Note

The role of human basolateral amygdala in ambiguous social threat perception

Beatrice de Gelder\textsuperscript{a,b,c,*}, David Terburg\textsuperscript{d,e,1}, Barak Morgan\textsuperscript{f,1}, Ruud Hortensius\textsuperscript{b}, Dan J. Stein\textsuperscript{e} and Jack van Honk\textsuperscript{d,e}

\textsuperscript{a}Department of Psychology and Neuroscience, Maastricht University, The Netherlands
\textsuperscript{b}Cognitive and Affective Neuroscience Laboratory, Tilburg University, The Netherlands
\textsuperscript{c}Brain and Emotion Laboratory Leuven, Department of Neurosciences, Leuven University, Belgium
\textsuperscript{d}Experimental Psychology, Utrecht University, The Netherlands
\textsuperscript{e}Department of Psychiatry and Mental Health, University of Cape Town, South Africa
\textsuperscript{f}MRC Medical Imaging Research Unit, Department of Human Biology, University of Cape Town, South Africa

\textbf{A B S T R A C T}

Previous studies have shown that the amygdala (AMG) plays a role in how affective signals are processed. Animal research has allowed this role to be better understood and has assigned to the basolateral amygdala (BLA) an important role in threat perception. Here we show that, when passively exposed to bodily threat signals during a facial expressions recognition task, humans with bilateral BLA damage but with a functional central-medial amygdala (CMA) have a profound deficit in ignoring task-irrelevant bodily threat signals.

1. Introduction

It is a common experience that an angry face feels more menacing when accompanied by a pair of fists, but it is rather unsettling when the fists come with a smile. In that case we experience the overall signal as profoundly ambiguous. When instructed to attend to only the facial expression, the brain notices the conflict between the facial expression and the accompanying bodily expression in a matter of milliseconds (Meeren, van Heijnsbergen, & de Gelder, 2005).

A variety of functions related to affective processes have been attributed to the amygdala (AMG) including immediate perception of affective stimuli, learning and conditioning, as
well as emotional memory (Phelps & LeDoux, 2005). The AMG is also involved in modulating behavioral responses and has multiple connections to brain areas directly involved in behavioral output (Mosher, Zimmerman, & Gothard, 2010). There is also overwhelming evidence that the AMG plays an important role in regulating emotion perception and preparing adapted motor behavior (Phelps & LeDoux, 2005).

Previous research has shown that the AMG plays an important role in face (Costafreda, Brammer, David, & Fu, 2008) and body (de Gelder, Snyder, Greve, Gerard, & Hadjikhani, 2004) expression recognition and is also highly sensitive to ambiguous signals (Kim et al., 2004; Whalen, 1998). But further progress in understanding the AMG will require understanding the specific contribution of the multiple nuclei of the AMG. Functions or loss of functions ascribed to the AMG as a whole may in fact result from activation of AMG nuclei or inter- and intra-amygdala connectivity. For example, facial expression recognition has been attributed to the AMG as a whole (Rutishauser et al., 2011) and consequently it was assumed that AMG damage abolishes this (Adolphs, Tranel, Damasio, & Damasio, 1994, but see Tsuchiya, Moradi, Felsen, Yamazaki, & Adolphs, 2009). But more recently it was shown that an impairment of one of the AMG nuclei, the basolateral amygdala (BLA), leads to hypersensitivity for facial fear expressions (Terburg et al., 2012).

Similarly, the same complete AMG impairment does not seem to abolish body expression recognition (Atkinson, Heberlein, & Adolphs, 2007). This finding does not rule out that an impairment of a specific nucleus of the AMG does nevertheless have consequences for normal processing of body expressions. In the case of a complex structure like the AMG a functional role attributed to the AMG as a whole cannot be attributed automatically to each of its subnuclei.

We addressed the issue of the functional role of the BLA in ambiguous social threat perception using subjects with Urbach–Wiethe disease (UWD), a rare genetic disorder that in our sample has resulted in bilateral focal calcification of the BLA. We tested three subjects from the South African UWD cohort (Thornton et al., 2008) selected for this specific BLA damage (Morgan, Terburg, Thornton, Stein, & van Honk, 2012) and a group of matched controls on a series of face and body expression recognition tasks. Our goal was first, to investigate the specific role of the BLA in implicit bodily expression recognition and second, the role of the BLA in ambiguity perception. We used angry and fearful Face Body Compounds created by combining a facial expression with either a congruent or incongruent bodily expression. Using convergent evidence from behavior and eye tracking measures, we investigated how BLA damage affects the processing of affective information from body expressions of anger and fear that are, unattended, not task relevant, and presented in the periphery. We conjectured that under these conditions of implicit perception participants with BLA damage would still process the threatening body signals and these signals would be more salient than in normal controls. Therefore we expect an increased effect from threatening bodily expressions on facial expression perception in the UWD group.

### 2. Methods

#### 2.1. Subjects

Three subjects from the South African UWD cohort (Morgan et al., 2012; Thornton et al., 2008) without any history of secondary psychopathology or epileptic insults and 12 matched controls participated in the experiment. The UWD and control group were all female and matched for age and IQ (see Table 1 for demographic data). All participants were from mountain-desert villages near the Namibian border. Detailed neuropsychological assessment of the UWD group is described elsewhere (Morgan et al., 2012; Terburg et al., 2012). Structural and functional MRI assessment by means of cytoarchitectonic-probability labeling showed that bilateral calcification is restricted to the BLA (see Fig. 1). This study was approved by the Health Sciences Faculty Human Research Ethics Committee of the University of Cape Town. All participants provided written informed consent. We note that all UWD subjects reported here and in previous studies (Adolphs et al., 1994; Morgan et al., 2012; Terburg et al., 2012) are female and we cannot exclude that gender colors past and present results. Resolution of this issue must await availability of male UWD subjects.

#### 2.2. Tasks

##### 2.2.1. Face Body Compound task (FBC)

Congruent and incongruent threatening FBCs (Meeren et al., 2005) were constructed using angry and fearful bodies (de Gelder & Van den Stock, 2011) and angry and fearful faces (MacBrain Face Stimulus Set) (see Fig. 2A). Stimuli (12 per condition, 6 female) were on screen for 350 msec for behavioral testing and 2000 msec for eye tracking. Participants had to recognize the facial expression and ignore the bodily expression while accuracy and reaction time were recorded during behavioral testing.

##### 2.2.2. Sample-to-match task

The Bodily Expressive Action Stimulus Test (BEAST) (de Gelder & Van den Stock, 2011) was used to assess the perception of emotional whole bodily expression. Participants had to match angry, happy, fearful or sad bodily expressions with one of two simultaneously presented bodily expressions (12 per condition, 6 female). Both the target and distracter had different identities, while the distracter had a different

<table>
<thead>
<tr>
<th>Table 1 – Demographic data.</th>
<th>UWDs</th>
<th>Controls</th>
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<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>Mean</td>
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<tr>
<td></td>
<td>UWD 1</td>
<td>UWD 2</td>
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<tr>
<td>Age</td>
<td>24</td>
<td>31</td>
</tr>
<tr>
<td>VIQ</td>
<td>95</td>
<td>84</td>
</tr>
<tr>
<td>PIQ</td>
<td>98</td>
<td>86</td>
</tr>
<tr>
<td>FSIQ</td>
<td>97</td>
<td>84</td>
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VIQ: verbal IQ, PIQ: performance IQ, FSIQ: full-scale IQ. Means and standard deviations are reported.
emotional expression. Stimuli were presented on screen until response while reaction time and accuracy were recorded.

2.2.3. Three-alternative forced choice task
In a three-alternative forced choice task (3AFC) participants indicated if the expressed emotion of the presented body was angry, happy or fearful. Stimuli were on screen for either 350 msec for behavioral testing or 2000 msec for eye tracking. Stimuli (12 per condition, 6 female) were from the same stimulus database as used in the sample-to-match task, but with different actors. Accuracy was measured for both durations, while reaction time was recorded for the 350 msec task.

2.2.4. Flanker task
A modified Erikson flanker task was used to test for interference of non social-emotional information. The task was similar to the one described by Cavanagh and Allen (2008), and involved 300 trials requiring the participants to identify a middle target letter flanked by 4 distractors. Half of the targets were flanked by the same letters (e.g., XXXXX, congruent trial), and half were flanked by a different letter (e.g., XXYXX, incongruent trial). A trial consisted of a blank screen (100 msec), a fixation screen (700 msec), a flanker screen (e.g., XX XX, 135 msec), a target screen (e.g., XXXXX, 265 msec), and a fixation cross screen (600 msec). Participants had to indicate as fast as possible what the middle target letter was by pressing the correct button (2AFC, left-hand or right-hand button). Participants received feedback only after incorrect trials in which the word ‘VERKEERD’ (Afrikaans for ‘wrong’) was presented. When participants did not respond within 1000 msec after flanker screen presentation feedback was given by presenting the word ‘TE STADIG’ (Afrikaans for ‘too slow’). The flanker screen preceded the target screen to increase task difficulty (Cavanagh & Allen, 2008). The task was divided in 10 blocks of 30 trials. Within each block 2 target/distractor letters were used, and each target letter was assigned to either the left or right-hand button counter-balanced across blocks (i.e., MN, NM, FE, EF, QO, OQ, VU, UV, TI, IT). Participants first practiced the task using the letter combinations XY and YX, and preceding each block the new target/button combinations were explained. The blocks only commenced after the participants correctly identified the target/button combinations. Accuracy and reaction times were recorded for all trials.

2.3. Procedure
Session-1 started with the 3AFC task followed by the FBC task. For both tasks the eye tracking session was presented first to ensure eye-movements were not biased by previous exposure.
to the stimuli. The sample-to-match task was done in session-2. Instructions were presented in Afrikaans. The flanker task was conducted two years earlier than the other experiments, but the same three UWD subjects participated together with ten age (29.9 years old, \(SD = 5.8\)) and IQ (VIQ: 85.9, \(SD = 4.7\); PIQ: 84.8, \(SD = 8.0\), FSIQ: 83.7, \(SD = 6.1\)) matched controls living in the same area as the UWD subjects.

2.4. Data analysis

Reaction times <150 msec and >2 SD of the subject’s mean were removed from the analysis. Incongruence scores were calculated by subtracting recognition accuracy for congruent from incongruent FBCs for each facial expression (FBC task) and by subtracting average accuracy and reaction time for congruent from incongruent trials (Flanker task). Group differences were tested cell-by-cell with two-tailed non-parametric Mann–Whitney U tests.

2.5. Eye tracking

The tasks were presented and eye-movements recorded with a Tobii-1750 eye tracker, sampling at 50 Hz, with .5° accuracy. Trials only commenced when subjects fixated gaze somewhere in a rectangle with the exact size and position of the stimuli, which ensures valid eye tracking data without biasing fixation positions. Gaze-fixations were defined as the average location of all subsequent gaze points within 1° visual angle, with a minimal duration of 100 msec (Tobii Technology, Danderyd, Sweden). Gaze-fixations were mapped onto a priori determined areas of interest (AOI); face, torso, arms, hands and legs. For each AOI mean fixation duration (FD) and proportion of number of fixations relative to all fixations (NF) was computed and used for further analysis.

3. Results

In line with previous results (Terburg et al., 2012), UWD participants performed as well as the controls in recognizing congruent fearful and angry Face Body Compounds (U = 7, \(p = .14\) and U = 6, \(p = .13\), respectively). However, recognition of fearful (U = 1.5, \(p = .02\), \(r = -.62\)) and angry faces (U = 5, \(p = .06\), \(r = - .49\)) was impaired when a face was combined with an incongruent bodily expression (see Fig. 2B). This incongruence effect was significantly larger in the UWD group compared to the control group for fearful as well as for angry faces, U = .5, \(p = .007\), \(r = -.66\) and U = 2.5, \(p = .02\), \(r = -.58\) respectively (see Fig. 2C). No differences in reaction times were found between the UWD and control group (p’s < .37).

Importantly, the groups were not different in gaze behavior. Consistent with the task instructions, both the UWD and control group predominantly looked at the faces (76%
vs 65%) (see Fig. 2D), which rules out a possible attention deficit underlying the current finding.

As revealed by the control experiments, recognition of bodily expressions, including fear, was intact in the UWD group. No differences in accuracy and reaction time were found in recognizing whole body expressions in either a sample-to-match ($p' > .14$) or the 3AFC task (long stimulus duration; $p' > .18$, short stimulus duration; $p' > .18$, see Fig. 3). Eye tracking data also showed no abnormalities in terms of gaze duration or fixation patterns. No group differences were found on the eye-track measures NF or FD in the 3AFC or in the congruent and incongruent conditions of the FBC task (all $p' > .1$, see also Fig. 2D). Crucially, visual attention in the FBC task was predominantly directed to the face compared to the other AOI’s on both measures (all $p' < .05$), and compared to the 3AFC task, visual attention in the FBC task was longer and more often directed to the face part of the stimuli (all $p' < .001$). These results confirm that the subjects’ visual attention was increasingly directed to the faces when they were asked to judge the facial emotion, and

Fig. 3 – Recognition accuracy and reaction times for the sample-to-match task (top), the 3AFC task with 2000 msec (middle) and 350 msec stimulus duration (bottom). No differences were found between the UWD and control group.
that this was not different for UWD subjects and the controls. Furthermore, no significant correlation between recognition accuracy on congruent and incongruent FBC trials were found for both fearful ($r_{\text{ho}} (15) = -0.36, p = 0.19$) and angry faces ($r_{\text{ho}} (15) = -0.12, p = 0.68$), which confirms that task instructions to recognize the facial emotion were followed.

Average reaction times and error-rates in the flanker task are summarized in Table 2. As expected the error-rate was significantly ($Z = -2.8, p = .005$) higher, and reaction time significantly ($Z = -3.1, p = .002$) slower, in the incongruent compared to congruent trials. The congruency effect on error-rate was not significantly ($U = 11, p = .57$) different between groups, which was also the case when error-rates were tested separately for congruent ($U = 12, p = .69$) and incongruent ($U = 12, p = .69$) conditions. The congruency effect on reaction time was also not significantly ($U = 6, p = .16$) different between groups, which was also the case when tested separately for congruent ($U = 15, p = 1$) and incongruent ($U = 10, p = .47$) conditions. In sum, the flankers successfully evoked interference, but this was not different between the UWD and control groups.

### 4. Discussion

Our main result shows a strong and selective effect of unattended body signals on facial expression recognition in UWD subjects. This means that BLA damage leads to a stronger interference in a simple facial expression recognition task when the target face is paired with a bodily expression shown in the periphery that is not attended to. In other words, ignoring the role of the task-irrelevant stimulus part appears harder for UWD subjects and their impaired facial expression judgments reflect this. Importantly, this effect was obtained in the UWD group while their gaze behavior was not different from that of the controls. The results from the flanker task show that this effect is specific for affective stimuli and thereby underscore that the AMG is at the core of this process. Future research needs to clarify whether these data reflect heightened threat value of bodily expressions, heightened sensitivity to ambiguous affective signals or a combination of both.

Previous studies of a UWD subject have reported that AMG impairment has only implications for facial (Adolphs et al., 1994, but see Tsuchiya et al., 2009) and not for bodily expressions (Atkinson et al., 2007). The strong effect of bodily expressions on facial expression recognition seen here indicates that as in the case of faces, the AMG is important for processing affective expressions of bodies and this is consistent with similar AMG activation to facial and bodily expressions in normal subjects (de Gelder et al., 2004; de Gelder, Hortensius, & Tamietto, 2012). Fearful bodily expressions are known to trigger automatic action preparation (de Gelder et al., 2004) and the AMG is known to play a role in triggering adaptive emotional reactions (Pessoa, 2010). Rather than illustrating that the AMG is not needed for body expression processing, the present findings are compatible with a role of the BLA in generating hypervigilance for threat signals as previously argued for fearful facial expressions (Terburg et al., 2012). Conversely, our results show that the ambiguity of the emotional signal created by incongruent Face Body Compounds is fully noticed by the UWD subjects. The effects of this stimulus ambiguity and thus of the threatening body expressions are much stronger in the UWD subjects than in the controls. This excessive effect of ambiguity is also consistent with earlier studies on the role of the AMG in ambiguous decision-making (Brand, Grabenhorst, Starcke, Vandekerckhove, & Markowitz, 2007), ambiguous affect perception (Kim et al., 2004; Whalen, 1998) and conflict resolution (Etkin, Egner, Peraza, Kandel, & Hirsch, 2006; Etkin et al., 2004).

There are thus two possible explanations for the finding of more ambiguity sensitivity in the UWD group. The body expressions have a stronger impact because in the absence of the BLA, they are experienced as more salient. This may be for example because the central-medial amygdala (CMA) is not controlled by the BLA and affective signals are over-represented, similar to the explanation of hypervigilance in Terburg et al. (2012). Furthermore, it may be that without BLA there is a heightened sensitivity to the ambiguous meaning of the compound stimuli. The latter presupposes that at the level of perception of the face and of the body expressions there is no difference between the UWD subjects and controls but that the difference is generated once the face and the body percept are combined in later integration processing stages. One may then argue that the BLA deficit creates this heightened sensitivity to ambiguity.

While there were no significant differences in terms of gaze duration or fixation, indicating that both the UWD and control group followed task instructions and looked at the faces, we cannot discard the possibility that there might be small differences in gaze behavior (e.g., scan-paths) between groups that would underlie the found effect. Thus, the UWD group could still pay more attention to the irrelevant bodies. However, while not significant the number of fixations on the face was higher in the UWD group compared with the control group. This is in line with the notion that the distracting threatening body has a stronger impact or heightened sensitivity to the ambiguous threat signal due to BLA deficits.

The present report represents therefore significant theoretical and methodological advances. Methodologically, the present cases are unique in the AMG literature because they have focal damage involving only the basolateral nucleus. Our study combines behavioral methods with spontaneous eye movement recordings. Finally, we use facial expressions but also whole body expressions and face and body combinations that make for highly ambiguous signals and we use implicit perception measures.

### Table 2 – Average reaction times and error-rates (with their range) in the flanker task.

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<thead>
<tr>
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<th>UWD</th>
<th>Control</th>
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<tbody>
<tr>
<td>Average reaction time (ms)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Congruent trials</td>
<td>430 (387–493)</td>
<td>421 (366–532)</td>
</tr>
<tr>
<td>Incongruent trials</td>
<td>443 (401–523)</td>
<td>455 (405–562)</td>
</tr>
<tr>
<td>Average error-rate (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Congruent trials</td>
<td>15 (4–25)</td>
<td>11 (8–19)</td>
</tr>
<tr>
<td>Incongruent trials</td>
<td>18 (9–25)</td>
<td>17 (8–24)</td>
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To conclude, we show that the BLA is important for processing ambiguous social information. In addition to focusing on specific emotions (e.g., fear) or specific categories (e.g., facial expressions) or specific attributes of emotional stimuli (e.g., salience), AMG research would benefit from concentrating on the interplay between the affective context, the different AMG subnuclei, and their connectivity.

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Conflict of interest

None.

References


