Brain activation and defensive response mobilization during sustained exposure to phobia-related and other affective pictures in spider phobia

JULIA WENDT,[a] MARTIN LOTZE,[b] ALMUT I. WEIKE,[a] NORBERT HOSTEN,[c] AND ALFONS O. HAMM[a]

[a]Department of Psychology, University of Greifswald, Greifswald, Germany
[b]Functional Imaging Unit, Department for Diagnostic Radiology and Neuroradiology, University of Greifswald, Greifswald, Germany
[c]Department for Diagnostic Radiology and Neuroradiology, University of Greifswald, Greifswald, Germany

Abstract

This study explored defensive response mobilization as well as fMRI responses during sustained exposure to phobia-relevant stimuli. To test the specificity of affective physiology and brain activation, neutral and other affective stimuli were included. Phobia-specific startle potentiation was maintained and autonomic responses even increased during sustained phobic stimulation. Viewing of spider pictures also resulted in increased activation of the amygdala in spider-phobic participants. This effect, however, was not fear specific because other affective materials evoked comparable signal strength in the amygdala. In contrast, insula activation was specifically increased during sustained phobic exposure in phobic volunteers. These data suggest that the activation of the amygdala in fMRI studies primarily indexes the detection of motivationally relevant stimuli whereas the insula might be more specifically linked to defensive response mobilization.

Descriptors: Spider phobia, Amygdala, Insula, fMRI, Startle

Fear is an aversive emotional state elicited by threatening cues that activate the organism's defense system. In people with specific phobias this defense system seems to be overreactive, as it is easily activated even by symbolic representations of their feared objects. Accordingly, when individuals with animal phobia are confronted with pictures of their feared animals, they exhibit marked sympathetic activations, as indexed by a rapid increase in heart rate, skin conductance, and blood pressure (Fredrikson, 1981; Globisch, Hamm, Esteves, & Öhman, 1999; Hamm, Cuthbert, Globisch, & Vaitl, 1997). Moreover, people with specific phobia also exhibit a significant potentiation of the startle reflex when exposed to pictures of their feared objects (Globisch et al., 1999; Hamm et al., 1997; Sabatinelli, Bradley, & Lang, 2001). This phobia-specific startle potentiation can be observed for different types of phobia and is also fear specific, that is, more pronounced than startle facilitation evoked by other standard unpleasant materials (Hamm et al., 1997). In the current experiment we investigated whether defensive response mobilization would be maintained during sustained exposure to phobic material as would be suggested by several studies that examined startle

modulation during sustained exposure to standard affective pictures (Bradley, Cuthbert, & Lang, 1996; Smith, Bradley, & Lang, 2005; Sutton, Davidson, Donzella, Irwin, & Dottl, 1997). Substantial evidence suggests that such defensive response mobilization as indexed by startle potentiation is mediated by the amygdala. Lesions of the central nucleus of the amygdala completely block startle potentiation after fear conditioning in rodents (Davis, 2000), and unilateral amygdala damage impair fear potentiated startle in humans as well as startle facilitation during viewing of highly arousing unpleasant pictures (Buchanan, Tranel, & Adolphs, 2004; Funayama, Grillon, Davis, & Phelps, 2001; Weike et al., 2005). Hence, many theorists assume that the amygdala is the core structure involved in fear learning and activation of fear and phobia as well (LeDoux, 1996; Öhman & Mineka, 2001; Rosen & Schulkin, 1998). Indeed, recent studies exploring the hemodynamic responses of the human brain during viewing of phobia-related stimuli showed that animal phobics respond with increased amygdala activation when confronted with pictures of their feared animals (Carlsson et al., 2004; Dilger et al., 2003; Fredrikson & Furmark, 2003; Sabatinelli, Bradley, Fitzsimmons, & Lang, 2005; Schienle, Schäfer, Walter, Stark, & Vaitl, 2005; Straube, Mentzel, & Miltner, 2006). Whether this increased activation of the amygdala in the fMRI studies indexes increased aversive response mobilization or increased selective attention during encoding of these motivationally relevant stimuli—at least for phobia patients—is currently under debate (Pessoa, Japee, Sturman, & Ungerleider,

2006; Davis & Whalen, 2001). Several studies suggest that pictures of emotional faces reliably elicit increased signal strength in the amygdala in the absence of any defensive response mobilization as indexed by autonomic arousal or verbal report (Adolphs & Spezio, 2006; Duncan & Barrett, 2007; Morris, Öhman, & Dolan, 1998; Whalen et al., 1998). Animal data clearly show that the amygdala comprises many subnuclei, with the basolateral nucleus serving as input region and the central nucleus involved in organizing defensive response mobilization (Davis, 2000; Maren & Quirk, 2004). The current imaging technology cannot yet discriminate these individual subregions within the amygdala in a reasonable time resolution. Thus it is not clear whether increased amygdala activation during phobic stimulation indexes increased attention to these stimuli (increased activity of the input region) or augmented phobia-specific defensive response mobilization. One way to disentangle both processes might be to include motivationally significant but appetitive stimuli that do not evoke any defensive responses. Unfortunately, there is only one study so far that explicitly compared the brain activation in the amygdala evoked by phobic and pleasant arousing pictures. In this study increased activation of the amygdala was observed for both phobia-relevant and erotic pictures in individuals with snake phobia with no difference between pleasant and phobic pictures (Sabatinelli et al., 2005). The present study followed up on this research by not only comparing the brain responses to phobia-relevant and neutral materials but also including arousing pleasant and unpleasant materials.

Another integral part of the affective processing network is the insular cortex (Carlsson et al., 2004; Critchley, 2005). Increased activation of the insula was repeatedly reported during processing of phobia-relevant relative to neutral stimuli in phobic individuals (Carlsson et al., 2004; Dilger et al., 2003; Rauch et al., 1995; Straube, Glauer, Dilger, Mentzel, & Miltner, 2006; Straube, Mentzel, et al., 2006). Moreover, representation of visceral sensations (Craig, 2002), pain, and temperature perception (Davis, Kwan, Crawley, & Mikulis, 1998; Peyron, Laurent, & Garcia-Larrea, 2000) converge in the insula and the surrounding operculum, suggesting that especially the insula is involved in the representation of the internal body state and interoception. Thus, it has been suggested that the insula is involved in integrating the emotionally relevant information from the environment (possibly mediated via strong connections from and to the amygdala; Reynolds & Zahm, 2005; Shi & Cassell, 1998) with the effects that these stimuli may have on the body state (Paulus & Stein, 2006). Accordingly, Anders, Lotze, Erb, Grodd, and Birbaumer (2004) found a positive correlation between reported valence of evoked feeling states and the activity of the insular cortex. In the same vein, Critchley, Wiens, Rotshtein, Öhman, and Dolan (2004) demonstrated that accuracy in a heart beat detection task was positively related to the activity in the right anterior insular cortex. This interoceptive accuracy and insular activation was also correlated with anxiety symptoms in these normal volunteers, as assessed by the Hamilton anxiety scale. Moreover, anticipation of aversive events was also associated with increased activation of the anterior insular cortex during aversive conditioning (Büchel, Morris, Dolan, & Friston, 1998), and during the anticipation of aversive pictures (Nitschke, Sarinopoulos, Mackiewicz, Schaefer, & Davidson, 2006). This activation of the insular cortex during anticipation of aversive pictures was stronger for anxiety-prone participants (Simmons, Strigo, Mathews, Paulus, & Stein, 2006). Using an instructed fear paradigm in which

participants were informed that they would receive an electric shock during the threat condition but not in the safe condition, Phelps and collaborators (2001) also found a pronounced activation of the anterior insular cortex. Interestingly, although the stronger activation of the amygdala was attenuated within the 18-s threat condition, the extensive and strong activation in the left insula and the somewhat more limited activation of the right insula were maintained during the 18-s threat condition. Accordingly, studies that used sustained exposure to phobic stimuli (e.g., film clips of crawling spiders) did not always find increased amygdala activation during phobic stimulation but strong and extensive activation of the insular cortex (Rauch et al., 1995; Straube, Glauer, et al., 2006). Moreover, exposure therapy mainly reduces the insular activation, suggesting that the intensity of the phobic fear and anxiety as well as the experienced distress might be mediated by the anterior insular cortex (Straube, Glauer, et al., 2006). Although there is increasing evidence of the broader role of the insula during processing of aversive emotional stimuli (Phan, Wager, Taylor, & Liberzon, 2002; Schienle et al., 2002), it is unclear whether such activation of this brain structure is specific for aversive or even phobic stimulation. The only study that compared brain activation during processing of phobia-relevant and pleasant arousing stimuli (Sabatinelli et al., 2005) has not included the insular cortex as a region of interest. Therefore, the present study was designed to investigate three questions: (1) We explored whether a sustained exposure to phobia-relevant stimuli would elicit a continuous defensive response mobilization in people with specific phobia as indexed by a phobia-specific startle potentiation and prolonged increased autonomic arousal. We expected a phobia startle potentiation that would be maintained during sustained exposure to phobia-relevant stimuli. (2) We investigated whether increased activation of the amygdala during phobia-relevant compared to neutral stimuli would be fear specific or would rather be independent of defensive response mobilization, thus primarily indexing the detection of motivationally salient cues from the environment. In this case amygdala activation should be comparable for phobia-relevant and arousing pleasant pictures, as suggested by the findings of Sabatinelli et al. (2005). (3) Finally, we explored whether the activation of the insula would be selectively increased during processing of phobia-relevant relative to other affective materials and whether this activation would be specific for participants with animal phobia. We expected increased activation in the insula during processing of phobia-relevant materials but only in the group of participants with specific phobia.

Methods

Participants

Thirty-two women were selected from a screening of 211 students of the University of Greifswald. The participants completed the German version of the Spider Phobia Questionnaire (SPQ; Hamm, 2006). Women were included if scores were either ≥ 17 in the SPQ (phobia group), that is, above the 85th percentile of the distribution, or ≤ 4 (control group), that is, below the lower 15th percentile of the SPQ. Sixteen participants with spider phobia (age range 19–31; $M = 23.1$) had a mean SPQ score of 19.8 (range: 17–23), while 16 other participants (age range 19–26; $M = 21.3$) with a mean SPQ score of 2.4 (range 0–4) served as

controls. Because of equipment failure, the fMRI data of 6 participants (3 controls) had to be excluded from further analysis; thus data of 26 participants (phobia group: $N = 13$, age range 19–31, $M = 23.2$; control group: $N = 13$, age range 19–26, $M = 21.1$) were included in the preprocessing and statistical analyses of the fMRI data.

Stimulus Materials and Procedure

Participants viewed 150 color pictures mainly selected from the International Affective Picture System (Lang, Bradley, & Cuthbert, 2005).[1] The pictures were derived from five different categories, which were spiders (phobia relevant), mushrooms (neutral pictures for the contrasts to spiders), pleasant contents (babies, young animals, erotica), unpleasant contents (animal attack, mutilations, human terror), and complex neutral pictures (buildings, neutral people, cars).[2] Each category included 30 pictures that were grouped into blocks of 10 pictures each. Each picture was shown for 3 s; thus, each block lasted for 30 s. During the 15-s interblock intervals a black screen with a white fixation cross was presented.

The experiment was divided into two sessions. During the first session participants were exposed to three semirandomly presented blocks of each of the five picture categories in a 1.5 T magnetic resonance scanner (Siemens Magnetom Symphony). The pictures were back-projected onto a translucent screen and were viewed by participants through a mirror affixed at the head coil. Prior to scanning, the participants were instructed to watch the pictures attentively and not to close their eyes even if some of the pictures might be very unpleasant. Ratings of the subjective experience of valence and arousal induced by each picture were obtained immediately after the scanning session outside of the scanner using the Self Assessment Manikin Scale (Bradley & Lang, 1994).

Finally, all participants viewed the pictures in a second session assessing psychophysiological response patterns. For one group of participants ($N = 12$), the time between the first and the second session ranged from 2 days to 20 days; for the other participants

the time lag ranged between 6 and 10 months.[3] The experimental procedure was identical to the fMRI scanning session except that the pictures were presented on a white wall approximately 2 m in front of the participants, who were exposed to six semirandomly presented blocks of each of the five picture categories. Additionally, an acoustic startle eliciting stimulus (100 dB[A]) was presented binaurally through headphones at defined time windows at the beginning and the end of each block as well as in the interblock intervals.[4] All participants gave their written informed consent before participation, and the study was approved by the Ethics committee of the University of Greifswald.

Apparatus and Data Acquisition

Physiological recordings. Recordings of electromyographic (EMG) activity over the left orbicularis oculi muscle served to measure the eyeblink component of the startle response. Two Ag/AgCl miniature surface electrodes (Sensormedics) filled with electrolyte were attached beneath the left eye. The raw EMG signal was amplified using a Coulbourn S75-01 bioamplifier with a 30-Hz high-pass filter and a 400-Hz Kemo KEM-VBF8-03 low-pass filter. Digital sampling was set to 1000 Hz from 100 ms prior to the onset of the startle probe until 400 ms after it. The EMG signal was filtered off-line through a 60-Hz high-pass filter and was rectified and integrated (time constant: 10 ms) using a digital filter. Corrugator EMG activity was recorded above the left eye with two miniature Ag/AgCl surface electrodes (Sensormedics), using the placement recommended by Fridlund and Cacioppo (1986). Amplification and filtering of the raw EMG was identical to that described for the orbicularis oculi. The signal was rectified and integrated using a Coulbourn S76-01 contour-following integrator with a time constant set at 500 ms. The integrated signal was continuously sampled with a rate of 10 Hz.

Skin conductance was recorded at the hypothenar eminence of the participant's right hand, using two adjacently placed Hellige AG/AgCl standard surface electrodes (8 mm diameter) filled with a 0.05 M sodium chloride electrolyte medium (Venables & Christie, 1980). A Coulbourn S71-22 skin conductance coupler that processed the signal with a resolution of 0.001 μS provided a constant 0.5 V across electrodes. The analog signal was continuously sampled at a rate of 10 Hz. The electrocardiogram (Eindhoven Lead II) was measured using two Hellige AG/AgCl standard electrodes (8 mm diameter). R waves were identified by means of a custom-made heart rate trigger (modified according to Shimizu, 1978), and the analog signal was continuously sampled at a rate of 100 Hz.

fMRI data. MRI data were collected using a 1.5 Tesla Magnetom Symphony system (Siemens) that was additionally equipped with an 8-channel headcoil. Field homogeneity was optimized prior to each session by using a shimming sequence. Then a T1-weighted anatomical volume (TR 368 ms, TE 4.88 ms, flip angle 40°, FoV 192 mm, matrix 256 × 256,

---

[1]The International Affective Picture System identification numbers are the following: 1440, 1441, 1463, 1710, 1722, 2040, 2050, 2057, 2058, 2070, 2080, 2153, 2260, 2332, 2341, 4650, 4651, 4652, 4653, 4658, 4659, 4660, 4670, 4672, 4680, 4681, 4689, 4690, 4694, 4695 (pleasant pictures); 1300, 1302, 1303, 1321, 1930, 2811, 3000, 3010, 3015, 3030, 3051, 3053, 3060, 3061, 3062, 3168, 3170, 3191, 3225, 3266, 3400, 6022, 6550, 9040, 9254, 9400, 9404, 9410, 9420, 9426 (unpleasant pictures); 2102, 2221, 2383, 2396, 2515, 2530, 5250, 5635, 5820, 5900, 7130, 7140, 7491, 7500, 7510, 7546, 7547, 7550, 7595, 7700 (neutral pictures); 1201, 1205, 1220, 1230, 1240 (spider pictures); 5510, 5520, 5534 (mushroom pictures). Further spider and mushroom pictures were selected from picture sets that were successfully utilized in previous studies (e.g., Globisch et al., 1999).

[2]Pictures of mushrooms were included as a neutral category because in most fMRI studies with animal phobics contrasts are calculated between BOLD responses to phobia-related pictures (snakes or spiders) and those elicited by mushrooms. Mushrooms serve as a good control condition for this contrast because (1) there is a higher homogeneity of objects within both categories relative to the pleasant and unpleasant pictures and (2) no differentiation between human scenes and nonhuman objects is necessary. Such differentiation is always implied for the rather heterogeneous pleasant and unpleasant category. Therefore, pictures depicting complex neutral scenes and neutral human people were included as an additional neutral category to calculate contrasts to the more heterogeneous pleasant and unpleasant categories. As expected, a whole-brain analysis revealed a significantly stronger activation of the fusiform gyrus for the heterogeneous neutral picture category compared to the mushroom category ($t = 7.05$, $p_{FDR} < .001$).

---

[3]Short- and long-time lag between sessions did not influence any of the peripheral measures that might count as further evidence for the stability of response output patterns to specific stimuli.

[4]Acoustic startle probes were randomly presented during the second or third picture of each block (i.e., either at 4, 4.5, 5, 7, 7.5, or 8 s after block onset) and during the eighth or ninth picture of the block (i.e., either at 22, 22.5, 23, 25, 25.5, or 26 s after block onset). Control startles were presented at the beginning (i.e., at 2, 2.5, or 3 s after block offset) and the end of the interblock- intervals (i.e., at 12, 12.2, or 13 s after block offset).

voxel size $0.75 \times 0.75 \times 3$ mm) was recorded. During the presentation of each block of pictures, 230 volumes with 33 slices each (3 mm thick, 0.75 mm gap) were acquired in the transversal direction parallel to the AC-PC line using echo-planar images (EPIs; TR 3000 ms, TE 50 ms, flip angle 90°, FoV 192 mm, matrix 64×64, voxel size $3 \times 3 \times 3$ mm).

Data Reduction and Analysis

The magnitude of the startle eyeblink was scored off-line using a computer algorithm (Globisch, Hamm, Schneider, & Vaitl, 1993) that automatically identified latency of blink onset and peak amplitude in microvolts. Only blinks starting 20–100 ms after probe onset and peaking within 150 ms were scored. No detectable blinks were scored as zero responses. Trials with excessive baseline activity or recording artifacts were rejected (157 out of 3780 trials, 4.2%). Prior to statistical data analyses all missing values were replaced individually for each participant by the individual overall mean blink magnitude for this participant across all 60 trials. To ensure that each participant contributed equally to the group's mean, blink magnitudes were standardized for each participant using a z score transformation. The standardized responses of each participant were then converted to T scores [$50 + (z \times 10)$]. Digital values of the corrugator EMG activity were converted to microvolts. Corrugator activity was scored by subtracting the 2-s preblock level from half second values and calculating the average change during each 3-s picture viewing period, resulting in 10 corrugator EMG change scores for the entire picture block. As for startle responses, raw EMG change scores were standardized within each participant and then converted to T scores.

Digital values of skin conductance were converted to micro-siemens. Skin conductance level (SCL) was scored by subtracting the 2-s preblock level from half second values and calculating the average SCL change during each 3-s picture viewing period, again resulting in 10 SCL change values for the entire block. Logarithms of these values were computed to normalize the distribution (Venables & Christie, 1980). To ensure that each participant contributed equally to the group's mean, the log values were range corrected as recommended by Lykken, Rose, Luther, and Maley (1966; [$SCL - SCL_{min}]/[SCL_{max} - SCL_{min}]$). Interbeat (R-R) intervals were converted to heart rate (beats per minute, bpm) in half-second bins. Baseline heart rate (2-s preblock) was subtracted from the heart rate for every half second during the 30-s picture blocks. Difference scores were again averaged across each 3-s picture presentation, resulting in 10 change scores per block. A mixed model analysis of variance (ANOVA) included the factor Group (phobics vs. controls) as a between-subjects factor and the factors Category (spiders vs. mushrooms vs. complex neutral vs. unpleasant vs. pleasant) and Time (10 levels during each block for corrugator, SCL, and HR responses and 2 levels representing early and late probe times for analysis of startle eyeblink magnitude) as within-subject factors.[5] Unless otherwise noted, all statistical tests concerning the

peripheral physiology data used the .05 level of significance and were accomplished with SPSS 12.0 (SPSS for Windows, SPSS Inc.). Greenhouse–Geisser adjustments of degrees of freedom were used to control for all effects involving repeated measures factors. Nominal degrees of freedom are reported along with epsilon values.

fMRI data. Preprocessing and statistical analyses were realized using the statistical parametric mapping software (SPM5, Wellcome Department of Imaging Neuroscience, London, UK). Preprocessing included spatial realignment, normalization into the MNI space, and spatial smoothing (FWHM 12 mm). To correct for low-frequency components, a high-pass filter with a cutoff of 1/90 Hz was used. Statistical analyses were performed using the general linear model as implemented in SPM5. For each participant a design matrix was created using a canonical hemodynamic response function for each of the five stimulus conditions. The six movement parameters estimated during the realignment procedure were introduced as covariates into the model. The resulting beta images were then taken to the second level full factorial model in SPM5. Exploratory analyses of the whole brain volume were computed with an intensity threshold of $p < .05$ corrected (false discovery rate, FDR; Genovese, Lazar, & Nichols, 2002) and with an extent threshold of 10 contiguous voxels. Additionally, a regions of interest (ROI) analysis for the amygdala and insula was conducted by using the anatomical masks of the "Automated Anatomical Labeling" software (Tzourio-Mazoyer et al., 2002). For whole-brain analysis as for region-of-interest analysis, phobia-relevant contrasts were calculated separately for both groups, whereas contrasts concerning the standard affective categories were calculated as overall analysis including all participants. For each stimulus condition, differences between phobic participants and the control group in evoked brain activation were computed.

Results

Physiological Responses

Startle response magnitudes. As expected, blink magnitudes were substantially modulated by the affective categories, Category $F(4,100) = 15.23$, $p < .001$, $\eta^2 = .38$. Although startle responses were potentiated during viewing of unpleasant pictures relative to neutral contents, $F(1,26) = 11.66$, $p < .01$, $\eta^2 = .31$, viewing of pleasant pictures relative to neutral pictures resulted in a marginally significant inhibition of the blink magnitudes, $F(1,26) = 3.23$, $p = .08$, $\eta^2 = .11$. Most importantly, this response pattern did not differ significantly between both groups for these standard affective categories, Category $\times$ Group $F(2,50) = 1.21$, n.s. In contrast, participants with animal phobia but not controls exhibited a significant potentiation of the startle response during viewing of spider pictures relative to the mushroom picture block, phobia group: $F(1,13) = 21.83$, $p < .001$, $\eta^2 = .63$; control group: $F(1,12) = 2.15$, $p > .15$. This effect was substantiated by a significant Category $\times$ Group interaction, $F(1,25) = 7.03$, $p < .05$, $\eta^2 = .22$. A between-group comparison also confirmed that blink magnitudes during viewing of spider pictures were significantly larger in the phobia relative to the control group, Group $F(1,25) = 13.3$, $p < .01$, $\eta^2 = .35$, whereas blink magnitudes during viewing of mushrooms pictures did not differ between phobics and controls, $F < 1$. Phobia-specific startle potentiation did not differ for the two probe times,

---

[5] One spider-phobic participant did not reappear to the second experimental session and the peripheral physiological data of a participant of the control group were lost due to technical problems. Three other participants (2 controls) were excluded from further analyses of the startle eyeblink because they did not meet the criteria of showing at least 40% valid responses. One phobic participant was excluded from analysis of the skin conductance level because of equipment failure and one other participant of the control group was excluded from data analyses of the heart rate date because of frequent extrasystoles.
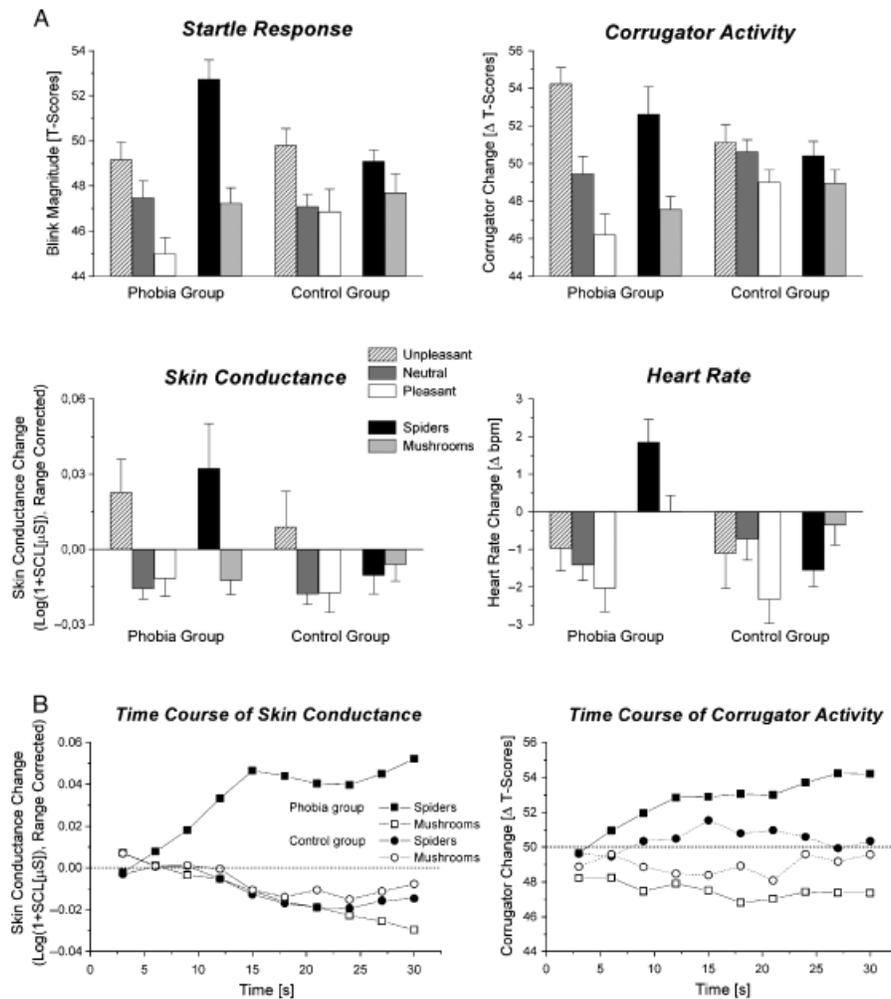
Figure 1. Response output patterns to phobia-relevant and standard affective pictures in spider-phobic participants and the control group. A: Mean startle blink magnitudes (upper left), mean corrugator activity change (upper right), mean skin conductance level change (lower left), and mean heart rate change (lower right) during viewing of phobia-relevant and standard affective pictures in spider-phobic participants and the control group. B: Change of skin conductance (left) and corrugator activity (right) in the time course of phobia-relevant picture presentation for spider-phobic participants and the control group.

$F(1,25) = 1.46$, $p > .2$) suggesting that the defensive response mobilization was maintained throughout the entire block of spider pictures. Mean startle response magnitudes to all stimulus categories are presented in the upper left panel of Figure 1A.

Corrugator activity. As expected, corrugator EMG also varied significantly for the different affective categories, $F(4,112) = 9.01$, $p < .001$, $\eta^2 = .24$. Although there was a clear increase in corrugator activity during viewing of unpleasant pictures relative to neutral contents, $F(1,29) = 6.05$, $p < .05$, $\eta^2 = .17$, corrugator activity significantly decreased during viewing of pleasant pictures, $F(1,29) = 9.81$, $p < .01$, $\eta^2 = .25$ (see upper right panel of Figure 1A). Only animal phobics showed increasing corrugator activity during viewing of spider pictures, and the differences between phobics and controls accumulated during the course of the block, Category × Time × Group $F(9,252) = 2.57$, $p < .01$ $\eta^2 = .08$ (see right panel of Figure 1B).

Skin conductance level. Skin conductance level also varied for the different affective categories, $F(4,108) = 5.23$, $p < .01$, $\eta^2 = .16$ (see lower left panel of Figure 1A). Although all par-

ticipants showed a significant increase in SCL during viewing of unpleasant relative to neutral pictures, $F(1,28) = 10.29$, $p < .01$, $\eta^2 = .24$, only animal phobics but not controls responded with increased SCL during viewing of spider pictures relative to the neutral mushrooms contents, Category × Group $F(1,27) = 5.19$, $p < .05$, $\eta^2 = .16$. Participants with specific phobia exhibited significantly stronger sympathetic activation during viewing of spider pictures than controls, Group $F(1,27) = 5.29$, $p < .05$, $\eta^2 = .16$. Again this increased sympathetic activation was maintained in the phobia group throughout the entire picture block, Category × Time × Group $F(9,243) = 5.01$, $p < .05$ $\eta^2 = .15$ (see left panel of Figure 1B). SCL changes during pleasant blocks did not differ from the neutral picture blocks, $F < 1$.

Heart rate. Whereas participants with specific phobia responded with a clear cardiac acceleration during viewing of spider pictures relative to mushroom pictures, $F(1,14) = 7.20$, $p < .05$, $\eta^2 = .34$, the control group showed increased orienting to the spider pictures as indexed by a marginally significant stronger heart deceleration relative to mushrooms, $F(1,13) = 3.38$, $p = .09$, $\eta^2 = .21$ (see lower right panel of Figure 1A). This pattern

Table 1. *Summary of Results for Contrasts of the Standard Affective Categories with Significant Activated Regions, MNI Coordinates, and Cluster Size (k_E)*

Region	Side	MNI coordinates x	y	z	k_E	t score	p_{FDR}
Overall							
Unpleasant > complex neutral							
Fusiform gyrus	L	−45	−51	−21	5878	6.01	.000
Inferior frontal gyrus	L	−57	30	15	3373	5.56	.000
Superior frontal gyrus		−3	66	24	1248	4.79	.001
Postcentral gyrus	R	48	−27	45	95	3.13	.014
Middle temporal gyrus	R	60	−6	−18	38	3.10	.015
Cerebellum	L	−21	−39	−48	10	2.96	.020
Gyrus rectus		0	39	−21	78	2.77	.028
Amygdala	R	21	0	−15		4.39	ROI .001
Amygdala	L	−21	0	−12		3.44	ROI .002
Insula	L	−27	12	−18		4.55	ROI .007
Insula	R	36	27	6		3.35	ROI .021
Complex neutral > unpleasant		No differential activation					
Pleasant > complex neutral							
Middle temporal gyrus	R	51	−66	12	882	6.28	.000
Middle temporal gyrus	L	−54	−66	15	1261	6.11	.000
Gyrus rectus		3	39	−18	925	4.59	.001
Amygdala	R	21	0	−18	145	4.40	.001
Postcentral gyrus	R	39	−33	51	217	4.29	.002
Insula	L	−27	12	−18	81	4.10	.003
Cerebellum		−3	−78	−36	20	3.68	.008
Postcentral gyrus	L	−33	−42	60	91	3.68	.008
Inferior frontal gyrus	L	−54	33	12	37	3.29	.019
Putamen	L	−24	3	−3	22	3.24	.021
Precentral gyrus	L	−57	0	39	53	3.20	.023
Middle temporal gyrus	R	60	−12	−21	12	3.14	.026
Paracentral lobule		−6	−33	66	10	2.96	.037
Amygdala	L	−27	0	−24		3.35	ROI .002
Complex neutral > pleasant		No differential activation					
Phobia group > control group							
Unpleasant		No differential activation					
Pleasant		No differential activation					
Complex neutral		No differential activation					
Control group > phobia group							
Unpleasant		No differential activation					
Pleasant		No differential activation					
Complex neutral		No differential activation					

of results was confirmed by a significant Category × Group interaction, $F(1,27) = 10.01$, $p < .005$, $\eta^2 = .27$.

Valence and arousal ratings. The valence and arousal ratings of unpleasant, neutral, and pleasant pictures corresponded to the normative ratings, which determined the preselection of the pictures, and the actual valence and arousal ratings of the standard affective categories did not differ between groups. In contrast, pictures of spiders were rated as more unpleasant and arousing in the phobia group than in the control group, $F(1,31) = 67.53$, $p < .001$, $\eta^2 = .69$; $F(1,31) = 47.52$, $p < .001$, $\eta^2 = .61$, for the valence and arousal ratings, respectively.

fMRI Activation Patterns

Whole brain analysis. As expected, affective stimuli evoked stronger responses than neutral stimuli in the amygdala, insula, and the inferior temporal visual stream supporting the ROI selection (see Table 1). Both groups did not differ in brain responses to standard affective materials (all between-group comparisons n.s.; see Table 1). In controls, however, viewing of

pleasant pictures compared to spider pictures evoked stronger bilateral activations of the middle temporal gyri (left: −57 −69 15; $t = 5.34$, $p_{FDR} < .01$; right: 51 −72 15; $t = 4.02$, $p_{FDR} < .05$) and of the left fusiform gyrus (−42 −51 −24; $t = 3.91$, $p_{FDR} < .05$; Table 2).

Amygdala activation. ROI analysis revealed that participants with animal phobia showed significantly stronger activation of both amygdalae when exposed to pictures of spiders relative to mushroom pictures (right: 30 0 −15; $t = 3.54$, $p_{FDR} < .05$; left: −30 0 −18, $t = 2.76$, $p_{FDR} < .05$; see Figures 2A,3A; Table 2). As expected, this contrast was not significant in the control group (see Figure 2A; Table 2). The between-group comparison of the amygdala activation during viewing of spider pictures, however, was not significant. By contrasting fMRI maps evoked by pleasant pictures with those evoked by neutral ones, bilateral amygdala activations (right: 21 0 −18; $t = 4.40$, $p_{FDR} < .01$; left: −27 0 −24; $t = 3.35$, $p_{FDR} < .05$) were observed in all participants with no differences between groups (see Table 1). As expected, viewing unpleasant compared to neutral materials

Table 2. *Summary of Results for Phobia-Relevant Contrasts with Significant Activated Regions, MNI Coordinates, and Cluster Size (k_E)*

| Region	Side	MNI coordinates			k_E	t score	p_{FDR}
		x	y	z			
Phobia group							
Spiders > mushrooms							
Insula	L	−45	9	−9	19	4.62	.048
Supramarginal gyrus	R	63	−30	33	19	4.25	.048
Rolandic operculum	R	63	9	3	12	4.37	.048
Insula	R	39	−3	−12		3.27	ROI .014
Amygdala	R	30	0	−15		3.54	ROI .025
Amygdala	L	−30	0	−18		2.76	ROI .027
Mushrooms > spiders		No differential activation					
Spiders > pleasant		No differential activation					
Pleasant > spiders		No differential activation					
Control group							
Spiders > mushrooms		No differential activation					
Mushrooms > spiders		No differential activation					
Spiders > pleasant		No differential activation					
Pleasant > spiders							
Middle temporal gyrus	L	−57	−69	15	202	5.34	.008
Middle temporal gyrus	R	51	−72	15	31	4.02	.028
Fusiform gyrus	L	−42	−51	−24	12	3.91	.035
Phobia group > control group							
Spiders							
Insula	R	39	6	6		3.39	ROI .032
Insula	L	−42	9	−9		3.24	ROI .032
Mushrooms		No differential activation					
Control group > phobia group							
Spiders		No differential activation					
Mushrooms		No differential activation					
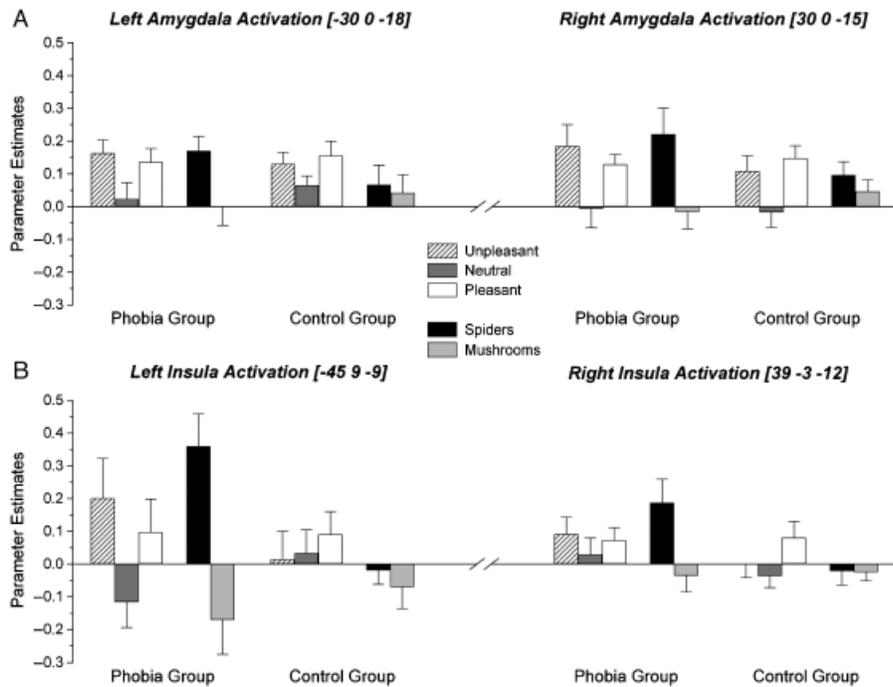
Figure 2. A: Hemodynamic responses to different picture categories for left and right amygdala in spider-phobic participants and the control group. Mean parameter estimates for the phobia and control group were derived from the clusters (6 mm sphere, 33 voxels) of maximum activation within the amygdalae for the contrast spiders > mushrooms in spider-phobic participants. B: Hemodynamic responses to different picture categories for left and right insula in spider-phobic participants and the control group. Mean parameter estimates for the phobia and control group were derived from the clusters (6 mm sphere, 33 voxels) of maximum activation within the insulae for the contrast spiders > mushrooms in spider phobic participants.
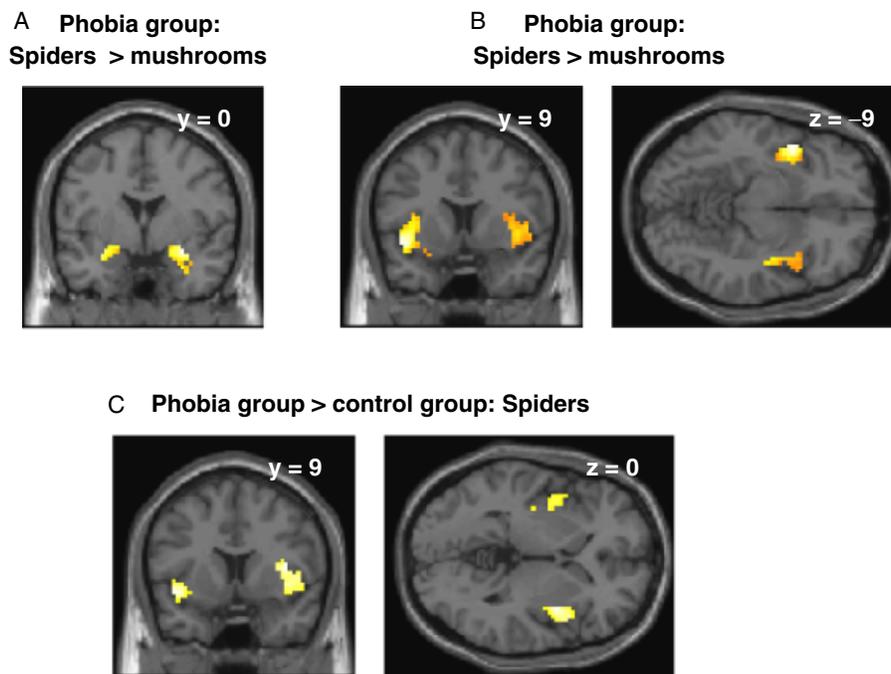
A   **Phobia group:**
     Spiders > mushrooms

B   **Phobia group:**
     Spiders > mushrooms

C   **Phobia group > control group: Spiders**

Figure 3. Region of interest (ROI) activation. A: Amygdala activation for the contrast spiders > mushrooms in spider phobics. B: Insula activation for the contrast spider > mushrooms in spider phobics. C: Insula activation for the contrast phobia group > control group during viewing of spider pictures. Activation foci are overlaid on a T1 scan.

also significantly increased bilateral amygdala activations (right: 21 0 − 15; $t = 4.39$, $p_{FDR} < .01$; left: − 21 0 − 12; $t = 3.44$; $p_{FDR} < .01$) in all participants (see Table 1). Importantly, amygdala activation during processing of pleasant pictures did not differ from amygdala activation prompted during viewing of phobia-relevant material in participants with specific phobia, suggesting that increased amygdala activation was not fear specific (phobia group: spiders > pleasant; right: $t = 1.62$, n.s.; left: $t = 1.42$, n.s.).

Insula activation. During viewing of spider pictures, phobic participants also showed increased bilateral insula activation as compared to when viewing mushroom pictures (left: − 45 9 − 9; $t = 4.62$, $p_{FDR} < .05$; right: 39 − 3 − 12; $t = 3.27$, $p_{FDR} < .05$; Table 2; see Figures 2B, 3B), whereas this contrast was not significant in the control group (see Figure 2B; Table 2). Moreover, ROI analysis revealed that phobics exhibited a significantly stronger insula activation during viewing of spider pictures than controls in the between-group comparison (right: 39 6 6; $t = 3.39$, $p_{FDR} < .05$; left: − 42 9 − 9; $t = 3.24$, $p_{FDR} < .05$; Table 2; Figure 3C). Contrasting pleasant with neutral picture conditions revealed increased left insula activation (− 27 12 − 18; $t = 4.10$, $p_{FDR} < .01$) during viewing of pleasant pictures in all participants with no differences between groups (see Table 1). Moreover, viewing unpleasant compared to neutral materials also resulted in increased bilateral insular activations (left: − 27 12 − 18; $t = 4.55$, $p_{FDR} < .01$; right: 36 27 6; $t = 3.44$, $p_{FDR} < .05$) in all participants (see Table 1).

Time course analysis. Exploratory analyses of the time course of the hemodynamic brain response during the spider picture block used parameter estimates that were extracted using a finite impulse response function (time bin = 3 s, time window = 30 s) for spheres of 6 mm centered around voxels of maximum activation within amygdala and insula for the contrast spi-

ders > mushrooms in spider-phobic participants. The extracted values were averaged for the first and the second halves of the picture block. Analyses revealed that during viewing of spider pictures phobic participants showed elevated activation in both insulae compared to controls during the first and the second halves of the block (early: left, $t = 3.61$, $p < .01$, right, $t = 3.66$, $p < .01$; late: left, $t = 2.27$, $p < .05$, right, $t = 2.80$, $p < .05$). In contrast, activation of the right amygdala during viewing of spider pictures only differed significantly between phobics and controls during the first half of the block ($t = 2.64$, $p < .05$). During the second half, these group differences were reduced and were only marginally significant ($t = 1.76$, $p = .09$). Activation of the left amygdala did not differ significantly between both groups during either part of the spider block presentation.

Discussion

Sustained Phobic Stimulation and Defensive Response Mobilization

As predicted, participants with small animal phobia showed increased defensive response mobilization during sustained exposure to phobia-relevant stimuli as indexed by phobia-specific potentiation of the startle reflex, increased corrugator activity, and autonomic arousal. Phobia specific potentiation of the startle reflex was maintained throughout the entire block of pictures, suggesting that the defensive response mobilization did not habituate throughout the 30-s block of repeated picture presentation. The autonomic measures support this interpretation because skin conductance levels even further increased during the sustained phobic stimulation. Moreover, participants with specific phobia responded with a cardiac acceleration to their feared objects whereas all other affective materials prompted a heart rate deceleration in this group. In the control group there was a general heart deceleration to all affective contents.

These findings confirm and extend previous data by Bradley et al. (1996) as well as Sutton et al. (1997), showing that extended processing of a series of pictures of the same affective valence led to either increases or maintenance of affective responding across trials in a block. The current results demonstrate that phobia-specific defensive mobilization extends throughout repeated presentation of phobia-relevant pictures in a block of 30 s. Again, replicating previous findings, potentiation of the startle reflex during processing of phobia-relevant stimuli was specific for the fear-relevant content, whereas there were no group differences for the other affective categories.

Sustained Phobic Stimulation and Brain Activation

Participants with animal phobia showed increased activation of the *amygdala* during exposure to phobia-related picture materials relative to neutral stimuli—a result that is in line with previous findings (Carlsson et al., 2004; Dilger et al., 2003; Schienle, Schäfer, Walter, et al., 2005; Straube, Mentzel, et al., 2006). However, such increased amygdala activation does not seem to be specific for sustained phobic stimulation. Increased activation of the amygdala was also found during viewing of other unpleasant pictures and also during viewing of pleasant ones. These results confirm previous reports of increased amygdala activation to happy faces (Yang et al., 2002), erotic scenes (Stark et al., 2005), and other pleasant pictures (Hamann, Ely, Grafton, & Kilts, 1999; Liberzon, Phan, Decker, & Taylor, 2003). These data are also in line with recent evidence from a study of Sabatinelli and coworkers (2005), who found increased amygdala activation in snake-phobic participants when they were exposed to pictures of snakes using an event-related design. As in the current study, however, the amygdala activation during phobic stimulation did not differ from that evoked during viewing of erotica and mutilation pictures in this group of participants. Time course analyses of the current data revealed that participants with specific phobia showed stronger activation of the right amygdala during viewing of spider pictures relative to controls, but only at the beginning of the sustained phobic stimulation. During the second half of the block no significant group differences emerged. These data are in line with previous findings showing that amygdala activation attenuated during later trials of cued shock anticipation (Phelps et al., 2001) and support the hypothesis that the amygdala BOLD signal primarily indexes the detection of motivationally significant stimuli in the environment.

In contrast, phobics exhibited significantly increased activations of the *insula* during viewing of spider pictures both relative to neutral pictures and compared to controls, replicating previous findings (Carlsson et al., 2004; Dilger et al., 2003; Straube, Glauer, et al., 2006). Importantly, additional analyses revealed that increased activation of the left mid-insular cortex seemed to be specific for the processing of fear-related stimuli, because increased activity of this part of the insula during sustained exposure to phobia-relevant stimuli was stronger than the activity evoked by pleasant pictures.[6] Increased activation of the insular cortex during sustained exposure to feared objects

(e.g., touching a glass container housing the feared animal or touching a "contaminated" object or listening to traumatic scenes) has been demonstrated for a variety of anxiety disorders, including patients with specific phobia, obsessive–compulsive disorder, or posttraumatic stress disorder (see Rauch, Savage, Alpert, Fischman, & Jenike, 1997). Interestingly, this increased activation of the insular cortex in participants with anxiety disorders during sustained exposure of their feared objects clearly contrasts the brain activation during processing of emotional pictures in participants with psychopathic behavior. These individuals with psychopathic behavior display a brief activation of the amygdala but no further activation of the insula during processing of unpleasant pictures (Birbaumer et al. 2005). These data support the view that the insular cortex is critically involved in integrating threatening information from the environment with the interoception of the defensive mobilization of the body leading to an increased experience or awareness of the fear state. Accordingly, insula hyperactivity seems to be a common feature in persons with elevated trait anxiety (Simmons et al., 2006; Stein, Simmons, Feinstein, & Paulus, 2007). In the current study sustained exposure to phobia-relevant objects prompted phobia-specific startle potentiation, cardiac acceleration, and increase in skin conductance accompanied by reports of intense unpleasant feeling states in participants with specific phobia. These participants also showed phobia-specific activation of the insula. Although the relatively long temporal gap between the assessment of the physiological responses and the brain imaging session did not allow for meaningful correlational analyses between these variables, the different measures were obtained within the same group of participants. Assuming that the exposure to the feared objects prompted a comparable fear state in both sessions, these data suggest that the phobia-specific activation of the insular cortex goes in parallel with the strong defensive response mobilization and an increase in autonomic arousal integrated to an intense emotional experience.

Additionally, spider-phobic participants showed activations of the gyrus supramarginalis and the rolandic operculum during viewing of spider pictures compared to mushroom pictures. Increased activation of the right supramarginal gyrus has been previously described in a study presenting fear-relevant materials to patients with obsessive–compulsive disorder (Schienle, Schäfer, Stark, Walter, & Vaitl, 2005). This response pattern might be related to the activation of a motor program involved in the action tendency to escape. The pronounced increase in heart rate in phobics during their viewing of spider pictures is in line with such an interpretation. The bilateral posterior rolandic operculum has been described recently as being involved in the processing of emotional music (Koelsch, Fritz, von Cramon, Müller, & Friederici, 2006). This area, anatomically adjacent to the insula, might therefore contribute to the emotional evaluation of auditory but also visual material.

Conclusions

In summary, the present findings provide further evidence that sustained exposure to symbolic representations of phobia-relevant objects evoke robust defensive response mobilization in participants with specific phobia accompanied by a strong sympathetic activation, that is, an increase in sweat gland activity and a marked cardiac acceleration. Moreover, this phobia-specific defensive response mobilization was associated with enhanced activation of the left mid-insula in individuals with animal phobia, supporting the view that this brain region might serve an

---

[6]To test the specificity of insula activations evoked by phobia-related stimuli, the region of interest was reduced to a sphere of 5 mm (19 voxels) centered around the voxel of highest activation in the left insula ($-45\ 9\ -9$). Analyses revealed a significantly stronger activation during viewing of spider pictures than during pleasant pictures in spider-phobic participants ($t = 2.23$, $p_{FDR} = .046$), but not in controls (see Figure 2B), thus showing phobia-specific activation of this region.

important role in integrating the information of aversive stimuli in the environment with the bodily symptoms evoked by the feared objects. Accordingly, patients with various anxiety disorders exhibit increased activation of the insular cortex during exposure to their feared objects (Rauch et al., 1997). Interestingly, successful behavior therapy reduces insular activation during exposure to the feared objects in individuals with animal phobia (Straube, Glauer, et al., 2006). In contrast, amygdala activation seems to be less phobia specific during sustained stimulation but rather serves as a first stage discriminator for motivationally relevant stimuli. Enhanced amygdala activation was observed during viewing of generally unpleasant and pleasant stimuli in all participants and in the phobia group during phobia-related stimulation but with no significant differences between the emotionally relevant categories. Compared to controls, phobic individuals showed enhanced activation of the right amygdala only during the first half of sustained presentation of spider pictures. In a similar vein, Larson et al. (2006) found that animal phobics respond with a strong but brief

amygdala activation to phobic stimulation, whereas responses of nonphobic controls were weaker but more sustained. Therefore, amygdala BOLD responses seem to reflect the detection of significant stimuli. Thus amygdala activation is specifically increased when phobia-relevant stimuli are difficult to detect, that is, if phobic stimuli are masked (Carlsson et al., 2004), presented very briefly (Schienle, Schäfer, Walter, et al., 2005), or are presented in an event-related procedure (e.g., Dilger et al., 2003), but less pronounced during sustained exposure to phobia-relevant pictures. In contrast, activation of the insula is more pronounced during unmasked relative to masked presentation of phobia-related pictures in spider phobic individuals (Carlsson et al., 2004) or when participants with specific phobia are concentrated on the identification of phobic stimuli but not during a distraction task (Straube, Mentzel, et al., 2006). Thus, activation of the insular cortex seems to be associated with an actual or anticipated (Phelps et al., 2001) defensive body state that might trigger an increase in anxious affect that is typical for various patients with anxiety disorders.

REFERENCES

Adolphs, R., & Spezio, M. (2006). Role of the amygdala in processing visual social stimuli. *Progress in Brain Research*, *156*, 363–378.

Anders, S., Lotze, M., Erb, M., Grodd, W., & Birbaumer, N. (2004). Brain activity underlying emotional valence and arousal: A response-related fMRI study. *Human Brain Mapping*, *23*, 200–209.

Birbaumer, N., Veit, R., Lotze, M., Erb, M., Hermann, C., Grodd, W., et al. (2005). Deficient fear conditioning in psychopathy: A functional magnetic resonance imaging study. *Archives of General Psychiatry*, *62*, 799–805.

Bradley, M. M., Cuthbert, B. N., & Lang, P. J. (1996). Picture media and emotion: Effects of a sustained affective context. *Psychophysiology*, *33*, 662–670.

Bradley, M. M., & Lang, P. J. (1994). Measuring emotion: The self-assessment manikin and the semantic differential. *Journal of Behavior Therapy and Experimental Psychiatry*, *25*, 49–59.

Buchanan, T. W., Tranel, D., & Adolphs, R. (2004). Anteromedial temporal lobe damage blocks startle modulation by fear and disgust. *Behavioral Neuroscience*, *118*, 429–437.

Büchel, C., Morris, J., Dolan, R. J., & Friston, K. J. (1998). Brain systems mediating aversive conditioning: An event-related fMRI study. *Neuron*, *20*, 947–957.

Carlsson, K., Petersson, K. M., Lundqvist, D., Karlsson, A., Ingvar, M., & Öhman, A. (2004). Fear and the amygdala: Manipulation of awareness generates differential cerebral responses to phobic and fear-relevant (but nonfeared) stimuli. *Emotion*, *4*, 340–353.

Craig, A. D. (2002). How do you feel? Interoception: The sense of the physiological condition of the body. *Nature Reviews. Neuroscience*, *3*, 655–666.

Critchley, H. D. (2005). Neural mechanisms of autonomic, affective, and cognitive integration. *Journal of Comparative Neurology*, *493*, 154–166.

Critchley, H. D., Wiens, S., Rotshtein, P., Öhman, A., & Dolan, R. J. (2004). Neural systems supporting interoceptive awareness. *Nature Neuroscience*, *7*, 189–195.

Davis, K. D., Kwan, C. L., Crawley, A. P., & Mikulis, D. J. (1998). Event-related fMRI of pain: Entering a new era in imaging pain. *NeuroReport*, *9*, 3019–3023.

Davis, M. (2000). The role of the amygdala in conditioned and unconditioned fear and anxiety. In J. P. Aggleton (Ed.), *The amygdala: A functional analysis* (2nd ed, pp. 213–287). Oxford: Oxford University Press.

Davis, M., & Whalen, P. J. (2001). The amygdala: Vigilance and emotion. *Molecular Psychiatry*, *6*, 13–34.

Dilger, S., Straube, T., Mentzel, H.-J., Fitzek, C., Reichenbach, J. R., Hecht, H., et al. (2003). Brain activation to phobia-related pictures in spider phobic humans: An event-related functional magnetic resonance imaging study. *Neuroscience Letters*, *348*, 29–32.

Duncan, S. L., & Barrett, L. F. (2007). The amygdala in visual awareness. *Trends in Cognitive Sciences*, *11*, 190–192.

Fredrikson, M. (1981). Orienting and defense reactions to phobic and conditioned fear stimuli in phobics and normals. *Psychophysiology*, *18*, 456–465.

Fredrikson, M., & Furmark, T. (2003). Amygdaloid regional cerebral blood flow and subjective fear during symptom provocation in anxiety disorders. *Annals of the New York Academy of Science*, *985*, 341–347.

Fridlund, A. J., & Cacioppo, J. T. (1986). Guidelines for human electromyographic research. *Psychophysiology*, *23*, 567–589.

Funayama, E. S., Grillon, C., Davis, M., & Phelps, E. A. (2001). A double dissociation in the affective modulation of startle in humans: Effects of unilateral temporal lobectomy. *Journal of Cognitive Neuroscience*, *13*, 721–729.

Genovese, C. R., Lazar, N. A., & Nichols, T. (2002). Thresholding of statistical maps in functional neuroimaging using the false discovery rate. *NeuroImage*, *15*, 870–878.

Globisch, J., Hamm, A. O., Esteves, F., & Öhman, A. (1999). Fear appears fast: Temporal course of startle reflex potentiation in animal fearful subjects. *Psychophysiology*, *36*, 66–75.

Globisch, J., Hamm, A. O., Schneider, R., & Vaitl, D. (1993). A computer program for scoring reflex eyeblink and electrodermal responses written in PASCAL [Abstract]. *Psychophysiology*, *30*(Suppl.), 13.

Hamann, S. B., Ely, T. D., Grafton, S. T., & Kilts, C. D. (1999). Amygdala activity to enhanced memory for pleasant and aversive stimuli. *Nature Neuroscience*, *2*, 289–293.

Hamm, A. O. (2006). *Spezifische Phobien* [Specific Phobias]. Göttingen: Hogrefe Verlag.

Hamm, A. O., Cuthbert, B. N., Globisch, J., & Vaitl, D. (1997). Fear and the startle reflex: Blink modulation and autonomic response patterns in animal and mutilation fearful subjects. *Psychophysiology*, *34*, 97–107.

Koelsch, S., Fritz, T., von Cramon, D. Y., Müller, K., & Friederici, A. D. (2006). Investigating emotion with music: An fMRI study. *Human Brain Mapping*, *27*, 239–250.

Lang, P. J., Bradley, M. M., & Cuthbert, B. N. (2005). *International affective picture system (IAPS): Digitized photographs, instruction manual and affective ratings*. Technical Report A-6. Gainesville, FL: University of Florida.

Larson, C. L., Schaefer, H. S., Siegle, G. J., Jackson, C. A., Anderle, M. J., & Davidson, R. J. (2006). Fear is fast in phobic individuals: Amygdala activation in response to fear-relevant stimuli. *Biological Psychiatry*, *60*, 410–417.

LeDoux, J. E. (1996). *The emotional brain*. New York: Simon & Schuster.

Liberzon, I., Phan, K. L., Decker, L. R., & Taylor, S. F. (2003). Extended amygdala and emotional salience: A PET investigation of positive and negative affect. *Neuropsychopharmacology*, *28*, 726–733.

Lykken, D. T., Rose, R. J., Luther, B., & Maley, M. (1966). Correcting psychophysiological measures for individual differences in range. *Psychological Bulletin*, *66*, 481–484.

Maren, S., & Quirk, G. J. (2004). Neuronal signalling of fear memory. *Nature Reviews. Neuroscience*, *5*, 844–852.

Morris, J. S., Öhman, A., & Dolan, R. J. (1998). Conscious and unconscious emotional learning in the human amygdala. *Nature*, *393*, 467–470.

Nitschke, J. B., Sarinopoulos, I., Mackiewicz, K. L., Schaefer, H. S., & Davidson, R. J. (2006). Functional neuroanatomy of aversion and its anticipation. *NeuroImage*, *29*, 106–116.

Öhman, A., & Mineka, S. (2001). Fears, phobias, and preparedness: Toward an evolved module of fear and fear learning. *Psychological Review*, *108*, 483–522.

Paulus, M. P., & Stein, M. B. (2006). An insular view of anxiety. *Biological Psychiatry*, *60*, 383–387.

Pessoa, L., Japee, S., Sturman, D., & Ungerleider, L. G. (2006). Target visibility and visual awareness modulate amygdala responses to fearful faces. *Cerebral Cortex*, *16*, 366–375.

Peyron, R., Laurent, B., & Garcia-Larrea, L. (2000). Functional imaging of brain responses to pain. A review and meta-analysis. *Neurophysiologie Clinique*, *30*, 263–288.

Phan, K. L., Wager, T., Taylor, S. F., & Liberzon, I. (2002). Functional neuroanatomy of emotion: A meta-analysis of emotion activation studies in PET and fMRI. *NeuroImage*, *16*, 331–348.

Phelps, E. A., O'Connor, K. J., Gatenby, J. C., Gore, J. C., Grillon, C., & Davis, M. (2001). Activation of the left amygdala to a cognitive representation of fear. *Nature Neuroscience*, *4*, 437–441.

Rauch, S. L., Savage, C. R., Alpert, N. M., Fischman, A. J., & Jenike, M. A. (1997). The functional neuroanatomy of anxiety: A study of three disorders using positron emission tomography and symptom provocation. *Biological Psychiatry*, *42*, 446–452.

Rauch, S. L., Savage, C. R., Alpert, N. M., Miguel, E. C., Baer, L., Breiter, H. C., et al. (1995). A positron emission tomographic study of simple phobic symptom provocation. *Archives of General Psychiatry*, *52*, 20–28.

Reynolds, S. M., & Zahm, D. S. (2005). Specificity in the projections of prefrontal and insular cortex to ventral striatopallidum and the extended amygdala. *Journal of Neuroscience*, *25*, 11757–11767.

Rosen, J. B., & Schulkin, J. (1998). From normal fear to pathological anxiety. *Psychological Review*, *105*, 325–350.

Sabatinelli, D., Bradley, M. M., Fitzsimmons, J. R., & Lang, P. J. (2005). Parallel amygdala and inferotemporal activation reflect emotional intensity and fear relevance. *NeuroImage*, *24*, 1265–1270.

Sabatinelli, D., Bradley, M. M., & Lang, P. J. (2001). Affective startle modulation in anticipation and perception. *Psychophysiology*, *38*, 719–722.

Schienle, A., Schäfer, A., Stark, R., Walter, B., & Vaitl, D. (2005). Neural responses of OCD patients towards disorder-relevant, generally disgust-inducing and fear-inducing pictures. *International Journal of Psychophysiology*, *57*, 69–77.

Schienle, A., Schäfer, A., Walter, B., Stark, R., & Vaitl, D. (2005). Brain activation of spider phobics towards disorder-relevant, generally disgust- and fear-inducing pictures. *Neuroscience Letters*, *388*, 1–6.

Schienle, A., Stark, R., Walter, B., Blecker, C., Ott, U., Kirsch, P., et al. (2002). The insula is not specifically involved in disgust processing: An fMRI study. *NeuroReport*, *13*, 2023–2026.

Shi, C. J., & Cassell, M. D. (1998). Cortical, thalamic, and amygdaloid connections of the anterior and posterior insular cortices. *Journal of Comparative Neurology*, *399*, 440–468.

Shimizu, H. (1978). Reliable and precise identification of R-waves in the EKG with a simple peak detector. *Psychophysiology*, *15*, 499–501.

Simmons, A., Strigo, I., Matthews, S. C., Paulus, M. P., & Stein, M. B. (2006). Anticipation of aversive visual stimuli is associated with increased insula activation in anxiety-prone subjects. *Biological Psychiatry*, *60*, 402–409.

Smith, J. C., Bradley, M. M., & Lang, P. J. (2005). State anxiety and affective physiology: Effects of sustained exposure to affective pictures. *Biological Psychiatry*, *69*, 247–260.

Stark, R., Schienle, A., Girod, C., Walter, B., Kirsch, P., Blecker, C., et al. (2005). Erotic and disgust-inducing pictures—Differences in the hemodynamic responses of the brain. *Biological Psychology*, *70*, 19–29.

Stein, M. B., Simmons, A. N., Feinstein, J. S., & Paulus, M. P. (2007). Increased amygdala and insula activation during emotion processing in anxiety-prone subjects. *American Journal of Psychiatry*, *164*, 318–327.

Straube, T., Glauer, M., Dilger, S., Mentzel, H.-J., & Miltner, W. H. R. (2006). Effects of cognitive-behavioral therapy on brain activation in specific phobia. *NeuroImage*, *29*, 125–135.

Straube, T., Mentzel, H.-J., & Miltner, W. H. R. (2006). Neural mechanisms of automatic and direct processing of phobogenic stimuli in specific phobia. *Biological Psychiatry*, *59*, 162–170.

Sutton, S. K., Davidson, R. J., Donzella, B., Irwin, W., & Dottle, D. A. (1997). Manipulating affective state using extended picture presentation. *Psychophysiology*, *34*, 217–226.

Tzourio-Mazoyer, N., Landeau, B., Papathanassiou, D., Crivello, F., Etard, O., Delcroix, N., et al. (2002). Automated anatomical labeling of activations in SPM using a macroscopic anatomical parcellation of the MNI MRI single-subject brain. *NeuroImage*, *15*, 273–289.

Venables, P. H., & Christie, M. J. (1980). Electrodermal activity. In I. Martin & P. H. Venables (Eds.), *Techniques in psychophysiology* (pp. 4–67). New York: Wiley.

Weike, A. I., Hamm, A. O., Schupp, H. T., Runge, U., Schroeder, H. W. S., & Kessler, C. (2005). Fear conditioning following unilateral temporal lobectomy: Dissociation of conditioned startle potentiation and autonomic learning. *Journal of Neuroscience*, *25*, 11117–11124.

Whalen, P. J., Rauch, S. L., Etcoff, N. L., McInerney, S. C., Lee, M., & Jenike, M. A. (1998). Masked presentations of emotional facial expressions modulate amygdala activity without explicit knowledge. *Journal of Neuroscience*, *18*, 411–418.

Yang, T. T., Menon, V., Eliez, S., Blasey, C., White, C. D., Reid, A. J., et al. (2002). Amygdalar activation associated with positive and negative facial expressions. *NeuroReport*, *13*, 1737–1741.