



Event-related brain potential correlates of conscious and non-conscious processing in anxiety. (Bial Foundation 144/06: awarded to Professor Anne Richards and Dr A Holmes.)

Final Report

The original BIAL proposal described a research programme to examine conscious and non-conscious processing in anxiety and to examine the resolution of ambiguity in anxiety. The first phase of the research manipulated emotional connotation by using an affective conditioning paradigm (see previously sent Progress Report). We found some interesting effects from this phase of the research but these were variable and the size of the effects were very small. We therefore decided to change the design of the experiments in Phase II in order to examine the same research questions but using a different methodology. In one series of experiments, we used the adaptation paradigm to manipulate the perception of emotionally ambiguous facial stimuli. We manipulated the perception of emotionally ambiguous facial expressions by using an adaptation paradigm. The first experiment was a pilot study carried out in order to select appropriate stimuli for the main study. In the main experiment, we measured both behavioural and electrophysiological responses. In the second series of experiments, we investigated the extent to which information relating to emotional facial expression is extracted under conditions of limited awareness by using the oddball paradigm. We gave an oral presentation of the results of the adaptation experiment at International Organisation of Psychophysiology, Budapest early this year. In addition, we have two papers in preparation (both due to be submitted at the end of this month). The findings from the research have also been discussed at informal meetings in the UK and in Taiwan. An outline of these two series of experiments followed by a summary of the pilot fEMG work is presented below:

Adaptation experiments

Prolonged exposure to a stimulus biases perception of a subsequent stimulus away from the adapting stimulus (Clifford & Rhodes, 2005). These are called after-effects and they have been observed with a wide variety of stimuli, including facial identity (Rhodes & Jeffery, 2006) and emotional facial expression (e.g., Webster et al., 2004). In the current experiment, we used this paradigm to examine the effect of anxiety on the perception of ambiguous emotional stimuli.

Electrophysiological research has identified a series of specific components are elicited in response to emotional stimuli. One such component is the early posterior negativity (EPN, see Schupp et al., 2004) component, which is an enhanced negativity over lateral posterior and occipital areas that begins after around 220 ms post-stimulus onset. The EPN is observable for negative relative to positive facial expressions. In addition to the EPN, there is an enhanced positivity effect for emotional (especially negative) expressions that begins within 200 ms post-stimulus onset and is located over prefrontal (fronto-central) electrodes. Following this early positivity is a later, sustained positivity that is more broadly distributed over fronto-parietal areas (Ashley et al, 2004; Krolak-Salmon et al., 2001). This enhanced positivity is referred to as the late positive potential (LPP) and occurs beyond 250 ms post-stimulus onset. The LPP is consistently elicited by negative facial expressions and there have been a few studies showing that the LPP is elicited by positive facial expressions too (Eimer et al., 2003; Marinkovic et al., 1998). ERPs have fine temporal resolution and therefore have the potential to dissociate processes that have different time courses (Holmes et al., 2009). It has been proposed that the early anterior positivity, which occurs within 200 ms of stimulus-onset, might be generated by rapid emotional detection mechanisms that are located in prefrontal or orbitofrontal areas, whereas the later more broadly distributed sustained positivity might reflect later emotional processes involved in decision-making and responding (Eimer and Holmes, 2007). In light of these proposals, it might be expected that the effects of adaptation on the processing of ambiguous emotional expressions is likely to be reflected in later rather than earlier

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ERP components. The current experiment will examine the EPN as well as early and late positive potentials.

Photographs of a male and a female model from the Karolinska Directed Emotional Faces set (KDEF; Lundqvist et al. 1998) were selected and a series of morphs created by interpolating a fear exemplar with a neutral exemplar for each model. We carried out a behavioural pilot study to establish a range of morphed expressions falling either side of the fear/neutral boundary, and we then used these in the main electrophysiological study. There were 16 adaptation blocks in the experiment proper, and each block began with an adapting phase in which participants viewed (and categorized) 35 presentations of one endpoint from the morph continuum. This was immediately followed by a test phase, during which participants viewed and categorized 33 images of the same model (8 dummy trials, 5 top-up trials and 20 test trials – 10 from either side of the categorical boundary). A total of 56 participants took part in this series of experiments.

We predicted that all test trials for both high and low anxious participants would be perceived as having an emotional expression opposite to that of the adapting stimulus (e.g., adaptation to fear would produce 'neutral' classifications for the test stimuli that were originally on the 'fear' side of the boundary). We were particularly interested in potential differences between the high and low anxious people. Using both behavioural and electrophysiological measures, we examined the mood-congruency hypothesis and the context-sensitivity hypothesis. The mood-congruency hypothesis would predict enhanced electrophysiological activity for anxious compared to non-anxious for the emotionally ambiguous morphs, irrespective of the nature of the preceding adapting stimuli. Behaviourally, the anxious should classify a greater proportion of the morphs as being 'fearful' than the non-anxious as 'fearful' emotion is congruent with the anxious participants' mood. The context-sensitivity hypothesis would predict enhanced electrophysiological activity for the anxious compared to the non-anxious following neutral adapt compared to fear adapt. Behaviourally, the context-sensitivity hypothesis would be supported by a greater increase for the anxious compared to the non-anxious in the proportion of 'fearful' classifications following neutral adapt than fear adapt. Such a

finding would support earlier research showing that anxious individuals are more influenced by contextual information than are non-anxious individuals (Blanchette & Richards, 2003, 2010; Richards, Blanchette & Munjiza, 2007).

EEG data were DC recorded (low-pass filter at 40 Hz, linked-earlobe reference) and digitized at a sampling rate of 500 Hz using a SynAmps amplifier (Neuroscan). Signals were recorded from 23 electrodes FP1, FP2, F3, Fz, F4, F7, F8, FC1, FCz, FC2, FC5, FC6, C3, Cz, C4, CP5, CP6, P3, P4, P7, P8, O1, O2 (according to the 10-20 system). Horizontal eye-movements (HEOG) were measured from two electrodes placed at the outer canthi of both eyes, and impedances for electrodes were kept below 5 K Ω . Following EEG recording, the EEG was epoched relative to a 100 ms pre-stimulus baseline. Trials with lateral eye movements (HEOG exceeding $\pm 30 \mu\text{V}$), as well as trials with vertical eye movements, eye blinks (FP1/FP2 exceeding $\pm 60 \mu\text{V}$), or other artifacts (a voltage exceeding $\pm 60 \mu\text{V}$ at any electrode) measured after stimulus onset were excluded from analysis. ERP mean amplitudes were obtained for specific sets of electrodes within predefined measurement intervals. One set of electrodes (FC1, FCz, FC2, C3, Cz, C4) was defined as a cluster representing frontocentral effects. Regional activity was analyzed at this cluster of electrodes within successive post-stimulus intervals of 160-220 ms (early positivity), 220-400 ms (early phase LPP), 400-600 ms (mid phase LPP), and 600-800 ms (late phase LPP). Activity was also analyzed across a second set of occipito-parietal electrodes (P7, P8, O1, O2) within a time window of 260-380 ms in order to examine EPN component effects. These electrode sites and time windows were determined on the basis of inspection of individual subject waveforms and previous reports (e.g., Eimer & Holmes, 2007; Holmes et al., 2009).

An analysis of the behavioural classification data revealed that adaptation to fear created a shift in the classification of morphs towards 'neutral' and adaptation to neutral created a shift towards 'fear'. This shift was equivalent in high and low anxiety. There was no evidence of a mood congruency effect from either the ERP or behavioural analyses. Compared to the non-anxious individuals, the anxious did not overall classify more expressions as fearful than neutral, and there was no evidence of an increase in electrophysiological activity either. The ERP data did not reveal any significant EPN components, but they did reveal a late positive potential in all

participants when processing emotionally ambiguous expressions following neutral adaptation compared to fearful adaptation. This shows that all participants were sensitive to the influence of the context. However, the late positive potential was significantly more pronounced in the high compared to the low anxiety group following adaptation to neutral compared to fear, showing an even greater sensitivity to contextual information in the anxious compared to the non-anxious individuals.

Oddball Experiments

In these two experiments we investigated the extent to which information relating to emotional facial expression is extracted under conditions of limited awareness. Previous ERP and fMRI studies have shown that when emotional faces fall within the focus of spatial attention, specific neurophysiological responses are elicited (e.g., the Late Positive Potential [LPP] in the case of ERPs, and amygdala activation in the case of fMRI). These neurophysiological markers of emotion processing are typically abolished when spatial attention is focused away from the faces (for reviews, see Eimer & Holmes, 2007; Vuilleumier, 2005). For many years, it was thought that the human brain was tuned for the automatic detection of motivationally relevant stimuli, but this recent neurophysiological evidence has challenged the prevailing view. The mechanisms underlying the impact of spatial attention on emotion processing, however, remain unclear. The elimination of emotion-related effects may occur because the low-level extraction of emotional expression features under conditions of non-attention is attenuated, thereby preventing any further analysis of higher level emotional meaning. Conversely, the featural analysis of emotional expressions may remain intact, but higher level affective evaluation may be inhibited. To help resolve this issue, we performed the present study to examine whether featural changes in facial expressions are detectable for stimuli presented outside the focus of spatial attention, consistent with a low-level analysis of physical characteristics. From such a result, it could be inferred that the gating effects of spatial attention are operating at a high level of expression analysis. If, however, there is no evidence of featural change detection

for emotional faces under conditions of inattention, it may be more consistent with a role of spatial attention in the attenuation of low-level physical properties of facial expressions.

A specific electrophysiological component known as the visual mismatch negativity (vMMN; Czigler, Balázs, & Pató, 2004) was selected to examine the extent of featural processing of non-attended facial expressions. Previous research has shown that task-irrelevant stimuli produce this component if the stimuli are presented infrequently within the context of a series of alternative frequent stimuli that differ in visual properties from the infrequent stimuli. The vMMN is suggested to be an index of an automatic and pre-attentive encoding of the violation of featural regularities in the visual world (see Czigler, 2007) and has been demonstrated for simple featural (e.g., colour) discrepancies between task-irrelevant stimuli presented either inside (e.g., Liu & Shi, 2008) or outside (e.g., Clifford, Holmes, Davies, & Franklin, in press) of foveal visual spatial attention. The vMMN has also been shown to be elicited by task-irrelevant (participants were engaged in an auditory task) emotional facial expressions falling within foveal visual spatial attention (Astikainen & Hietanen, 2009; Zhao & Li, 2006). The aim of the present study is to examine whether featural change for emotional expressions is detectable for infrequent stimuli that are both task-irrelevant and also fall *outside* of the focus of foveal visual spatial attention. A pattern of results showing a vMMN (indicative of featural change detection) for spatially non-attended stimuli, but an absence of classic emotional expression ERPs, would suggest that spatial attention is not a prerequisite for the extraction of low-level emotional expression features, but is nonetheless important for the processing of higher level affective meaning. If, however, a vMMN is not triggered by infrequent, unattended emotional expressions, it could be the case that changes in low-level features of emotional facial expressions are not processed, which would provide an alternative account of why ERPs triggered by emotional facial expressions have been absent in non-attended conditions in previous studies.

For the first experiment, the design was matched closely with that of two previous studies (Zhao & Li, 2006; Eimer, Holmes, & McGlone, 2003) in order to

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maximise the chances of: a) eliciting a vMMN to facial expression change; b) eliminating facial expression ERP effects by means of an attentional manipulation. Happy, fearful and neutral facial expressions were selected from one male face identity. Each block contained trials with happy-neutral, fearful-neutral and neutral-neutral face pairs, with each face within a pair presented bilaterally to the left and right of fixation. Attention was actively directed away from these face stimuli toward a demanding perceptual judgment task. Participants had to monitor a pair of vertical lines presented bilaterally close to fixation in order to decide on each trial whether the two lines were identical or different in length and to make a key press whenever they differed in length (10% of trials). Faces had to be ignored. Emotional faces appeared infrequently (10% fearful-neutral face pairs and 10% happy-neutral face pairs, against 80% neutral-neutral face pairs) and proportions of infrequent to frequent stimuli were held constant across target and non-target trials. Statistical analyses were performed on non-target trials only, in order to avoid motor artefacts generated by key presses. The number of neutral-neutral trials submitted for analysis was matched to the number of fearful-neutral and happy-neutral trials to help ensure equality of variances.

Fifteen participants took part in the experiment, and the following ERPs were analysed: vMMN (120-220 ms – posterior electrodes) to assess featural change detection of emotional faces; early frontal positivity (250-350 ms – frontal sites), late positive potential (LPP: 350-500 ms; 500-700 ms – frontal and posterior sites), and early posterior negativity (EPN: 250-350 ms – lateral posterior electrodes) to assess the standard emotional expression ERP components that reflect the processing of higher level affective properties of stimuli. The results showed, as predicted, that there were no emotional facial expression ERPs for happy-neutral or fearful-neutral faces compared with neutral-neutral faces. However, a vMMN was present for happy-neutral face pairs, although not for fearful-neutral face pairs. It would appear therefore that a feature change was detected for happy faces presented outside of spatial attention, but there was no equivalent access to featural changes for fearful faces.

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A second experiment was undertaken to verify that the emotional facial stimuli used in the first experiment were capable of eliciting emotional expression ERP effects. In this experiment, participants were required to focus their attention on the faces and to ignore the lines. Seventeen participants were told to make a key press whenever the faces differed in size. All other aspects of the design were identical to Experiment 1. As predicted, the results showed traditional patterns of ERP modulations to both happy and fearful face stimuli, with an enhanced LPP (500-700 ms) at frontal and posterior sites and also an EPN (250-350 ms) for both types of facial emotion.

The findings replicate previous results in demonstrating an elimination of emotional expression effects for faces presented outside of the focus of spatial attention. Despite this, there was evidence of featural change detection evoked by non-attended happy facial expressions, although not by fearful facial expressions. A vMMN in response to happy faces arose possibly because the mouth of happy faces is a highly salient visual feature, and argued to be responsible for a happy face recognition advantage (Calvo, Nummenmaa, & Avero, in press). On the basis of our results, we might conclude that the 'pre-attentive' encoding of featural change detection may arise only for happy faces and not for other less featurally salient facial expressions. The salience of happy expressions may have arisen in evolutionary terms because of their functional significance in instigating and maintaining social interactions and bonds. Alternatively, the enhanced recognition of happy expressions may be due to their being more familiar in everyday social encounters (see Calvo et al., in press). However, the absence of higher level facial expression ERP effects for non-attended happy faces that would appear to have undergone featural processing might suggest that spatial attention is critical for the processing of higher level affective meaning, in addition to being important for the low level processing of some (e.g., fearful) facial expressions. Further research is needed to determine the extent to which other facial expressions falling outside of focal spatial attention may be analysed in terms of their basic physical or featural characteristics.

In conclusion, these findings suggest that spatial attention is involved in the direct modulation of non-perceptual processes relating to the evaluation of emotional significance. They contribute to research into the interactions between attention and emotion, and hopefully extend our understanding of the limits to automaticity for the processing of emotional facial expressions.

Phase III

Due to the Stirling/Euro exchange rate, a small part of the research fund was unspent at the end of the grant period. We requested that this money be used to carryout some pilot work using facial electromyography (fEMG). In this pilot work, we used the same adaptation stimuli as used in the adaptation paradigm outlined in Phase II above. This research has been extremely useful in revealing the adaptation paradigm to be a useful technique for manipulating an individual's perception of emotionally ambiguous expressions. We plan to further this pilot work by looking at the possibility of recording micro-facial expressions during EEG recordings. Support from the BIAL for this proposed research has been of fundamental importance, and we hope to report interesting results following this pilot work.

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