Selective effects of social anxiety, anxiety sensitivity, and negative affectivity on the neural bases of emotional face processing

Tali Manber Ball a,b,* , Sarah Sullivan a , Taru Flagana , Carla A. Hitchcock a,c , Alan Simmonsa,d , Martin P. Paulusa,d , Murray B. Stein a,d,e

a Department of Psychiatry, University of California San Diego, San Diego, CA, USA
b San Diego State University/University of California San Diego Joint Doctoral Program in Clinical Psychology, San Diego, CA, USA
c Alliant International University San Diego, San Diego, CA, USA
d Psychiatry Service, Veterans Affairs, San Diego Healthcare System, San Diego, CA, USA
e Department of Family and Preventive Medicine, University of California San Diego, San Diego, CA, USA

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ABSTRACT

Individuals with high anxiety show heightened neural activation in affective processing regions, including the amygdala and insula. Activations have been shown to be correlated with anxiety severity, but although anxiety is a heterogeneous state, prior studies have not systematically disentangled whether neural activity in affective processing circuitry is uniquely related to specific domains of anxiety. Forty-five young adults were tested on an emotional face processing task during functional magnetic resonance imaging. Participants completed the Social Interpersonal Anxiety Scale, Anxiety Sensitivity Index, and Spielberger Trait Anxiety Inventory. Using a robust multiple regression approach, we examined the effects of social anxiety, anxiety sensitivity, and trait anxiety (which overlapped with depressive symptoms, and can therefore be considered a measure of negative affectivity) on activation in insula, amygdala, and medial prefrontal cortex in response to emotional faces. Adjusting for negative affectivity and anxiety sensitivity, social anxiety was associated with activity in left amygdala, right insula, and subgenual anterior cingulate across all emotional faces. When comparing negative and positive faces directly, greater negative affectivity was uniquely associated with less activity to positive faces in left amygdala, left anterior insula, and dorsal anterior cingulate. The current findings support the hypothesis that hyperactivity in brain areas during general emotional face processing is predominantly a function of social anxiety. In comparison, hypoactivity to positively valenced faces was predominantly associated with negative affectivity. Implications for the understanding of emotion processing in anxiety are discussed.

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Introduction

Anxiety disorders are highly prevalent (12 month prevalence of 18% Kessler et al., 2005), and cause significant distress, reduced quality of life (Barrera and Norton, 2009; Lochner et al., 2003; Mendlewicz and Stein, 2000), and loss of productivity (DuPont et al., 1996). Functional brain imaging of individuals that either have, or are at high risk to develop, anxiety disorders allows for the study of which neural substrates and cognitive or affective processes contribute to disordered anxiety. Such investigations have consistently found that anxiety-prone individuals show hyperactivity in affective circuitry during emotional processing tasks (Etkin and Wager, 2007). However, such studies have not systematically disentangled how different domains of anxiety are uniquely related to activity in affective processing circuitry.

Neural correlates of emotional face processing

One key approach to examine neural substrates of anxiety-related processes is to present individuals with emotional faces. Emotional faces are salient social cues that convey important information about a potential threat or reward in the environment (Haxby et al., 2000). Facial expressions reliably engage the visual system, particularly fusiform gyrus (Haxby et al., 2002; Kanwisher et al., 1997; Vuilleumier and Pourtois, 2007). Importantly, emotional faces also engage affective circuitry, particularly the amygdala and insula, as well as the medial prefrontal cortex (PFC) (Adolphs, 2002; Anderson et al., 2003; Haxby et al., 2002; Heberlein et al., 2008; Vuilleumier and Pourtois, 2007).

Neural basis of emotional face processing in anxiety

Increased activation in regions engaged by emotional faces, namely the amygdala, insula and medial PFC, has been seen across a range of anxiety diagnoses (for a review see Britton and Rauch, 2008). Viewing harsh or angry faces led to greater activity in amygdala,
insula, and dorsal anterior cingulate cortex (ACC) in individuals with social anxiety disorder (Etkin and Wager, 2007; Evans et al., 2008; Phan et al., 2006; Straube et al., 2004). In generalized anxiety disorder, fearful and angry faces have yielded increased activity in amygdala, ACC, and ventromedial PFC (McClure et al., 2007; Monk et al., 2008). Studies of posttraumatic stress disorder (PTSD) have also found hyperactivity in amygdala and insula to negatively valenced faces (Fonzo et al., 2010; Rauch et al., 2000), with hypoactivity in medial PFC in response to fearful facial expressions (Shin et al., 2006). Similar results have extended to non-clinical, high trait anxious populations, with greater activation in amygdala and insula during conscious (Stein et al., 2007) and non-conscious (Etkin et al., 2004) emotional face processing. Changes in rostral and dorsal ACC activity were also observed in non-clinically anxious individuals to ambiguous facial stimuli (Simmons et al., 2008). Emotional face processing tasks are therefore a commonly used and well-understood probe for examining neural dysfunction in affective processing circuitry in anxious individuals.

Components of anxiety

Although these findings have applied broadly to individuals who experience high anxiety, anxiety is not one homogeneous construct (Clark and Watson, 1991). Factor analytic studies have conceptualized proneness to experience negative affect (i.e., neuroticism) as an overarching factor, with specific sub-factors differentiating depression (i.e., lack of positive affect) and anxiety (i.e., anxious arousal) (Clark and Watson, 1991). Anxiety can be broken down further into more specific constructs (e.g., anxiety sensitivity), that sometimes, though not always, are associated with increased propensity to develop anxiety disorders (e.g., panic disorder). These constructs can be present in an individual either alone or in combination, with each at sub-clinical or clinical levels. The goal of this investigation was to examine whether brain activation underlying emotional face processing can be mapped parametrically onto different specific anxiety-related constructs.

In the present study, we focus on three important components of anxiety that have been used to describe highly anxious individuals. Trait anxiety refers to one's general proneness to experience anxiety. Within Clark's three-factor model, trait anxiety has been associated with both the general negative affectivity factor as well as the anxiety factor (Bieling et al., 1998). Second, social anxiety refers to concern about negative evaluation from others or other negative social consequences, and has been associated with social anxiety disorder (Stein and Stein, 2008). Third, anxiety sensitivity refers to catastrophic interpretations and potential negative consequences of physical symptoms of anxiety, and has been associated with panic disorder (Reiss et al., 1986). These anxiety domains are inter-correlated—if one is high there is an increased chance that others will be high as well—but they are still separable.

The present study

Although activations, particularly in amygdala and insula, have been shown to be generally correlated with anxiety severity (Phan et al., 2006; Rauch et al., 2000), prior studies have not systematically disentangled how separate domains of anxiety, such as trait anxiety, social anxiety, and anxiety sensitivity, are uniquely related to hyperactivity in affective circuitry. In a previous study from our group (Stein et al., 2007), we demonstrated that individuals high in trait anxiety had greater activity in amygdala and insula when processing emotional faces. Furthermore, activity in response to faces in both amygdala and insula was associated with trait anxiety, whereas only insula activity was associated with anxiety sensitivity. However, social anxiety was not examined. Social anxiety may be especially related to aberrant responses to emotional faces due to the inherently social nature of faces as stimuli.

The present study aimed to expand on these previous findings—in a new sample of subjects—by examining the independent effects of trait anxiety, anxiety sensitivity, and social anxiety on neural responses to emotional faces. We included all three of these domains of anxiety simultaneously in a multiple regression to predict neural activity, focusing specifically on important affective processing regions (i.e., amygdala, insula, and medial PFC), with fusiform gyrus as a visual processing control region. Using a multiple regression allowed us to examine the effects of each anxiety domain independently, while statistically controlling for the other two domains. We were also interested in whether these effects are present when processing emotional facial expressions broadly, or whether there are differential effects to specific types of faces (i.e., negatively or positively valenced faces).

Hypotheses

We hypothesized that social anxiety would specifically relate to amygdala activation to emotional faces. Emotional facial expressions are important social cues that can signal threat, especially to people who are particularly sensitive to the approval of others. The amygdala has been implicated in social information and threat processing (Haxby et al., 2002), which are aberrant in individuals with social anxiety (Clark and McManus, 2002). We also hypothesized that anxiety sensitivity would specifically relate to insula activation to emotional faces. The insula has been implicated in interoception, the perception of internal bodily sensations (Craig, 2009; Paulus and Stein, 2006). The insula’s hyperactivity in response to emotional faces in individuals with high anxiety may thus be specifically due to their sensitivity to the physical symptoms of anxiety (Paulus and Stein, 2006; Rosso et al., 2010; Stein et al., 2007).

Methods

Participants

The study was approved by the University of California, San Diego and San Diego State University Institutional Review Boards. Approximately 1000 undergraduate San Diego State University (SDSU) students were screened with the Spielberger Trait Anxiety Inventory (Spielberger et al., 1983). Participants who scored high in trait anxiety (upper 15th percentile) and subjects with normative levels of trait anxiety (40th to 60th percentile) were selected for further screening. Approximately 10% of these were willing and eligible to participate in a functional magnetic resonance imaging (fMRI) study. Of these, 24 high trait anxious participants and 24 normative trait anxious participants completed the emotional face task on a 3T scanner. The 24 high trait anxious participants (19 female) were aged 19.0 ± 1.8 (range 18–26). Self-reported ethnicities were 12 Caucasian, four Filipino, three Asian, two Latino, one African-American, and two Other. Average years of education completed was 12.7 (SD = 1.0). Of the 24 normative trait anxious participants who completed the task, three were excluded for excessive motion during scanning. The 21 normative trait anxious participants included in the analyses were aged 18.5 ± 0.8 (range 17–20) and included 14 females. Self-reported ethnicities were 16 Caucasian, two Filipino, two Latino, and one Asian. Average years of education completed was 12.4 (SD = 0.5). Age, years of education, ethnicity and gender did not differ significantly between high and normative trait anxiety groups (ps > .2; Table 1).

Participants underwent a semi-structured diagnostic interview (SCID, First et al., 2002). None of the normative trait anxious participants met criteria for any current DSM-IV Axis I diagnosis. Of the 24 high trait anxious participants included in the study, two did not
felt in interpersonal situations (Heimberg et al., 1993). The Physical Social Interaction Anxiety Scale, a 20-item self-report measure of anxiety of the fear of anxiety-related sensations (Reiss et al., 1986), and the also completed the Anxiety Sensitivity Index, a 16-item self-report measure of anxiety.

**Measures**

In addition to the Spielberger Trait Anxiety Inventory, participants also completed the Anxiety Sensitivity Index, a 16-item self-report measure of the fear of anxiety-related sensations (Reiss et al., 1986), and the Social Interaction Anxiety Scale, a 20-item self-report measure of anxiety in interpersonal situations (Heimberg et al., 1993). The Physical Concerns subscale of the Anxiety Sensitivity Index was used for all analyses, in order to better differentiate physical from social concerns (the Anxiety Sensitivity Index also has a Social Concerns subscale). The coefficients of variance for the trait anxiety, social anxiety, and anxiety sensitivity measures were 25%, 59%, and 72% respectively. In addition to these primary measures of interest, participants also completed the Beck Depression Inventory (Beck et al., 1988), a 21-item measure of depressive symptoms. Because scores from the Beck Depression Inventory were highly correlated with the Spielberger Trait Anxiety Inventory in this sample ($r = 0.86$, $p < .001$), we excluded the depression scale as a covariate in order to better focus our analyses on anxiety. The high correlation between trait anxiety and depression supports the notion that individuals who with greater scores on these measures are best characterized as having greater negative affectivity.

**Task**

Participants were trained on a modified version of a widely used emotional face task (Hariri et al., 2002; Paulus et al., 2005; see Fig. 1) prior to testing during fMRI scanning. During each 5 s trial, participants were to press the left or right key on a button box to indicate which of two response options presented at the bottom of the screen matched the target presented at the top of the screen. In the Face condition, stimuli were emotional faces from a standardized set (Matsumoto and Ekman, 1998), and participants were instructed to match based on the emotional expression of the target face. Trials were presented in blocks of six consecutive trials in which the target face was angry, fearful, or happy. Each block was evenly divided between the two possible response option combinations (i.e., three trials of each face pair combination per block). In the sensorimotor control, or Shape condition, stimuli were wide or tall ovals or circles and participants were instructed to match based on shape. Each block of Faces or Shapes was presented three times in a pseudo-randomized order. A fixation cross was presented for 8 s between each block and at the beginning and end of the task (14 fixation periods total). Response accuracy and reaction time was obtained for each trial. The task and scan lasted 512 s (8 min and 32 s).

**Image acquisition**

During fMRI scanning, blood-oxygen-level-dependent (BOLD) signal was collected using a Signa Excite (GE Healthcare, USA) 3.0 T scanner (T2*-weighted echo planar imaging [EPI], repetition time = 2000 ms, echo time = 32 ms, field of view = 250 × 250 mm, 64 × 64 matrix, 30 2.6 mm axial slices with 1.4 mm gap, 256 repetitions). During the same session, a high resolution T1-weighted image (172 sagittally acquired spoiled gradient recalled [SPGR] 1 mm thick slices, inversion time = 450 ms, repetition time = 8 s, echo time = 4 ms, flip angle = 12°, field of view = 250 mm × 250 mm, 256 × 256 matrix) was obtained for anatomical reference.

**Image processing**

All structural and functional image processing was done with the Analysis of Functional Neuroimages (AFNI) software package (Cox, 1996) and R statistical package (http://cran.r-project.org). Functional voxels were 4 × 4 × 4 mm (64 mm³ or 64 μL). Those time points with over two standard deviations more outlier voxels than the subject’s mean were excluded from analysis (as determined by the AFNI function 3dToutcount). Voxel time series were interpolated to correct for

**Table 1**

Demographic and clinical features of participants.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Anxiety-normative (N = 21)</th>
<th>Anxiety-prone (N = 24)</th>
<th>Analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>SD</td>
<td>Mean</td>
</tr>
<tr>
<td>Demographics</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>18.5</td>
<td>0.7</td>
<td>19.0</td>
</tr>
<tr>
<td>Education (years)</td>
<td>12.4</td>
<td>0.5</td>
<td>12.7</td>
</tr>
<tr>
<td>Anxiety</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Trait anxiety</td>
<td>34.5</td>
<td>5.5</td>
<td>53.5</td>
</tr>
<tr>
<td>Social anxiety</td>
<td>17.0</td>
<td>8.7</td>
<td>35.3</td>
</tr>
<tr>
<td>Anxiety sensitivity—physical</td>
<td>6.5</td>
<td>5.4</td>
<td>11.2</td>
</tr>
<tr>
<td>Anxiety sensitivity—total</td>
<td>13.9</td>
<td>7.1</td>
<td>22.6</td>
</tr>
<tr>
<td>Depression</td>
<td>3.8</td>
<td>3.5</td>
<td>15.6</td>
</tr>
</tbody>
</table>

Fig. 1. Design of the emotional face processing task. The left half of the figure is an example of a trial in the Face condition, and the right half of the figure demonstrates a trial in the Shape condition. During each 5 s trial, participants were to press the left or right key on a button box to indicate which of two response options presented at the bottom of the screen matched the target at the top of the screen.
non-simultaneous slice acquisition within each volume and corrected for three-dimensional motion. Anatomical and echo planar volumes were co-registered using an algorithm that minimizes the amount of image translation and rotation (Saad et al., 2009). Three normative trait anxious participants were excluded for excessive motion during the task, defined as maximum movement greater than 4 mm and average displacement from the co-registration base image greater than 1 mm.

Individual participant time series data were analyzed with AFNI’s 3dDeconvolve program. The orthogonal regressors of interest were the happy, angry, fearful, and shape blocks. Regressors were convolved with a modified gamma variate function to account for the hemodynamic response (Friston et al., 1995; Cohen, 1997). The time series alignments in the roll, pitch, and yaw directions were used to create motion regressors for each participant, which were included in the general linear model as nuisance regressors to account for motion artifacts. Additional regressors were used to model the baseline, linear, and quadratic trends in the time series. Following deconvolution, data were converted to percent signal change, which was obtained by dividing the regressor coefficient by the zero-order regressor within each voxel. Data were normalized to Talairach coordinates and subjected to a 4 mm Gaussian blur for spatial smoothing.

Analysis was limited to a priori regions of interest using masks defined by the Talairach atlas (Lancaster et al., 2000; Talairach and Tournoux, 1988) of the bilateral amygdala, insula, medial prefrontal cortex (mPFC), and fusiform gyrus. To protect for multiple comparisons, Monte Carlo simulations (N = 10,000) using AFNI’s AlphaSim program demonstrated that for a per voxel threshold of α = .01 (one-sided), minimum volume thresholds of 152 mm³ (3 voxels) in the amygdala and 256 mm³ (4 voxels) in the insula, fusiform, and ventromedial prefrontal cortex would preserve α < .05 at the cluster level.

Statistical analysis

Analyses of self-report and behavioral data were performed with PASW Statistics (Release Version 18.0.0, Chicago IL). For the imaging analysis, the main contrast of interest was face blocks versus shape blocks. A secondary contrast of interest was negative (angry, fearful) versus positive (happy) face blocks. The primary analysis was a robust multiple regression, in which BOLD percent signal change was entered as the dependent variable, and trait anxiety, social anxiety and anxiety sensitivity to physical symptoms were the independent variables. This was implemented using the statistical programming language R (http://cran.r-project.org), specifically the rlm program from the robustbase library. Robust regressions are inherently more conservative and remove bias from outliers by weighting each observation based on the distribution of the data (Huber, 1973). As such they have been recommended for use in neuroimaging analysis (Wager et al., 2005). To obtain unbiased estimates, t-statistics for each coefficient were computed using a bootstrap estimate of each partial regression coefficient’s standard error. This procedure was performed using the boot library of R statistical package.

In order to buttress our results, we considered two complementary approaches. First, to confirm activations seen previously, task effects were examined with two one-sample t-tests across all participants, one examining the effects of face versus shape blocks, the other comparing negative versus positive faces. Average activation across all voxels in each significant task–effect cluster was then used as the dependent variable in a regression. Results from this task–effect region of interest analysis are reported along with the task effects themselves in Supplemental Tables 1 and 2. However, this approach limits the search space for associations with anxiety-related constructs to regions that are commonly recruited across participants. It is also likely that certain types of participants (e.g., those high on one facet of anxiety) recruit different regions altogether, or activate certain regions to a greater or lesser spatial extent. The approach described above would be insensitive to such effects, as the new regions or additional voxels would likely not emerge as significant across all participants when defining the functional regions of interest.

Thus a second approach was implemented, to fully explore the anatomical regions of interest. Specifically, a voxel-wise regression was performed within the anatomically defined regions of interest, and joint probability and volume thresholds (as described above) were used to control for type I error. In order to not artificially inflate correlations due to averaging activation across pre-selected voxels, the voxel-wise regression coefficients and t-statistics were averaged across all voxels within a cluster (see Tables 2 and 3). In addition, three robust simple regressions, using each independent variable as a single predictor of BOLD percent signal change, were also implemented on a voxel-wise basis. After identifying functional regions of interest that were associated with specific domains of anxiety, we explored associations between activity in previously identified regions and individual difference variables such as task performance (i.e., action time and accuracy). Finally, to better understand results from the negative versus positive face contrast, secondary regression analyses examining negative faces versus shape blocks and positive faces versus shape blocks were conducted.

Results

Self-report data and behavioral data

The self-report measures of the three components of anxiety—trait anxiety, social anxiety, and anxiety sensitivity—were all significantly inter-correlated across the 45 participants (r = .45–.57, all ps < .001). There were no significant correlations between either trait anxiety or social anxiety and either response time or accuracy to any of the four conditions—shapes, angry targets, fearful targets, or happy targets (all ps > .15). Interestingly, greater anxiety sensitivity was associated with slower reaction times to fearful target faces (r = .31, p < .05)

Table 2

<table>
<thead>
<tr>
<th>Region</th>
<th>Peak x</th>
<th>Peak y</th>
<th>Peak z</th>
<th>Vol</th>
<th>Trait anxiety coefficient (t-stat)</th>
<th>Social anxiety coefficient (t-stat)</th>
<th>Anxiety sensitivity coefficient (t-stat)</th>
</tr>
</thead>
<tbody>
<tr>
<td>R anterior insula</td>
<td>34</td>
<td>34</td>
<td>0</td>
<td>320</td>
<td>-.012 (-2.22)</td>
<td>.010 (2.88)*</td>
<td>-.0002 (0.14)</td>
</tr>
<tr>
<td>R posterior insula</td>
<td>42</td>
<td>-13</td>
<td>-8</td>
<td>256</td>
<td>-.010 (-2.17)</td>
<td>.008 (2.70)*</td>
<td>.0002 (0.04)</td>
</tr>
<tr>
<td>L amygdala</td>
<td>-26</td>
<td>-9</td>
<td>-16</td>
<td>256</td>
<td>-.006 (-1.45)</td>
<td>.007 (4.50)*</td>
<td>.002 (0.33)</td>
</tr>
<tr>
<td>Subgenual ACC</td>
<td>-2</td>
<td>3</td>
<td>-8</td>
<td>256</td>
<td>-.007 (-1.98)</td>
<td>.008 (3.04)*</td>
<td>-.011 (-1.85)</td>
</tr>
<tr>
<td>Rostral ACC</td>
<td>-10</td>
<td>43</td>
<td>12</td>
<td>832</td>
<td>-.001 (-0.44)</td>
<td>-.001 (-1.04)</td>
<td>.009 (2.94)*</td>
</tr>
<tr>
<td>Dorsal ACC</td>
<td>14</td>
<td>11</td>
<td>24</td>
<td>768</td>
<td>-.003 (-1.24)</td>
<td>-.002 (-1.12)</td>
<td>.009 (3.33)*</td>
</tr>
<tr>
<td>V dorsal ACC</td>
<td>-10</td>
<td>35</td>
<td>24</td>
<td>512</td>
<td>-.003 (-1.49)</td>
<td>.001 (0.31)</td>
<td>.010 (2.80)*</td>
</tr>
</tbody>
</table>

Note. Unstandardized beta coefficients and their associated t-statistics are presented. Voxel size is 4 x 4 x 4 mm. Vol = volume in mm³, R = right, L = left, ACC = anterior cingulate cortex.

* p < .05.
and lower accuracy to all four conditions ($r = -0.33$ to $-0.44$, all $p$s < .05). Independent sample t-tests between the anxiety-normative and anxiety-prone groups did not reveal significant differences in response time or accuracy to any of the four conditions (all $p$s > .35, see Fig. 2).

**Task effects**

Greater activation to faces than shapes was observed in bilateral fusiform gyrus, bilateral posterior and anterior insula, bilateral amygdala, and subgenual anterior cingulate cortex (ACC). Greater activation to shapes than faces was observed in rostral ACC and medial PFC (Brodmann area 10). Of these significant task effect regions, only left amygdala and left anterior insula demonstrated a significant relationship with any of the three independent variables. In both these regions, differential activity to faces versus shapes was significantly associated with social anxiety, controlling for trait anxiety and anxiety sensitivity. In addition, there was a trend ($p < .07$) for greater activation with higher levels of social anxiety in the right anterior insula. Full results are presented in Supplemental Table 1.

In the valence contrast, negative target faces (fearful, angry) yielded greater activation than happy target faces in bilateral anterior insula, bilateral fusiform gyrus, dorsal ACC, and right amygdala. A region of medial PFC demonstrated greater activation in happy than negative target faces. Of these significant task effect regions, only left anterior insula and dorsal ACC demonstrated a significant relationship with any of the three independent variables. In both these regions, differential activity to negative versus positive faces was significantly associated with trait anxiety, controlling for social anxiety and anxiety sensitivity (see Supplemental Table 2).

### Voxel-wise robust multiple regression: faces versus shapes

There were no significant associations in the simple regression between trait anxiety and BOLD response to emotional faces within medial PFC, amygdala, insula, or fusiform gyrus. A robust multiple regression controlling for trait anxiety and anxiety sensitivity revealed significant associations between social anxiety and greater BOLD response to emotional faces in left amygdala, right anterior and posterior insula, and subgenual ACC (Fig. 3A). When controlling for trait anxiety and social anxiety, anxiety sensitivity was significantly associated with greater activity in dorsal and rostral ACC (Table 2).

### Voxel-wise robust multiple regression: valence contrast

Significant associations between trait anxiety and differential BOLD response to negative versus positive face targets were found in the simple regression in bilateral posterior insula, left anterior insula, left amygdala, and dorsal ACC. Of these, only differential BOLD responses in left anterior insula, left amygdala, and dorsal ACC demonstrated significant associations with trait anxiety when controlling for social anxiety and anxiety sensitivity in the multiple regression (Fig. 3B). When controlling for trait anxiety and anxiety sensitivity, social anxiety was significantly associated with greater BOLD response to negative (versus positive) faces in right posterior insula, and right amygdala. When controlling for trait anxiety and social anxiety, anxiety sensitivity was significantly inversely associated with BOLD response to negative (versus positive) faces in subgenual ACC (Table 3).

To better understand these results, the relationships between the three anxiety domains and BOLD response was determined for negative faces versus shapes and positive faces versus shapes separately, in each of the regions listed in Table 3. This analysis revealed that for left amygdala and left anterior insula, the association with trait anxiety was primarily driven by an inverse relationship between trait anxiety and BOLD response during positive face target blocks. No other regions
Fig. 3. Activations associated with specific domains of anxiety. A: Response in left amygdala and right anterior insula to emotional faces was significantly associated with social anxiety. B: Response in left amygdala and left anterior insula to negative faces was significantly associated with trait anxiety. Note. The scatterplots show simple correlations of activity from brain region identified in the multiple regression against anxiety scores. In other words, while neural activity was associated with one domain of anxiety when controlling for the other two domains, the scatterplot does not adjust or control for other scores.
listed in Table 3 demonstrated significant relationships between either negative faces versus shapes or positive faces versus shapes and any of the three anxiety domains.

Additional brain–behavior relationships

We explored associations between brain activity in the functional regions of interest from the multiple regression and task performance (reaction time and accuracy). The region of rostral ACC that was specifically associated with anxiety sensitivity was also inversely associated with accuracy to both the face ($r = -0.38, p < 0.05$) and shape ($r = -0.39, p < 0.05$) conditions. No other regions that were significantly associated with an anxiety-related construct (i.e., listed in Table 2) were associated with accuracy or reaction time to faces or shapes.

Several participants in our sample met criteria not only for an anxiety disorder, but also for major depression, which has previously been associated with a bias for negative information. Therefore we also examined the relationship between brain activity during the valence contrast and depressive symptoms. There was a significant relationship between depression symptom scores and activity to negative faces in the three regions that were specifically associated with trait anxiety: left amygdala ($r = 0.45, p < 0.005$), left insula ($r = 0.38, p < 0.05$), and dorsal ACC ($r = 0.35, p < 0.05$). These results were expected due to the large correlation between depression symptoms and trait anxiety in this sample. The other regions listed in Table 3 were not associated with depressive symptoms.

Discussion

The present study aimed to parse the effects of trait anxiety, social anxiety, and anxiety sensitivity on neural responses to emotional faces, by including all three domains of anxiety simultaneously in a multiple regression to predict neural activity. We focused our analyses on key affective circuitry—amygdala, insula, and medial PFC—with fusiform gyrus as a visual processing control region. We hypothesized that amygdala activation to emotional faces would specifically relate to social anxiety, whereas insula activation would relate to anxiety sensitivity.

Selective effects of social anxiety

In agreement with our hypothesis, we found that left amygdala activity during emotional face processing was associated with social anxiety, even controlling for trait anxiety and anxiety sensitivity. This result was present both when performing the regression on average activity from the task effect region of interest, as well as in the voxel-wise search. However, we found that insula activity was associated with social anxiety, rather than anxiety sensitivity as we had predicted. Anxiety sensitivity was associated with rostral and dorsal ACC activation.

These results suggest that the heightened activity in amygdala and insula during emotional face processing that is commonly observed in anxious individuals may be attributable to the extent to which such individuals are anxious about social judgment in particular. This result might help explain divergent findings in the literature. A previous study of high trait anxious young adults, using the same task, found that trait anxiety was associated with greater activity in amygdala and insula in response to emotional faces (Stein et al., 2007). We have not replicated this in the present sample. However, given that we found amygdala and insula activity to be related specifically to the domain of social anxiety, differences in sample composition on social anxiety levels could explain inconsistent levels of association between trait anxiety or negative affectivity and neural activity. Social anxiety disorder is a common condition (lifetime prevalence 12%) that is frequently comorbid with other mood and anxiety disorders (Ruscio et al., 2008).

Concerns about social judgment are likely to be present not only in social anxiety samples, but in other anxiety populations as well. The relationship between social anxiety and activity in the amygdala and insula is consistent with the role of these regions in affective processes. The amygdala has long been associated with fear conditioning (Davis and Whalen, 2001), and is especially engaged by social threat (Haxby et al., 2002). Accordingly, this may underlie the relationship we report here between activity in the amygdala and the extent to which participants are concerned with social judgment (and may therefore see emotional facial expressions as more threatening). Similarly, the insula has been shown to be a critical structure for interoception, the perception of internal bodily sensations (Craig, 2009; Paulus and Stein, 2006). Although we expected the insula to be associated with sensitivity to anxiety sensations, its association with social anxiety might reflect greater physiological arousal in participants with high levels of social anxiety. It is possible that the insula would show a relationship with anxiety sensitivity if physical sensations of anxiety were more strongly elicited across all participants.

Selective effects of anxiety sensitivity

The association between anxiety sensitivity and ACC activity was not hypothesized. The ACC is thought to support a complex set of cognitive processes, including error detection, detection of conflict between response options, and regulation of arousal and stress (Bush et al., 2000; Paus, 2001). It is possible that individuals high on anxiety sensitivity may be hypervigilant towards arousal, and may over-engage ACC in support of this vigilance. Alternatively, altered functioning in ACC may lead to dysregulation of stress and arousal, thereby generating greater anxiety sensitivity. In addition, both anxiety sensitivity and rostral ACC activity were inversely associated with accuracy of matching emotional facial expressions in this sample. Individuals who are high on anxiety sensitivity may be more distracted by symptoms of anxiety that arise from the experimental situation, and this may result in lower accuracy. However it should be noted that differences in accuracy were extremely small, with high overall accuracy (see Fig. 2). These speculations notwithstanding, the role of the ACC in this context, and its relationship to anxiety sensitivity remains unclear.

Selective effects of trait anxiety

We were also interested in how specific anxiety domains relate to negatively compared to positively valenced faces. We found that the differential neural response to negative versus positive faces in amygdala, insula, and dorsal ACC was associated with trait anxiety. This result in insula and dorsal ACC was present both when performing the regression on average activity from the task effect region of interest, as well as in the voxel-wise search. The association between the trait anxiety measure and differential responses to negative and positive faces may be due to the relationship between trait anxiety and general negative affectivity (Bieling et al., 1998). This is supported by the fact that regions specifically associated with trait anxiety were also positively related to symptoms of depression, an expected result given the high correlation between depression symptoms and trait anxiety in this sample. This conclusion is also supported by the inverse relationship between trait anxiety and positive (i.e., happy) faces. Therefore this trait is best referred to as general negative affectivity, a construct that underlies both anxiety and depression (Watson et al., 1988).

Conclusions

The current results support the hypothesis that whereas individuals with social anxiety exhibit increased sensitivity to all emotional faces, people with high trait anxiety may show a bias towards processing negative faces or against processing positive faces. These results were supported in both complementary analysis approaches, with the
This finding is consistent with an emerging literature emphasizing that social anxiety involves not only fear of negative evaluation, but fear of positive evaluation as well (Weeks et al., 2008). Positive evaluation (e.g., happy facial expressions) can signal not only positive regard, but also a social interaction that must be navigated in order to avoid embarrassment. The association between high trait anxiety and increased differential neural activity to negative over positive facial expressions is consistent with the idea that trait anxiety may also be more related to general negative affectivity than specifically to anxiety per se (Bieling et al., 1998). This is further supported by the high correlation between trait anxiety and depressive symptoms in this sample. Trait anxiety, depression, and negative affectivity have all been associated with a negative information processing bias (Beck and Clark, 1988; Chan et al., 2007; Macleod and Rutherford, 1992), and anhedonia has been associated with decreased neural response to positive stimuli in the amygdala (Keedwell et al., 2005), consistent with the present findings.

**Limitations and future directions**

Anxiety is a complex phenomenon and can be parsed into different anxiety-related constructs that ultimately may relate to specific anxiety disorders. Here we have only examined three common domains: negative affectivity, social anxiety, and anxiety sensitivity. Future work could focus on other related domains such as worry, avoidance, hypervigilance, or neuroticism. Although measures for these constructs are often highly inter-correlated, the approach proposed here may be a first step towards identifying the specific neural structures and processes that differentiate specific domains of anxiety. We would hypothesize that the increased activity within neural structures that are related to specific anxiety constructs may play a critical role in the development and maintenance of anxiety disorders, and would encourage longitudinal research along these lines. It will also be critical to examine emotional and cognitive processes that are common across anxiety domains, or integrate across anxiety-related constructs. This would allow both for localization of function to specific brain areas and networks, but also more precise association of anxiety problems to specific processing abnormalities.

We limited our analysis to four a priori regions of interest—medial PFC, amygdala, insula, and fusiform gyrus. Although these are key regions in understanding the neural substrates of anxiety disorders, other circuits are also important to examine. For example, prefrontal regions implicated in emotion regulation and their functional connectivity to affective circuitry would be important targets of future investigation. Furthermore, the task utilized an explicit evaluation of an emotional stimulus. Other emotion processing paradigms have used incidental emotional processing (e.g., matching emotional faces by gender), or emotion detection within the self or another. The neural substrates underlying diverse types of emotion processing have been shown to differ, with amygdala activity across most paradigms, and insula and rostral ACC activity greatest during the evaluation of one’s own emotion (Lee and Siegle, in press). Prior investigations of emotion-al face processing have also differentiated between direct and averted eye gaze in fearful and angry facial expressions, with greater amygdala activity to fearful faces with direct gaze, and angry faces with indirect gaze (Adams et al., 2003). Future work may benefit from examining relationships between domains of anxiety and the distinct types of emotion and threat processing described above.

The present sample was comprised of college undergraduates, and was therefore relatively homogeneous with regards to age and education, which potentially reduces the generalizability of these findings. However, young adults who experience high anxiety but are not seeking treatment are a key population to study given the typical timeline of anxiety disorder onset (Pine et al., 1998). Future work should explore whether activity in amygdala, insula, or medial PFC can help differentiate individuals with different types of anxiety, and predict which anxious young adults will go on to develop disorders and seek treatment.

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