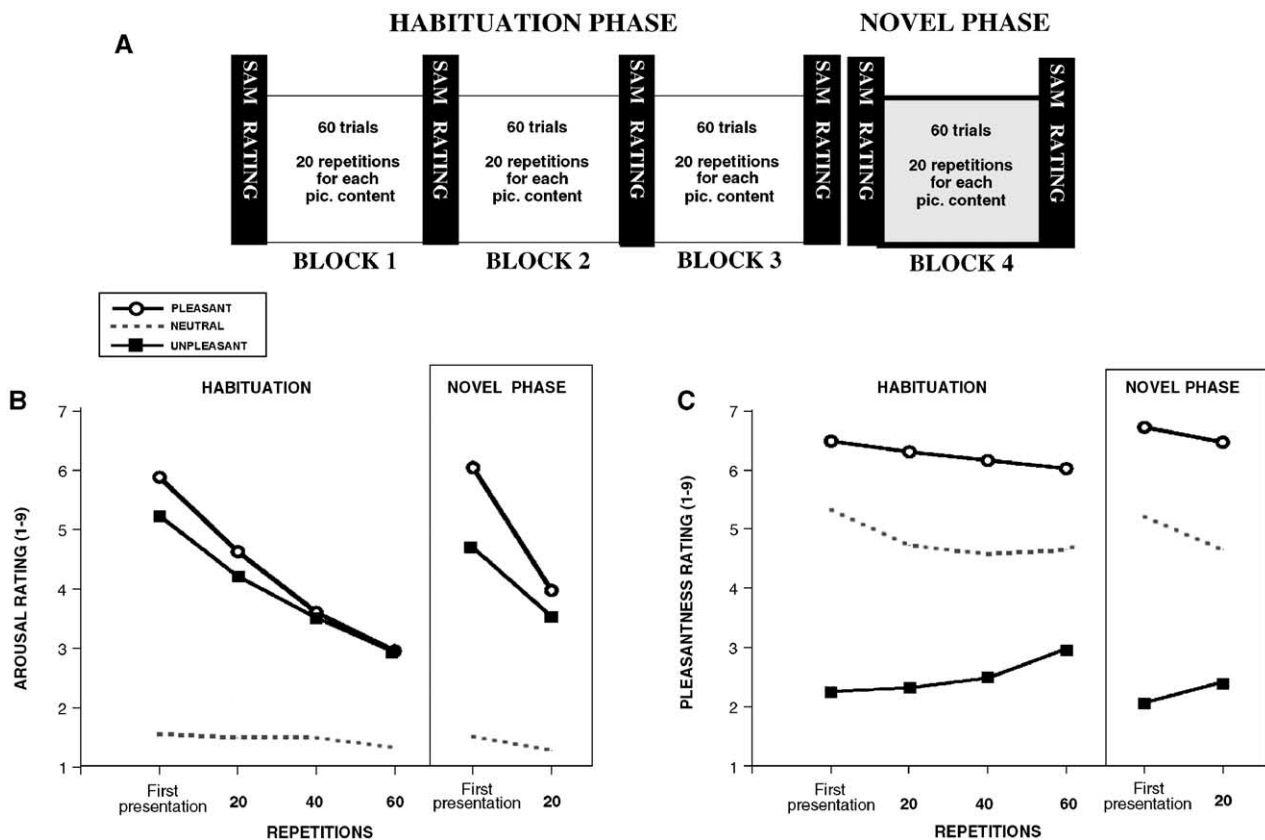


Fig. 1 – (A) Schematic diagram showing the sequence of events in the present study. SAM ratings of arousal (B) and pleasure (C) for pleasant, neutral, and unpleasant pictures at the beginning of the first block (new), at the end of the first block (after 20 repetitions), at the end of the second block (after 40 repetitions), at the end of the third block (after 60 repetitions), at the beginning of the novel phase (new), and at the end of the novel phase (after 20 repetitions).

In order to attribute potential decreases in responsivity to a central, rather than to a peripheral process (e.g., fatigue), we introduced a novel set of stimuli following the habituation phase to assess reactivity and expected full recovery of all habituated responses in this phase of the experiment.

2. Results

2.1. Self-report measures

As illustrated in Fig. 1B, arousal ratings were affected by picture repetition, picture content \times block, $F(10,39) = 9.91, P < 0.001$. Reports of arousal decreased across the habituation blocks for emotional pictures (first presentation vs. block 3 [$F(1,48) > 54, P_s < 0.001$]) but remained unchanged for neutral stimuli [first presentation vs. block 3 [$F(1,48) < 2, P > 0.05$]]. However, even after the third block, emotional pictures were rated as more arousing than neutral pictures [$F(1,48) > 29, P_s < 0.001$]. Arousal ratings for new emotional pictures in the novel phase were higher than for those rated in the last block of the habituation phase which had been repeated 60 times [$F(1,48) > 52, P_s < 0.001$] but no different than the initial ratings of the repeated picture set [$F(1,48) < 2, P_s > 0.05$].

As illustrated in Fig. 1C, unpleasant pictures were judged less pleasant than neutral pictures, which were in turn rated less positively than pleasant pictures. A significant interaction of picture content and block, $F(10,39) = 3.57, P < 0.005$, indicated that, while pleasantness ratings for pleasant pictures did not change across blocks, neutral, and unpleasant pictures were affected by repetition. Across time in the habituation phase, neutral pictures were judged as less pleasant [first presentation vs. block 3, $F(1,48) = 8.58, P < 0.01$] and unpleasant pictures as less unpleasant [first presentation vs. block 3, $F(1,48) = 11.67, P < 0.005$]; in the novel phase, both neutral and unpleasant pictures regained the same ratings as those seen in the beginning of the habituation phase.

2.2. Physiological measures

A significant three-way interaction of measure (3) by block (4) by picture content [$F(12,33) = 2.6, P < 0.05$] indicated that the LPP, heart rate and skin conductance were differentially affected by stimulus repetition and picture content. In addition, each measure was differentially affected by picture repetition, measure by block interaction [$F(6,39) = 11.5, P < 0.001$]. To followup the significant three-way interaction, effects of block and picture content were assessed for each measure separately.

2.3. Event-related potentials

Replicating previous findings (Cuthbert et al., 1995), the late positive potential was modulated by picture content [$F(2,45) = 70.5, P < 0.001$] (see Fig. 2), with pleasant and unpleasant pictures eliciting a more pronounced late positive potential (LPP) compared with neutral pictures [$F(1,46) > 107, P_s < 0.001$]. LPP magnitude for pleasant and unpleasant pictures did not differ [$F(1,46) < 2$].

A main effect of Block [$F(3,44) = 15.1, P < 0.001$], indicated that across the habituation phase the LPP decreased in amplitude [block1 vs. block3, $F(1,46) = 27.7, P < 0.001$]. Introduction of novel pictures following the habituation phase elicited a significant reinstatement of cortical positivity [block 4 vs. block 3, $F(1,46) = 23.7, P < 0.001$], and LPP amplitude during the novel phase was not statistically different from that elicited in the first block of the habituation phase [$F(1,46) < 1, P > 0.05$]. Further analysis using blocks of ten repetitions instead of twenty revealed that cortical positivity reached a plateau after approximately thirty repetitions; in particular, block one was not different from block two [$F(1,46) < 2, P > 0.05$]; block two was different from block three [$F(1,46) = 7.3, P < 0.01$], block three was not different from the following habituation blocks [$F(1,46) < 2, P > 0.05$].

Modulation of the LPP by picture content did not vary across blocks, however, [$F(6,41) = 1.8, P > 0.05$], indicating that the enhanced LPP for emotional stimuli, compared to neutral ones, persisted regardless of picture repetition (see Fig. 3A). In fact, even after sixty repetitions, the effect of picture content was highly significant [$F(2,45) = 34, P < 0.001$].

A significant main effect of location was found [$F(2,45) = 105.5, P < 0.001$], with the largest positivity observed at Pz, intermediate at Cz and least at Fz, all location comparison [$F(1,46) > 56, P < 0.001$]. Replicating previous studies (e.g., Cuthbert et al., 2000; Schupp et al., 2000), the LPP was significantly modulated as a function of picture content in all sites [$F(2,45) > 51, P < 0.001$]. Also, picture content interacted with site [$F(4,43) = 11.4, P < 0.001$], showing that affective modulation (emotional minus neutral pictures) was greatest at Cz and Pz compared to Fz [$F(2,45) > 4.5, P < 0.05$]. The interaction of location and block was not significant [$F(6,41) = 2.0, P > 0.05$].

Modulation of the LPP by picture content across blocks did not change as a function of sensor location, location by block by picture content interaction [$F(12,35) = 0.9, P > 0.05$]. In particular, the block by picture content interaction was not significant at Fz [$F(6,41) = 1.9, P > 0.05$], at Cz [$F(6,41) = 1.5, P > 0.05$], and at Pz [$F(6,41) = 2.1, P > 0.05$]. During the third block of the habituation phase emotional pictures (pleasant and unpleasant) continued to elicit larger LPP compared to neutral ones in all sites, Fz [$F(2,45) = 20.1, P < 0.001$], Cz [$F(2,45) = 30.1, P < 0.001$], Pz [$F(2,45) = 31.3, P < 0.001$].

Further analyses were conducted to evaluate the effects of picture content and block on early ERPs components such as N1 and P2 as well as on successive time intervals following the late positive potential (from 800 to 5000 ms) for each location. Results indicated that only the N1 amplitude decreased across blocks [$F(2,45) = 3.7, P < 0.05$] and did not vary with picture content or electrode site. On the other hand, picture content affected ERPs (slow waves) until 3000 ms, whereas a decrease across blocks was observed in the first 1000 ms after stimulus onset (all $F_s > 3.5, P_s < 0.05$). No interactions of these factors with location were found.

2.4. Skin conductance responses

A significant interaction between picture content \times block [$F(6,42) = 2.98, P < 0.05$] indicated that skin conductance response magnitude evidenced a rapid habituation across blocks for

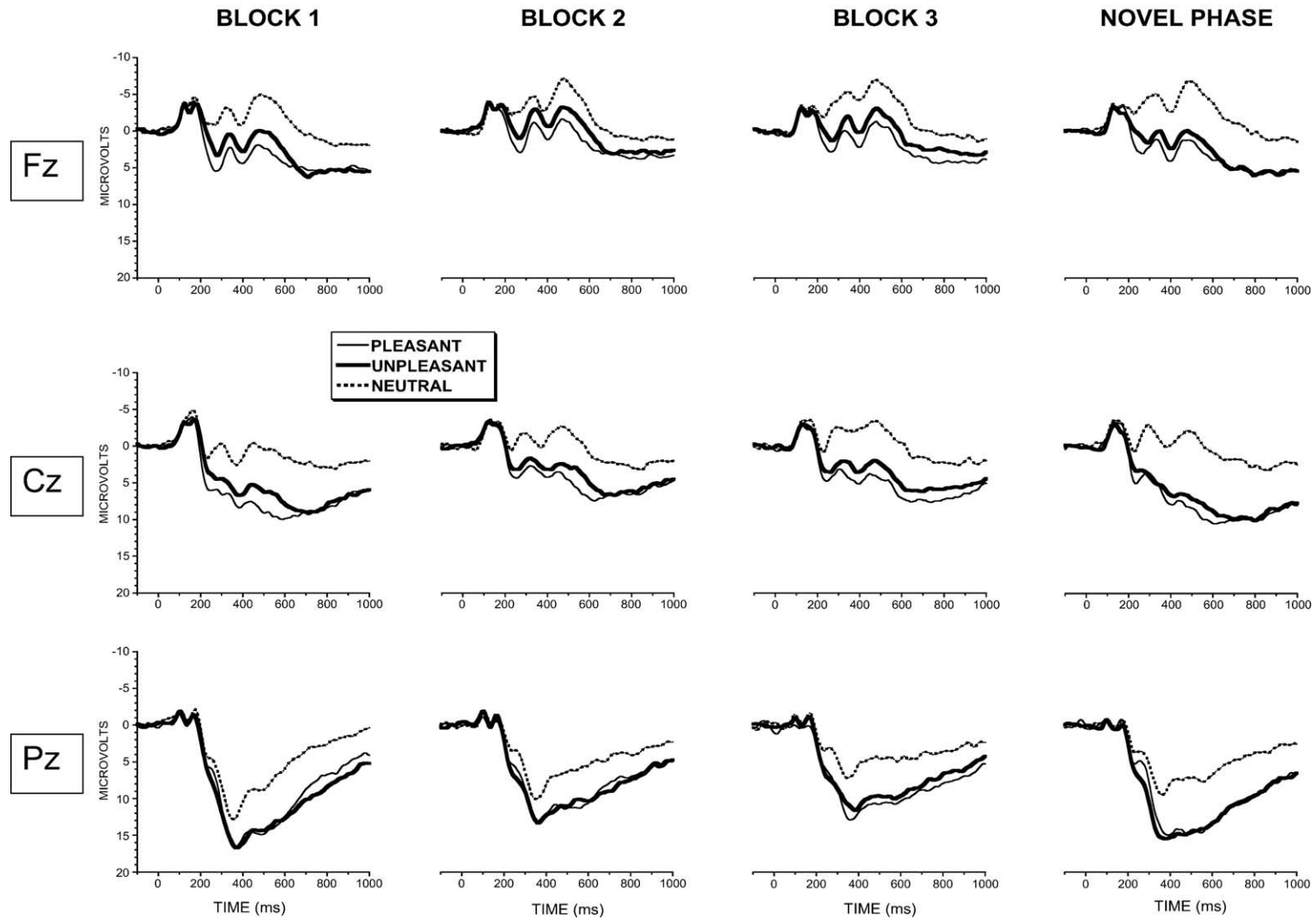


Fig. 2 – Grand-averaged ERP waveforms (Fz, Cz, Pz) elicited by pleasant, neutral, and unpleasant pictures during the habituation phase (three blocks) and the novel phase.

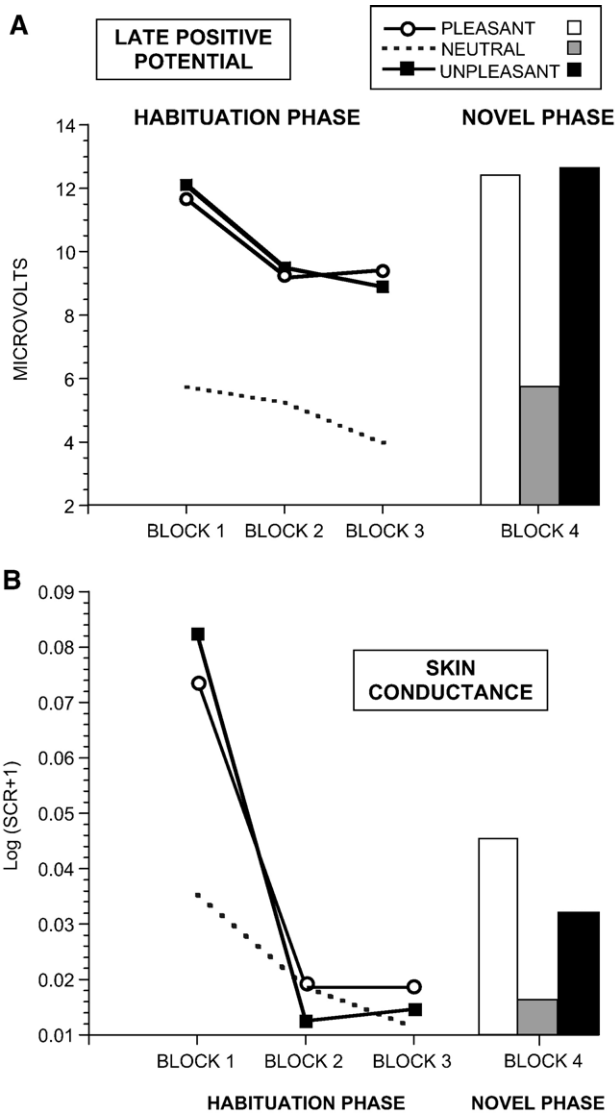


Fig. 3 – Late positive potential amplitude (Pz; 400–800 ms (A)) and skin conductance (B) changes elicited when viewing pleasant, neutral, and unpleasant pictures at each of the four blocks during the habituation and novel phases.

emotional stimuli (see Fig. 3B). In the first block, skin conductance replicated the typical arousal effect [$F(2,46) = 10.1, P < 0.001$], with larger changes when viewing pleasant and unpleasant, compared to neutral, stimuli [$F_s(1,47) > 10, P_s < 0.005$]; no differences were found between pleasant and unpleasant pictures, [$F(1,47) < 2, P > 0.05$]. The largest decrement in response magnitude occurred between Block 1 and 2 [unpleasant pictures, $F(1,47) = 10.56, P < 0.005$, pleasant pictures, $F(1,47) = 21.36, P < 0.005$, neutral pictures: ns.] During the second and third blocks of the habituation phase, skin conductance change did not discriminate between emotional and neutral stimuli [$F_s(2,46) < 2, P_s > 0.05$].

In the novel phase, the new set of pictures elicited reinstatement of the skin conductance changes that were similar in size to those elicited in the first block [block 1 vs. block 4: $F(1,47) = 2.6, P > 0.05$]. As in the first block of the

habituation, affective stimuli prompted larger electrodermal responses than neutral pictures, picture content [$F(2,46) = 4.19, P < 0.05$].

2.5. Heart rate responses

A significant main effect of block was obtained, [$F(3,45) = 5.9, P < 0.005$]. In the habituation phase, heart rate deceleration was larger in the first block compared to the second and third blocks, [$F_s(1,47) > 13, P_s < 0.001$], no differences were found between the second and the third block [$F(1,47) < 2, P > 0.05$]. In the novel phase, the presentation of a new set of pictures prompted larger heart rate deceleration compared to the last habituation block [novel phase vs. block 3, $F(1,47) = 8.4, P < 0.01$], this deceleration was not different with the one obtained during the first block of the habituation phase [$F_s(1,47) < 2, P_s > 0.05$].

The main effect of picture content and the interaction of picture content and block were not significant [$F_s < 2, P_s > 0.05$]. However, when analysis was confined to fewer repetitions (ten instead of twenty) for each content, the interaction of picture content and block was significant [$F(14,34) = 3.14, P < 0.005$] (see Table 1). In the first block of ten repetitions heart rate was affected by picture content [$F(2,46) = 4.9, P < 0.05$], showing larger deceleration during unpleasant picture viewing compared to neutral and pleasant pictures [$F_s(1,47) > 7.9, P_s < 0.005$], in the following habituation blocks no differences were found as a function of picture content [$F_s(14,34) < 2, P_s > 0.05$]. In the novel phase, during the first ten repetitions, the effect of picture content was again significant [$F(2,46) = 12.7, P < 0.001$] indicating larger deceleration during unpleasant picture viewing compared to neutral and pleasant stimuli [$F_s(1,47) > 4, P_s < 0.05$].

3. General discussion

Although the amplitude of the late positive potential elicited during picture viewing was attenuated by stimulus repetition, affective modulation of the LPP remained intact. Greater cortical positivity in a window from 400–800 ms after picture onset was preserved for emotionally arousing (pleasant and unpleasant) pictures, compared to neutral stimuli, throughout a long, repetitive habituation phase. The overall decrement in the LPP across repetitions is consistent with the hypothesis that attention to the picture declined with stimulus repetition. This decline was not due to general fatigue but was specific to

Table 1 – Mean scores (and standard errors) for heart rate (beats per minute) during each block of ten repetitions

		Pleasant	Neutral	Unpleasant
Habituation	Block 1	-0.39 (0.23)	-0.43 (0.21)	-1.14 (0.37)
	Block 2	-0.63 (0.25)	-0.50 (0.22)	-0.32 (0.24)
	Block 3	-0.36 (0.25)	-0.06 (0.27)	-0.66 (0.23)
	Block 4	-0.03 (0.28)	-0.18 (0.25)	-0.13 (0.23)
	Block 5	-0.46 (0.23)	0.16 (0.19)	0.20 (0.24)
	Block 6	0.14 (0.25)	-0.35 (0.24)	-0.01 (0.28)
Novel phase	Block 7	-0.91 (0.27)	0.01 (0.21)	-1.55 (0.30)
	Block 8	0.12 (0.25)	-0.49 (0.16)	-0.15 (0.29)

highly repeated stimuli, as clearly shown by the renewal of the LPP when a new set of pictures was presented in the novel phase.

In spite of an overall reduction in attention to repeated stimuli, the affective content of the stimuli continued to prompt a larger LPP, and this modulatory difference was unaffected by picture repetition. Based on the idea that resistance to habituation is one important feature of an obligatory process, these results suggest that affective discrimination of visual stimuli is obligatory, despite continuous repetitions, and that motivationally relevant picture contents automatically “grab” attention. Because participants were only asked to look at the pictures, without an additional task, however, we cannot rule out the possibility that affective modulation of the LPP results from a process in which greater attention is voluntarily allocated to affective pictures.

An interesting strategy for determining whether affective modulation of the LPP and its resistance to habituation is due to obligatory or voluntary processes would be to present affective pictures when participants are engaged in a competing task. Recently, Harris and Pashler (2004) have shown that although emotional words initially attract attention, after only a few repetitions these stimuli become less distracting in a performance task, and act more like neutral stimuli. This rapid habituation may be due to relatively weaker capacity of words, compared to pictures of natural scenes, to elicit emotional responses, as the authors noted. Assessing event-related potentials for affective pictures in the context of competing tasks could be informative, it could also affect the LPP by introducing new ERP components engaged by the task itself.

It is possible that the absence of affective habituation in the LPP is due to the use of a relatively long inter-stimulus interval (ITI); use of a shorter ITI might stress the habituation process. However, given that the main aim of the present study was to compare ERPs responses with autonomic measures in terms of habituation, we employed a long ITI because it was necessary for assessing habituation of the autonomic measures. On the other hand, the present findings do not support a hypothesis that differential LPP responses to emotional pictures reflect sheer differences in familiarity between affective and neutral pictures (e.g., that mutilation is less familiar than neutral faces): After up to sixty repetitions, presumably all of the pictures were quite familiar and yet larger LPPs continued to be elicited by affective stimuli.

Reports of pleasure and arousal changed across the habituation phase, as did autonomic responses (skin conductance and heart rate). In general, these changes indicated a decrease in reactions to emotional stimuli with increased repetition. After sixty repetitions, participants judged unpleasant pictures as less unpleasant, and both pleasant and unpleasant pictures as less arousing. However, it should be noted that reports of arousal changed more dramatically across the habituation phase compared to reports of pleasure, suggesting that picture repetition is more likely to change the intensity than hedonic value. Consistent with this, differentiation between emotional and neutral pictures for both skin conductance and heart rate rapidly habituated. In particular, fewer picture repetitions attenuated affective modulation in heart rate compared to skin conductance change. These results replicate a previous study (Bradley et al., 1993) which

also found rapid habituation of skin conductance and heart rate following a few presentations of the same neutral or emotional pictures. Again, these changes were not due to peripheral mechanisms such as fatigue, as affective differentiation was reinstated (that was not different from the initial block of habituation) when new pictures were presented in the novel phase.

The persistence of affective modulation in the late positive potential and the rapid decrease in differential autonomic responses to emotional stimuli raises questions regarding the function of different affective reactions. One hypothesis is that these responses reflect different processes (Bradley, 2002). According to this notion, heart rate and skin conductance reflect different facets of initial orienting responses, with heart rate reflecting processes related to sensory intake and skin conductance indexing preparation for action. Motivationally relevant stimuli generally prompt heightened responses in these measures, but orienting decreases when the stimulus is easily recognized (i.e., increased stimulus intake is not necessary), and it is clear that no adaptive action is necessary (as when the same picture is repeatedly presented).

On the other hand, stimulus detection and categorization, reflected in the LPP, continue to occur whenever a sensory stimulus is presented. The processing of the stimulus is mandatory, in the sense that, despite the number of repetitions, the new stimulus must be recognized before it can be categorized as previously presented. Affective stimuli are presumed to include associations to motivational systems in the brain (e.g., appetitive and defensive) (Lang et al., 1997) that activate outputs to the neural circuits involved in orienting and action (e.g., hypothalamic autonomic nuclei). One interpretation of our results is that stimulus repetition does not change the associative strength of connections to these subcortical motivational systems, as reflected in the LPP, whereas outputs to systems mediating orienting and action do habituate. According to this account, the LPP will primarily be changed by altering the affective association (e.g., by a counter-conditioning procedure).

In the present study, the analysis was restricted to ERPs waveforms at a few locations in the scalp. Future studies could investigate affective habituation using a high density electrode array as well as other neuroimaging techniques (functional magnetic resonance imaging and magneto-encephalography) in order to evaluate which cortical areas are involved in the reduction of autonomic responses. The results clearly indicate, however, that changes in the brain's response after 60 presentations of the same affective pictures are very similar to the brain's initial response to these stimuli and reflect continued discrimination between emotionally arousing and neutral stimuli long after the initial autonomic orienting responses have ceased.

4. Experimental procedures

4.1. Participants

Fifty volunteers (26 female, 22–34 years) gave written consent and participated in the study. Because the unpleasant pictures depicted a mutilated body, blood phobia was evaluated using

the Mutilation Questionnaire (Klorman et al., 1974), and participants scoring above 18 were excluded. Because of computer or experimenter error, some participants were excluded from analyses of some dependent measures. Final *N*s were as follows: pleasure and arousal ratings, *n* = 49; ERPs analysis, *n* = 47; heart rate, *n* = 48; skin conductance responses, *n* = 48.

4.2. Materials and design

Fifteen pictures were selected from the International Affective Picture System (IAPS; Center for the Study of Emotion and Attention, CSEA-NIMH, 2001; Lang et al., 2001), depicting 5 unpleasant events (mutilated bodies), 5 pleasant events (erotic couples), and 5 neutral faces of people. Five different sets of three pictures (one for each affective content) were created to increase the generalizability of the results beyond the specific features of a limited set of experimental stimuli. Each subject saw one set of pictures during the 180-trial habituation phase. Each of the three pictures was presented 60 different times. The pictures were arranged in three blocks of 60 trials, such that each of the three pictures was presented 20 times in each block.

After the 60 presentations of each of the three pictures, a novel, previously unseen set of three pictures was presented (novel phase). The novel phase was arranged in a block of 60 trials, consisting of 20 presentations of each new picture. Order of stimulus set was counterbalanced across subjects such that, across participants, each set of pictures was seen equally often in the habituation or novel phase. The order of the picture presentation was pseudo-randomized with the restriction that no more than two times consecutively the same picture could occur. Each picture was presented for 6-s viewing interval on a 19-in. (48.3 cm) monitor, situated approximately 80 cm from the participant.

Pleasure and arousal ratings for each picture were obtained by using the paper and pencil version of the Self-Assessment Manikin (SAM; Lang, 1980), which depicts a graphic figure that varies along two dimensions of pleasure and arousal, on a 9-point scale. SAM ranges from a smiling, happy figure to a frowning, unhappy figure when representing the pleasure dimension; on the other hand, SAM ranges from an excited, wide-eyed figure to a relaxed, sleepy figure for the arousal dimension. In the paper-and-pencil version of SAM, the subject can place an "X" over any of the five figures in each scale or between any two figures, which results in a 9-point rating scale for each dimension.

4.3. Physiological data collection and reduction

Peripheral and electroencephalogram (EEG) signals were recorded from 3 s before picture onset until 3 s after the end of the picture period. The International 10–20 system was followed for three electrode sites: Fz, Cz, Pz; in addition, Sensormedics silver/silver chloride miniature electrodes were placed on the mastoids (A1, A2). All channels were referenced to Cz and digitally re-referenced off-line to linked mastoids. Vertical and horizontal eye movements were recorded using Sensormedics silver/silver chloride miniature electrodes to account for ocular artifacts. A 30-Hz high-frequency cut off and a 10-s time constant were used to record all cortical and ocular channels. The sampling rate was 125 Hz.

Individual EEG trials that were off-scale at any time during the 12-s recording epoch were excluded from further analysis; on average, 10.1% of the trials were thus excluded. To correct for vertical and horizontal ocular artifacts, an eye movement artifact correction procedure (Gratton et al., 1983) was applied to the EEG recordings including 1-s baseline, picture, and post-picture periods. ERP grand average waveforms were calculated separately for each valence category and electrode site (Fz, Cz, Pz), referenced to a 100-ms baseline preceding picture onset.

Statistical analyses were conducted in the time window between 400 to 800 ms with the aim to evaluate the effect of repeated presentations on the LPP.

Skin conductance electrodes were placed adjacently on the hypothenar eminence of the left palmar surface, using Sensormedic standard electrodes filled with the recommended 0.05-m NaCl Unibase paste. The signal was calibrated before each session to detect activity in the range of 0–40 μ S. Skin conductance was sampled at 20 Hz, and averages were calculated off-line in 500 ms bins. Reactions in skin conductance were determined by subtracting activity in the 1 s before picture presentation from that occurring at each half second after picture onset. Skin conductance (SC) change was defined as the largest response occurring between 1 and 4 s after slide onset. A log transformation was performed to normalize the data.

The electrocardiogram was recorded from the left and right forearms, using large Sensormedic electrodes filled with electrolyte paste. The signal was filtered with a Coulbourn S75-01 bioamplifier, and a trigger interrupted the computer each time it detected a cardiac R-wave. Interbeat intervals were recorded to the nearest millisecond and reduced off-line by using VPM software (Cook, 1997) into heart rate (HR) in beats per minute, in half-second bins.

Heart rate waveform scores were computed by subtracting, for each participant and each trial, activity in the 1 s before picture presentation from that occurring at each half second after picture onset; the values of heart rate during 6 s of picture presentation were averaged for statistical analysis.

A multivariate analysis of variance (MANOVA) was first conducted, using Measure (LPP magnitude, heart rate change, and skin conductance change), Block (the first three blocks of habituation and the last novel block), and picture Content (pleasant, neutral, unpleasant). Separate 3 (content) \times 4 (block) repeated measures analyses of variance were then conducted on each measure as warranted by significant interaction in the MANOVA.

Given the rapid habituation in heart rate and skin conductance change, each block was further split into two subsets of ten repetitions and analyzed using a 3 (picture content) \times 8 (subsets) repeated measures ANOVA. Similarly, separate 3 \times 6 repeated-measures ANOVAs were conducted for pleasure and arousal ratings, with picture content (pleasant, neutral, unpleasant) and block (after the first presentation, after each of the three habituation blocks, after the first presentation of the new pictures in the novel phase, and at the end of the novel phase) as within-subjects factors. For all analyses that involved repeated measures variables, the multivariate test statistic (Wilks' lambda) is reported to avoid potential sphericity issues (Vasey and Thayer, 1987).

4.4. Procedure

After completing the informed consent form and the mutilation questionnaire, participants were seated in a recliner in a dimly lit, sound-attenuated room, and the physiological sensors were attached. Each participant was then familiarized with the SAM rating procedure, which involves ratings of pleasure and arousal. Following two practice trials, the participant was instructed that a series of pictures would be presented, and that each picture should be viewed the entire time it was on the screen.

On the first three trials (consisting of presentation of the 3 different pictures), the participant made SAM ratings of pleasure and arousal for each picture. These 3 pictures were then each repeated for 19 times until the first block of 20 trials ended (see Fig. 1A). Following each block of habituation, the last three repetitions of each picture were rated using SAM. In the novel phase, 3 new pictures were presented and participants rated them immediately after picture offset. Again, after 20 repetitions of each picture, participants rated them again for pleasure and arousal.

Pictures were displayed for 6 s followed by a randomly determined inter-picture interval, which varied between 12 s and 20 s. The inter-block interval was less than a minute.

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