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Functioning of Neural Systems Supporting Emotion Regulation in Anxiety-Prone Individuals

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Abstract

Previous neuroimaging studies suggest that prefrontal cortex (PFC) modulation of the amygdala and related limbic structures is an underlying neural substrate of effortful emotion regulation. Anxiety-prone individuals experience excessive negative emotions, signaling potential dysfunction of systems supporting down-regulation of negative emotions. We examined the hypothesis that anxious individuals require increased recruitment of lateral and medial PFC to decrease negative emotions. An emotion regulation task that involved viewing moderately negative images was presented during functional magnetic resonance imaging (fMRI). Participants with elevated trait anxiety scores (n = 13) and normal trait anxiety scores (n = 13) were trained to reduce negative emotions using cognitive reappraisal. Blood oxygenation level-dependent (BOLD) changes were contrasted for periods when participants were reducing emotions versus when they were maintaining emotions. Compared to healthy controls, anxious participants showed greater activation of brain regions implicated in effortful (lateral PFC) and automatic (subgenual anterior cingulate cortex) control of emotions during down-regulation of negative emotions. Left ventrolateral PFC activity was associated with greater self-reported reduction of distress in anxious participants, but not in healthy controls. These findings provide evidence of altered functioning of neural substrates of emotion regulation in anxiety-prone individuals. Anxious participants required greater engagement of lateral and medial PFC in order to successfully reduce negative emotions.

Keywords

Emotion; Emotion Regulation; fMRI; Anxiety; Prefrontal Cortex; Anterior Cingulate Cortex

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1. Introduction

Emotion regulation refers to a diverse set of processes that influence the occurrence, intensity, duration, and expression of emotion (Gross and Thompson, 2007). Effortful regulation of negative emotions constitutes one subtype of emotion regulation, in which individuals attempt to reduce negative emotions using deliberate strategies (e.g., distraction, re-interpretation). Empirical investigation has largely focused on this subtype of emotion regulation, given its relevance to emotional disorders and the potential that findings will translate to clinical intervention (Campbell-Sills and Barlow, 2007; Mennin et al., 2005).

Neuroimaging research has revealed neural substrates that contribute to effortful regulation of negative emotions (Ochsner et al., 2002, 2004; Levesque et al., 2003, 2004; Kalisch et al., 2005; Phan et al., 2005; Beauregard et al., 2006; Johnstone et al., 2007; Goldin et al., 2008, 2009a, 2009b; New et al., 2009; Koenigsberg et al., 2009, 2010). Many studies have focused on cognitive reappraisal, an effective strategy (Gross, 1998; Gross and John, 2003) that entails reinterpreting the meaning of a stimulus in such a way that its emotional impact is diminished. Converging evidence suggests that lateral and medial regions of the prefrontal cortex (PFC) [e.g., dorsolateral prefrontal cortex (DLPFC); dorsal anterior cingulate cortex (dACC)] down-regulate neural substrates that are primary emotion processing areas (e.g., amygdala) during cognitive reappraisal (Ochsner et al., 2002, 2004; Levesque et al., 2003; Kalisch et al., 2005; Phan et al., 2005; Goldin et al., 2008; Koenigsberg et al., 2010).

With the literature broadly supporting this model of cognitive reappraisal, investigation has recently shifted toward exploring individual differences in the functioning of neural circuitry that supports this strategy. Studies have now examined the functional differences between healthy individuals and patients suffering from major depressive disorder (MDD; Beauregard et al., 2006; Johnstone et al., 2007) social anxiety disorder (SAD; Goldin et al., 2009a, 2009b), posttraumatic stress disorder (PTSD; New et al., 2009), and borderline personality disorder (Koenigsberg et al., 2009). Although sufficient converging evidence is not yet available to support neural systems models of emotion regulation deficits associated with these disorders, several interesting findings have emerged. Studies of subjects with MDD have provided evidence of hyperactivity of “cognitive control” regions such as ventrolateral PFC (VLPFC; Johnstone et al., 2007) and dorsal anterior cingulate cortex (dACC; Beauregard et al., 2006) during down-regulation of negative emotions. A study of subjects with SAD also showed increased engagement of right DLPFC during regulation of responses to “physical threat” stimuli; however, during regulation of responses to “social threat” stimuli (which are most relevant to their disorder), socially anxious subjects engaged cognitive control regions (e.g., DLPFC, dACC) to a lesser degree than controls (Goldin et al., 2009a). Another study also showed decreased PFC recruitment in patients with PTSD versus controls during down-regulation of emotional responses to negative pictures (New et al., 2009).

The current study aimed to add to the emerging literature on individual differences in the functioning of neural systems supporting emotion regulation, by characterizing differences between subjects with high and normal levels of trait anxiety. The current sample differs from previous samples of patients with specific anxiety and mood disorders: the subjects recruited for the current study, while endorsing high levels of trait anxiety, have no history of treatment for anxiety or other emotional problems. Additionally, they do not self-identify as having a particular disorder (although the majority meet criteria for anxiety disorders upon examination). This sample therefore represents a different segment of the anxiety severity spectrum than previous studies of specific patient groups, and offers an opportunity to observe differences associated with anxiety status that are not confounded by general illness factors such as treatment history.

Subjects scoring in the high and normal ranges on a measure of trait anxiety were trained in cognitive reappraisal, after which they performed an emotion regulation task while undergoing functional magnetic resonance imaging (fMRI). We hypothesized that anxious participants would be able to learn and apply reappraisal, but would require greater top-down control to accomplish down-regulation of negative emotions. Thus, we expected to observe PFC hyperactivity in anxious participants during attempts to reduce negative emotions. We also predicted less attenuation of amygdala response in anxious participants during down-regulation of emotion (i.e., decreased efficacy of reappraisal).

2. Material and Methods

2.1 Participants

The institutional review boards of University of California San Diego and San Diego State University approved this study. All participants provided written informed consent.

Participants were drawn from a pool of undergraduates (N = 950) who completed questionnaires for course credit that included the State Trait Anxiety Inventory (STAI; Spielberger et al., 1983). Subjects scoring high in trait anxiety (upper 15th percentile of the STAI distribution) and in the normal range (40th–60th percentile) were contacted to assess interest in experimental and fMRI studies, and (if interested) to schedule an eligibility interview.

The eligibility assessment included the Structured Clinical Interview for DSM-IV (First et al., 1996). High Anxiety (HA) subjects could have any anxiety disorder diagnosis (full or subthreshold), but were not currently seeking nor had ever sought treatment in the past. Participants in the Normal Anxiety (NA) group were included if the interview revealed no history of anxiety or mood disorders. Exclusion criteria for both groups were: lifetime psychoactive medication or psychotherapy use, current use of >400 mg caffeine daily, current daily tobacco use, lifetime head injury, current alcohol or substance abuse, lifetime alcohol or substance dependence, active suicidality, and lifetime bipolar or psychotic disorders.

Twenty-six subjects (13 HA, 13 NA) were eligible and completed the study. The majority of the sample was female (n = 22; 84.6%) and the mean age was 19.15 (SD = 1.83). Participants identified themselves as Caucasian (n = 15), Asian (n = 4), Hispanic (n = 3), African American (n = 2), and Other (n = 2). The HA and NA groups did not differ on gender, χ^2 (1, 26) = 0.00, *ns*, age, t (24) = 1.08, *ns*, or ethnicity, χ^2 (4, 26) = 4.07, *ns*.

Upon evaluation, 12 of 13 HA participants met full criteria for one or more anxiety disorders [generalized anxiety disorder (GAD; n = 9); SAD (n = 8); and obsessive-compulsive disorder (n = 1)]. The remaining HA participant had subthreshold GAD and SAD. Three HA participants also reported a major depressive episode in the past six months. Results presented do not change significantly if participants with recent co-occurring depression (or the single participant who did not meet full criteria for an anxiety disorder) are excluded.

2.2 Task

The task was designed to isolate brain activation related to down-regulation of negative emotion. The condition of interest involved instructing participants to reduce their emotions using cognitive reappraisal (“Reduce”). Following two prior investigations (New et al., 2009; Phan et al., 2005), we selected a contrast condition where participants maintained their reactions to negative images (“Maintain”).[1]

The task consisted of 24 trials lasting 9.6 minutes. Before each trial, subjects were cued to either reduce or maintain the emotions elicited by the upcoming image. Subjects also were asked to rate their distress (via a button box) using a 4-point scale (1 = none; 2 = mild; 3 = moderate; 4 = severe) before and during presentation of each image.

Images were selected from the International Affective Picture System (IAPS; Lang et al., 1997). IAPS images are defined by valence (1 to 9; extremely negative to extremely positive) and arousal (1 to 9; no arousal to extreme arousal). Stimuli rated 2 to 3.5 on valence and 5 to 7 on arousal were considered for inclusion. Twenty-four images were selected for the fMRI task, and 10 images for the practice test.

To provide a baseline matched for luminescence and color, each image was subjected to a pixel-wise randomization routine, resulting in a scrambled image that was presented between each trial. The 24 target images were randomly assigned to either Reduce or Maintain (with the constraint that exactly 12 images were assigned to each). We checked to ensure that images assigned to the two conditions did not differ on valence or arousal, and that they contained equivalent portrayals of people, animals, and objects.[2] The images (and corresponding instructions) were arranged in a pseudorandom order with the restriction that no more than 3 consecutive trials of the same type were permitted. Order of presentation was identical for all participants.

Each trial lasted 24 seconds and began with a 12 second “baseline” to give participants time to recover from the previous trial. The visual presentation during the 12-second baseline was the scrambled image. For 1–3 seconds (jittered), participants viewed the scrambled image. Then “Rate Emotion (1–4)” appeared in yellow print below the scrambled image. This instruction lasted 3 seconds and then disappeared. Another 1–3 seconds passed before another instruction appeared. This was the instruction to either “Reduce Emotion” (appearing in blue) or “Keep Up Emotion” (appearing in red) on the next trial. The instruction lasted 3 seconds, then disappeared leaving only the scrambled image for 1–3 seconds.

The visual presentation for the next 12 seconds was the target image. Participants viewed the image for 4–6 seconds, while attempting to reduce or maintain emotions. Then “Rate Emotion (1–4)” appeared in yellow below the image for 3 seconds. The unit of analysis for this study was the 4–6 second period when participants were reducing or maintaining emotions, prior to the cue to rate how they were feeling.[3] This rating instruction disappeared and the target image persisted for 3–5 more seconds. When the target image disappeared, the trial was over. See Figure 1 for the task schematic and regressors for imaging analyses.

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[1]Benefits of using the “Maintain” condition as a comparator included its ability to control for picture viewing and meta-cognition involved in monitoring oneself for adequate task performance. We considered using a contrast condition that involved passive viewing of the stimuli (with no corresponding instructions); however, we opted against this because unlike the “Reduce” condition it would fail to provide any direction for participants’ behavior. Moreover, our pilot studies suggested that some subjects engaged spontaneously in reappraisal during passive viewing, raising the concern that passive viewing would not be adequately differentiated from “Reduce” trials for some subjects.

[2]Most of the images in the task (18 of 24) depicted scenes involving people in stressful or upsetting situations (e.g., being threatened with a weapon; in a hospital; visibly grieving); these were equally balanced across the Reduce (9) and Maintain (9) conditions. Animal images depicted snakes, spiders, and roaches and inanimate images depicted guns, a tornado, and a capsizing boat.

[3]Self-rating of emotions introduces a new task that engages different neural systems and also distracts participants from reducing or maintaining emotions. Therefore, to examine activation associated with Reduce and Maintain, we analyzed only the portion of these trials that occurred prior to the “Rate Emotion” cues. We also subtracted out the portion of the Baseline period that involved viewing and responding to the emotion rating cue (see Figure 1). We thank anonymous reviewers for this useful suggestion.

2.3 Procedure

Participants underwent a 30-minute training by one of the authors (L.C-S. or A.A.R.). For "Reduce" trials, they were taught to generate interpretations of the images that would help reduce negative emotions. For example, for a picture of a destroyed building, they might generate a thought about the building being torn down by a construction crew (less negative interpretation) instead of a thought about a bomb having destroyed the building (more negative interpretation). For "Maintain" trials, they were told to "notice what you are feeling without trying to change it" and to "maintain your emotional reaction until the picture disappears." See Supplement 1 for the handout that the experimenters used as a basis for the training session.

Subjects completed a 10-trial practice test, which familiarized them with the task and allowed the experimenter to determine whether they had become proficient in reappraisal. Subjects performed the same task they would in the scanner but verbalized their thought process for each trial. The experimenter gave corrective feedback. Subjects who had difficulty (2 HA subjects and 1 NA subject) repeated the handout and practice test. All participants were judged proficient by the end of two practices. They received an MRI orientation and completed the regulation task while undergoing fMRI.

2.4 Image Acquisition

During the task, one BOLD fMRI run was collected for each participant using a Signa EXCITE 3.0 Tesla-GE scanner (T2*-weighted echo planar imaging, TR=2000 ms, TE=32 ms, FOV = 230×230 mm^3, 64 × 64 matrix, thirty 2.6mm axial slices with a 1.4mm gap, 290 scans). For anatomical reference, a high resolution T1-weighted image (SPGR, TI = 450, TR = 8 ms, TE = 4 ms, FOV = 250×250 mm^3, flip angle = 12°, 172 sagittally acquired slices, ~1 mm^3 voxels) was obtained during the same session. For preprocessing, voxel time series were interpolated to correct for non-simultaneous slice acquisition within each volume. These interpolated values were then corrected for six-dimensional motion (3 linear and 3 rotational dimensions).

2.5 Image Processing and Analysis

All structural and functional image processing was done with Analysis of Functional Neuroimages (AFNI) software (Cox, 1996). Echoplanar images were realigned to a base using a Fourier transform—using the AFNI program 3dvolreg—and then time-corrected for slice acquisition order. The base image was selected from the middle acquisition of consistent high quality images using the AFNI program 3dToutcount. This resulted in the selection of the middle of the entire run (i.e., 145th acquisition) in almost all subjects.

Preprocessed time series data for each individual were analyzed using a multiple regression model. For this model, five orthogonal regressors of interest were created: (1) Reduce (the time between the onset of the image and the prompt to rate emotion during Reduce trials), (2) Post-Rating Reduce (the time between the offset of the prompt to rate emotion and the disappearance of the image during Reduce trials), (3) Maintain (the time between the onset of the image and the prompt to rate emotion during Maintain trials), (4) Post-Rating Maintain (the time between the offset of the prompt to rate emotion and the disappearance of the image during Maintain trials), and (5) all emotion rating periods regardless of whether they occurred during baseline periods, Reduce trials, or Maintain trials. In subsequent analyses, Reduce and Maintain were analyzed and contrasted. Regressors were convolved with a modified gamma variate function to account for the delay and dispersion of the hemodynamic response of the BOLD-fMRI signal. Additionally, five nuisance regressors were used to account for residual motion (roll, pitch, and yaw) and to eliminate slow signal drifts (baseline and linear trend). The ten regressors were applied to the AFNI program

3dDeconvolve to calculate the estimated voxelwise response amplitude. The resultant weightings were divided by the baseline regressor to determine the percent signal change. This percent signal change was used for all subsequent analyses. To account for individual variation of anatomical landmarks, a Gaussian filter with 6mm full width at half maximum was applied to the voxelwise percent signal change data.

Data for each participant were normalized to Talairach coordinates (Talairach, 1998). Whole brain analyses were corrected for multiple comparisons by clustering thresholds determined by the AFNI function AlphaSim; the resulting minimum cluster volume was 1408 ml. Analyses were conducted with a voxel-wise *a priori* probability of 0.01; this threshold for significance was maintained for the cluster using the threshold determined by AlphaSim.

2.6 Statistical Analyses

For the main between-groups analyses, the voxel-wise percent signal change data were entered into a mixed-model analysis of variance (ANOVA) with task contrast (Reduce vs. Maintain) and group (HA vs. NA) as fixed factors and participants as a random factor. For regions where a significant between-groups difference was found on the Reduce-Maintain contrast, the BOLD signal was extracted separately for the Reduce and Maintain conditions to clarify the nature and direction of the effect. Analyses of the relationships between BOLD response and distress ratings were conducted with SPSS 12.0.

3. Results

3.1 Group Differences in Self-Reported Distress

Bonferroni *t* tests ($\alpha = .0125$ to account for 4 comparisons based on the distress ratings) were used to examine the effect of group (HA vs. NA) on distress ratings during Baseline, Reduce, and Maintain periods (see Figure 2). HA subjects reported higher levels of distress during Baseline periods, $t\ (24) = 3.04$, $p < .01$. The HA and NA groups did not differ significantly on levels of distress reported during Reduce, $t\ (24) = 0.83$, *ns*, or Maintain periods, $t\ (24) = 2.29$, $p = .03$ (non-significant with Bonferroni correction).

To provide another index of subjects' success in applying cognitive reappraisal, we computed the percent reduction in distress they were able to attain during Reduce trials compared to Maintain trials. Percent reduction in distress was calculated for each subject as [(Mean rating during Reduce trials − Mean rating during Maintain trials)/Mean rating during Maintain trials] × −100. The groups did not differ on percent reduction in distress, $t\ (24) = 0.37$, *ns*, with the HA and NA groups achieving 33.6% and 31.8% mean reductions in distress, respectively. Overall, results suggest that HA and NA subjects were equally effective in using cognitive reappraisal to reduce distress.

3.2 Task Effects on BOLD Activity – Total Sample

Whole brain analyses showed that a distributed set of brain regions was more active during Reduce and Maintain trials when each was compared to Baseline periods (see Supplements 2 and 3). Regions that were engaged during Reduce periods included bilateral occipital cortex, bilateral thalamus, left anterior insula, and right dorsolateral PFC. Regions that were engaged during Maintain periods included bilateral occipital cortex, left thalamus, dACC, right anterior insula, and right lateral PFC.

Reduce and Maintain trials were contrasted in the total sample to isolate regions engaged by efforts to down-regulate negative emotion (Table 1 and Figure 3). The contrast revealed one cluster in right DLPFC (BA 9) that was more active during Reduce trials than Maintain trials.

3.3 Group Differences in BOLD Activity on the Reduce-Maintain Contrast

Between-group differences on the Reduce-Maintain contrast were the main focus of this investigation (Table 2 and Figure 4). Whole brain analysis revealed between-group differences in bilateral cuneus (BA 18), dorsomedial PFC/dACC (BA 6/32), and sgACC (BA 25); left DLPFC (BA 8/9), left ventrolateral PFC (VLPFC; BA 47), and right VLPFC (BA 44/45). The HA group demonstrated greater activation during Reduce trials than the NA group in all of these regions. NA subjects recruited some of these regions to a greater extent during Maintain trials (see bar graphs in Figure 4).

3.4 Relationship Between Percent Reduction in Distress and BOLD Activity

Signal change averaged across all voxels in functionally defined regions of interest (i.e., significant clusters on the between-groups Reduce-Maintain contrast) was correlated with percent reduction in distress. Spearman's rho was computed for the total sample and within the NA and HA groups. In the HA group, subjects with greater percent reduction in distress showed greater left VLPFC activity, $r = .59$, $p < .05$. No other correlations were significant.

4. Discussion

The current investigation highlights functional differences in neural systems supporting emotion regulation in individuals with high and normal levels of trait anxiety. During efforts to reduce emotions, anxious participants showed greater activity in brain regions integral to effortful emotional control (e.g., DLPFC, VLPFC; Ochsner and Gross, 2005; Philips et al., 2008) and automatic regulation of emotional processing (e.g., sgACC; Pezawas et al., 2005).

These results are consistent with our hypothesis that anxiety-prone individuals engage PFC systems more when down-regulating emotions. Subjective indices (e.g., distress ratings during Reduce trials and percent reduction in distress) indicated that anxious subjects were as successful as controls in reducing negative emotion using reappraisal. However, BOLD responses indicated that they required increased PFC engagement to accomplish down-regulation.

Our hypothesis that anxious subjects would show greater amygdala activation even after employing reappraisal was not supported. This hypothesis was based on our *a priori* assumption that anxious subjects would be less successful in employing reappraisal to decrease emotions. However, the self-report ratings indicated that anxious subjects were at least as successful as normal controls in reducing distress using reappraisal. Based on that finding, between-group differences in amygdala activation on the Reduce-Maintain contrast would not in fact be expected. It is possible that subjects high in trait anxiety would have more difficulty employing reappraisal (and demonstrate amygdale hyperactivity) on an emotion regulation task involving more intense task stimuli (e.g., extremely negative images); future research should explore this question.

4.1 Neural Activation During the Emotion Regulation Task

Across all subjects, we observed increased activation of right DLPFC during down-regulation of negative emotions. Right-lateralization of this effect may reflect greater engagement of the right hemisphere on tasks that involve visual processing, negative stimuli, and/or motivated processing (Craig, 2005).

DLPFC consistently emerges as a region that is more active during reappraisal than comparison conditions (Levesque et al., 2003; Ochsner et al., 2002, 2004; Phan et al., 2005; Goldin et al., 2008), with some investigators suggesting that right DLPFC specifically mediates essential components of cognitive reappraisal (Kalisch et al., 2005). DLPFC is

strongly implicated in higher-order executive functions (Aron, 2007; Miller and Cohen, 2001). In the context of reappraisal DLPFC may be essential for attentional control (e.g., holding appropriate interpretations "on-line") and inhibition of behavioral responses that might disrupt down-regulation (Philips et al., 2008).

4.2 Between-Group Differences During Down-Regulation of Emotion

Compared to controls, anxious participants displayed greater lateral (left DLPFC, bilateral VLPFC) and medial (dorsomedial PFC, sgACC) PFC activity during down-regulation of emotion. DLPFC, VLPFC, and dorsomedial PFC support cognitive reappraisal of emotional stimuli in healthy individuals (Ochsner et al., 2002, 2004; Phan et al., 2005; Koenigsberg et al., 2010) and have been conceptualized as critical elements of a top-down emotional control network (Ochsner and Gross, 2005; Philips et al., 2008). Hyperactivity of left DLPFC, bilateral VLPFC, and dorsomedial PFC in anxious subjects suggests that they required greater engagement of this network to accomplish down-regulation.

Anxious participants also showed greater sgACC activity, a structure that plays an important role in modulating amygdala processing of emotional salience. The sgACC features prominently in neural models of depression, with evidence of both structural and functional sgACC abnormalities in depressed samples (Drevets et al., 2008). Anxiety-prone individuals display reduced connectivity of the sgACC and amygdala, and decreased ability of the sgACC to modulate amygdala may create compensatory hyperactivity in other PFC regions (Pezawas et al., 2005). The PFC hyperactivity displayed by anxious subjects may have been partly a compensatory response triggered by the reduced ability of the sgACC to regulate amygdala response to task stimuli.

Activity in left VLPFC was positively correlated with percent reduction in distress in the anxious group. While strong inferences should not be drawn from such correlations (and we caution that this result was based on observation of only 13 subjects), it is possible that anxious subjects engaged this region in a compensatory manner that did in fact result in greater success in reducing negative emotions using reappraisal. One prior study also suggested that increased VLPFC activation served as a compensatory response for anxious adolescents (Monk et al., 2006).

Overall, the present results converge with evidence from studies of subjects with MDD. Depressed individuals in one study engaged dACC more than controls during down-regulation of emotions elicited by sad films (Beauregard et al., 2006). In another study, depressed individuals recruited right VLPFC more than controls during down-regulation (Johnstone et al., 2007). It is possible that PFC hyperactivity during down-regulation of negative emotion may be a shared feature of trait anxiety and depression.

The present findings only partly converge with results of investigations of patients with anxiety disorders. In one study, patients with SAD recruited DLPFC and dACC to a *lesser* extent than controls during regulation of emotions elicited by socially threatening stimuli (Goldin et al., 2009a), and in another study patients with PTSD engaged lateral PFC less than controls when regulating emotions elicited by negative pictures (New et al., 2009). These results contrast with our finding of PFC hyperactivity in anxious participants.

One factor that may have contributed to this discrepancy is variability in task stimuli. Disorder-specific stimuli (used by Goldin et al., 2009a) should be particularly emotionally provocative. Limbic response to disorder-specific stimuli may overwhelm the subject such that conscious efforts to reduce emotions (e.g., via lateral PFC) may be more difficult to generate. In contrast, subjects confronting moderately negative stimuli that are not disorder-specific (as in this study) may be less overwhelmed, but at the same time may need to "work

harder" and engage PFC systems more than controls to accomplish down-regulation. The same study that showed less PFC activity in patients with SAD who were regulating negative reactions to socially threatening stimuli (Goldin et al., 2009a) also showed that they engaged right DLPFC *more* than controls during regulation of emotions related to physically threatening stimuli. This constitutes one parallel to the present results (i.e., provides evidence of PFC hyperactivity in anxious subjects when they regulate emotions in response to stimuli that are not directly related to their disorder).

Another possibility relates to the length of the regulation period analyzed in this study (jittered to average 5 seconds). A recent study that examined reappraisal of negative self-beliefs (Goldin et al., 2009b) showed that patients with SAD displayed less PFC activity than controls early in the regulation process (0–3 seconds post-stimulus), but greater PFC activity later in the process (6–9 seconds post-stimulus). This points to increased PFC engagement by anxious subjects, albeit only during the later stages of the regulation process. It is possible that the PFC hyperactivity observed in our anxious subjects may have been more prominent toward the end of the 5-second regulation period; however, we have insufficient power to conduct this fine-grained analysis in the current study.

4.3 Between-Group Differences During Maintenance of Emotion

Between-group effects were partly attributable to the fact that some regions that were hyperactive in anxious participants during Reduce trials were engaged more by non-anxious subjects during Maintain trials (see bar graphs in Figure 4). Activation patterns during Reduce and Maintain trials are consistent with the idea that PFC systems are more or less engaged during emotion regulation, depending on how far individuals are from their homeostatic "set points" of emotional response. Anxious individuals who are already aroused at baseline and automatically appraise stimuli in a threat-laden manner (Bar-Haim et al., 2007) may require robust engagement of these systems to attenuate the emotional impact of moderately negative stimuli. In contrast, non-anxious individuals who are relaxed at baseline and interpret stimuli in a non-threatening manner may require engagement of the same systems to *maintain* emotions to moderately negative stimuli.

4.4 Limitations

Though comparable to the sample sizes of several other investigations of emotion regulation that have used fMRI, the sample size here is somewhat modest and therefore replication is warranted.

Although recruited as a "trait anxious" sample, most HA subjects in this study met criteria for anxiety disorders (which were essentially limited to GAD and SAD). The present findings may pertain to these specific conditions rather than the full anxiety disorder spectrum. Conversely, the heterogeneity of focus of subjects' anxiety may be viewed as a limitation. Although all HA subjects scored high on trait anxiety, some had predominantly social concerns (SAD) while others endorsed more diffuse worry (GAD). Though we conclude here that abnormal neural activation was associated with high trait anxiety, it is possible that differences in social anxiety or worry could explain the observed between-groups differences. Additional research with more homogenous samples of anxious individuals may be able to clarify this. The sample also consisted predominantly of young, female undergraduates; therefore, results may not generalize to samples more diverse in gender, age, or education.

This study relied on participants to carry out Reduce and Maintain instructions, which are somewhat open to interpretation. A rigorous pre-experiment training procedure was used, that aimed to balance standardization of participant behavior and allowance of individual

differences which were integral to the topic of study. It is also possible that demand characteristics influenced subjects' distress ratings, in that they may have felt obligated to report lower distress on Reduce trials and higher distress on Maintain trials. We attempted to minimize this by emphasizing during the training session that we wanted subjects to report what they were really feeling, rather than what they thought they should feel based on the Reduce and Maintain instructions.

Finally, the Maintain instructions may have prompted up-regulation, in that some participants may have needed to deliberately extend their emotions for the entire trial (cf. section 4.3 "Between-Group Differences in Maintenance of Emotion"). The effects reported here should be interpreted in light of this (i.e., anxious participants recruit PFC more than controls during reappraisal *relative to* maintenance of emotions).

4.5 Conclusions

Down-regulation of negative emotions via cognitive reappraisal in anxious participants was associated with greater engagement of prefrontal systems including left DLPFC, bilateral VLPFC, dorsomedial PFC, and sgACC. These results provide evidence of altered function of neural systems supporting down-regulation of negative emotion in anxiety-prone individuals. Future research should attempt to confirm these results in other samples of anxious individuals and to clarify whether dysfunction of neural systems supporting emotion regulation constitutes a marker of vulnerability to emotional disorders.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

Aron AR. The neural basis of inhibition in cognitive control. Neuroscientist 2007;13:214–228. [PubMed: 17519365]

Bar-Haim Y, Lamy D, Pergamin L, Bakermans-Kranenburg MJ, van Ijzendoorn MH. Threat-related attentional bias in anxious and nonanxious individuals: a meta-analytic study. Psychol Bull 2007;133:1–24. [PubMed: 17201568]

Beauregard M, Paquette V, Levesque J. Dysfunction in the neural circuitry of emotional self-regulation in major depressive disorder. NeuroReport 2006;17:843–846. [PubMed: 16708026]

Campbell-Sills, L.; Barlow, DH. Incorporating emotion regulation into conceptualizations and treatments of anxiety and mood disorders. In: Gross, JJ., editor. Handbook of Emotion Regulation. New York, NY: Guilford Press; 2007. p. 542-559.

Cox RW. AFNI: Software for analysis and visualization of functional magnetic resonance neuroimages. Comput Biomed Res 1996;29:162–173. [PubMed: 8812068]

Craig AD. Forebrain emotional asymmetry: a neuroanatomical basis? Trends Cogn Sci 2005;9:566–571. [PubMed: 16275155]

Drevets WC, Savitz J, Trimble M. The subgenual anterior cingulate cortex in mood disorders. CNS Spectr 2008;13:663–681. [PubMed: 18704022]

First, MB.; Spitzer, RL.; Gibbon, M.; Williams, JBW. Structured Clinical Interview for DSM-IV Axis I Disorders, Clinician Version. Washington, D.C.: American Psychiatric Press, Inc.; 1996.

Goldin PR, Manber T, Hakimi S, Canli T, Gross JJ. Neural bases of social anxiety disorder: emotional reactivity and cognitive regulation during social and physical threat. Arch Gen Psychiatry 2009a; 66:170–180. [PubMed: 19188539]

Goldin PR, Manber-Ball T, Werner K, Heimberg R, Gross JJ. Neural mechanisms of cognitive reappraisal of negative self-beliefs in social anxiety disorder. Biol Psychiatry 2009b;66:1091–1099. [PubMed: 19717138]

Goldin PR, McRae K, Ramel W, Gross JJ. The neural bases of emotion regulation: reappraisal and suppression of negative emotion. Biol Psychiatry 2008;63:577–586. [PubMed: 17888411]

Gross JJ. Antecedent- and response-focused emotion regulation: Divergent consequences for experience, expression, and physiology. J Pers Soc Psychol 1998;74:224–237. [PubMed: 9457784]

Gross JJ, John OP. Individual differences in two emotion regulation processes: Implications for Affect, relationships, and well-being. J Pers Soc Psychol 2003;85:348–362. [PubMed: 12916575]

Gross, JJ.; Thompson, RA. Emotion regulation: Conceptual foundations. In: Gross, JJ., editor. Handbook of Emotion Regulation. New York, NY: Guilford Press; 2007. p. 3-24.

Johnstone T, van Reekum CM, Urry HL, Kalin NH, Davidson RJ. Failure to regulate: counterproductive recruitment of top-down prefrontal-subcortical circuitry in major depression. J Neurosci 2007;27:8877–8884. [PubMed: 17699669]

Kalisch R, Wiech K, Critchley HD, Seymour B, O'Doherty JP, Oakley DA, et al. Anxiety reduction through detachment: subjective, physiological, and neural effects. J Cogn Neurosci 2005;17:874–883. [PubMed: 15969906]

Koenigsberg HW, Fan J, Ochsner KN, Liu X, Guise KG, Pizzarello S, et al. Neural correlates of the use of psychological distancing to regulate responses to negative social cues: A study of patients with borderline personality disorder. Biol Psychiatry 2009;66:854–863. [PubMed: 19651401]

Koenigsberg HW, Fan J, Ochsner KN, Liu X, Guise KG, Pizzarello S, et al. Neural correlates of using distancing to regulate emotional responses to social situations. Neuropsychologia 2010;48:1813–1822. [PubMed: 20226799]

Lang, PJ.; Bradley, MM.; Cuthbert, BN. International Affective Picture System (IAPS): Technical Manual and Affective Ratings. Gainesville, FL: NIMH Center for the Study of Emotion and Attention, University of Florida; 1997.

Levesque J, Eugene F, Joanette Y, Paquette V, Mensour B, Beaudoin G, et al. Neural circuitry underlying voluntary suppression of sadness. Biol Psychiatry 2003;53:502–510. [PubMed: 12644355]

Mennin DS, Heimberg RG, Turk CL, Fresco DM. Preliminary evidence for an emotion dysregulation model of generalized anxiety disorder. Behav Res Ther 2005;43:1281–1310. [PubMed: 16086981]

Miller EK, Cohen JD. An integrative theory of prefrontal cortex function. Ann Rev Neurosci 2001;24:167–202. [PubMed: 11283309]

Monk CS, Nelson EE, McClure EB, Mogg K, Bradely BP, Leibenluft E, et al. Ventrolateral prefrontal cortex activation and attentional bias in response to angry faces in adolescents with generalized anxiety disorder. Am J Psychiatry 2006;163:1091–1097. [PubMed: 16741211]

New AS, Fan J, Murrough JW, Liu X, Liebman RE, Guise KG, et al. A functional magnetic resonance imaging study of deliberate emotion regulation in resilience and posttraumatic stress disorder. Biol Psychiatry 2009;66:656–664. [PubMed: 19589502]

Ochsner KN, Bunge SA, Gross JJ, Gabrieli JDE. Rethinking feelings: An fMRI study of the cognitive regulation of emotion. J Cogn Neurosci 2002;14:1215–1229. [PubMed: 12495527]

Ochsner KN, Gross JJ. The cognitive control of emotion. Trends in Cogn Sci 2005;9:242–249. [PubMed: 15866151]

Ochsner KN, Ray RD, Cooper JC, Robertson ER, Chopra S, Gabrieli JD, Gross JJ. For better or for worse: neural systems supporting the cognitive down- and up-regulation of negative emotion. Neuroimage 2004;23:483–499. [PubMed: 15488398]

Pezawas L, Meyer-Lindenberg AM, Drabant EM, Verchinski BA, Munoz KE, Kolachana BS, et al. 5-HTTLPR polymorphism impacts human cingulate-amygdala interactions: A genetic susceptibility mechanism for depression. Nat Neurosci 2005;8:828–834. [PubMed: 15880108]

Phan KL, Fitzgerald DA, Nathan PJ, Moore GJ, Uhde TW, Tancer ME. Neural substrates for voluntary suppression of negative affect: a functional magnetic resonance imaging study. Biol Psychiatry 2005;57:210–219. [PubMed: 15691521]

Philips ML, Ladouceur CD, Drevets WC. A neural model of voluntary and automatic emotion regulation: implications for understanding the pathophysiology and neurodevelopment of bipolar disorder. Mol Psychiatry 2008;13:833–857.

Spielberger, CD.; Gorsuch, RL.; Lushene, R.; Vagg, PR.; Jacobs, GA. Manual for the State-Trait Anxiety Inventory (Form Y). Palo Alto, CA: Mind Garden; 1983.

Talairach, JTP. Co-planar Stereotaxic Atlas of the Human Brain: 3-Dimensional Proportional System: An Approach to Cerebral Imaging. New York, NY: Thieme Medical Publishers, Inc.; 1998.

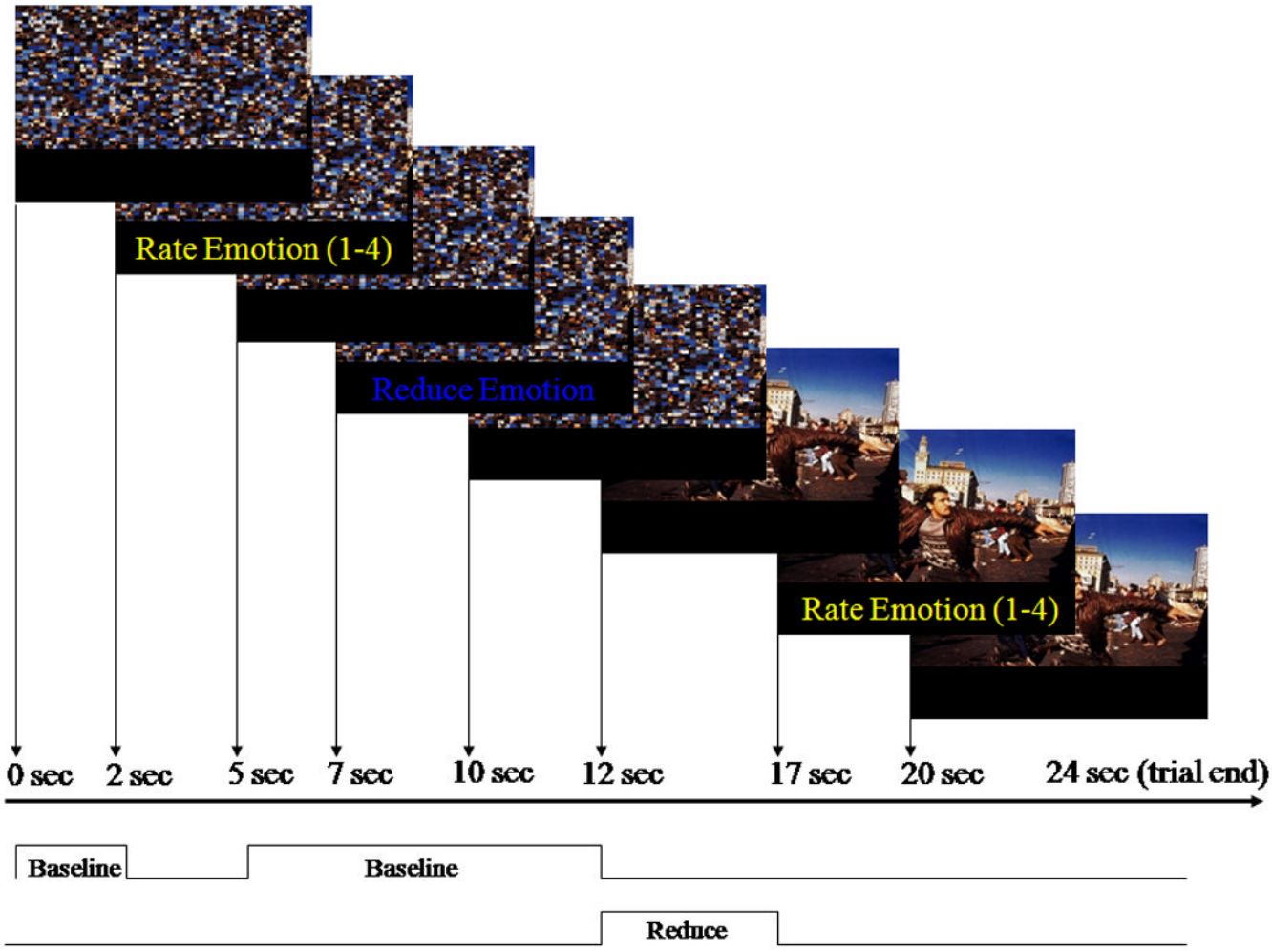

Figure 1.

Depiction of a Reduce Trial and Corresponding Regressors of Interest. The figure shows what subjects saw and responded to over the course of the 24-second trial. Line graphs below the timeline show how Reduce and Baseline regressors were defined. Maintain trials were identical except that subjects were provided with the instruction “Keep Up Emotion” 6–8 seconds into the trial. The Maintain regressor was defined analogously to the Reduce regressor (i.e., it was time-locked to the 4–6 seconds when subjects were viewing the image but had not yet been prompted to “Rate Emotion”).

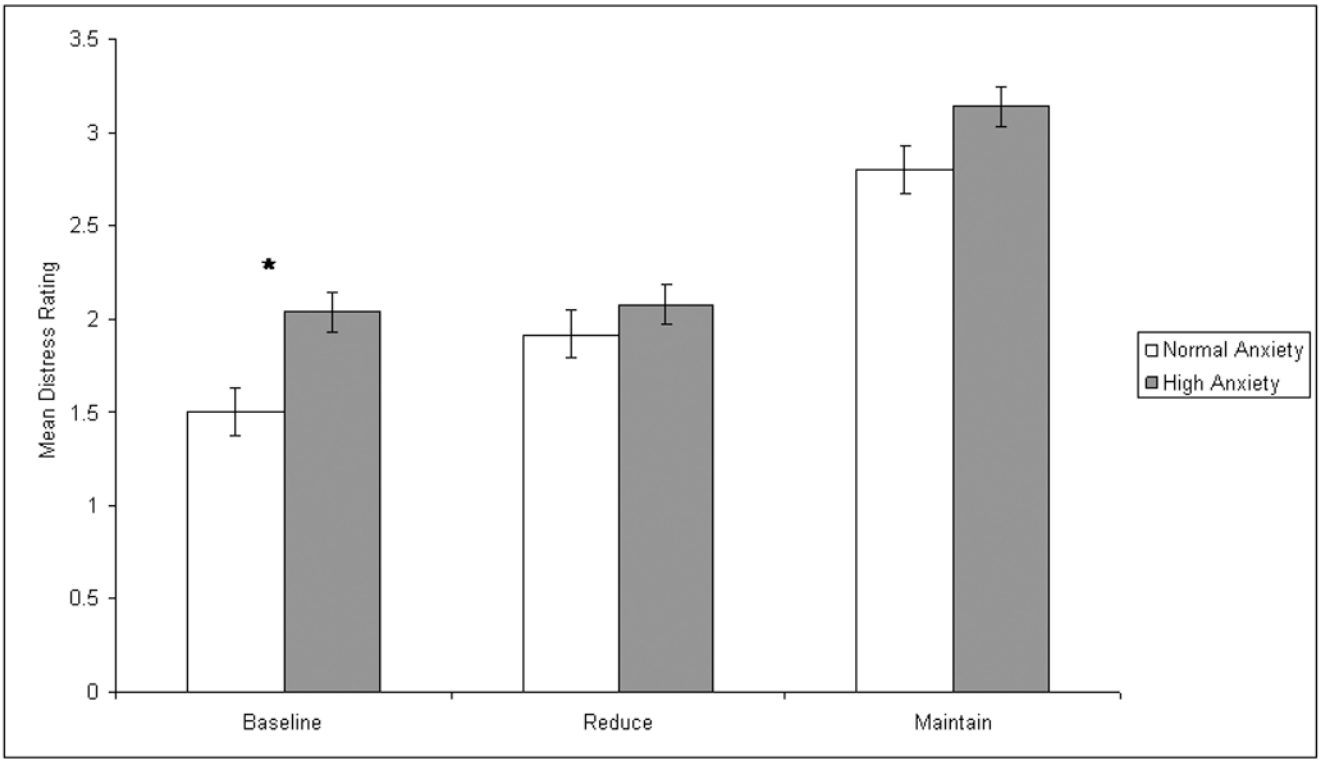

Figure 2.
Subjective Distress During Baseline, Reduce, and Maintain periods for Anxious and Non-Anxious Participants. The response scale for subjective distress was 1–4 (1 = none; 2 = mild; 3 = moderate; 4 = severe). * $p < .0125$ (Bonferroni correction).

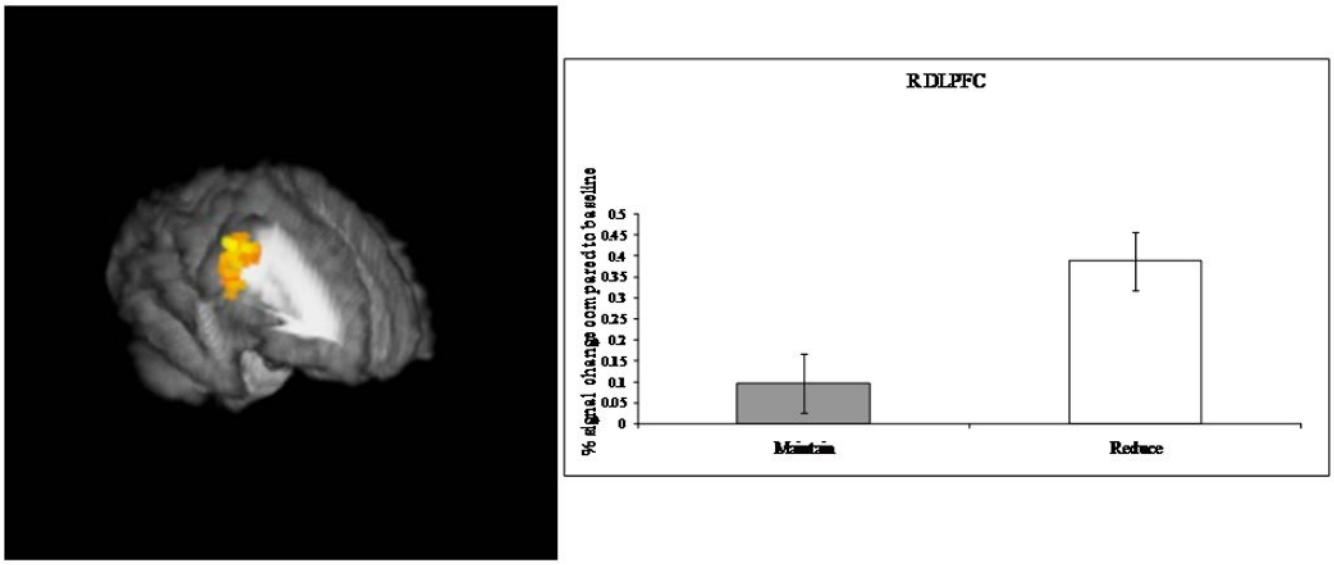

Figure 3.
Effects of the emotion regulation task (Reduce-Maintain contrast) across all participants (N = 26). The cuts in the image were made at x = 18, y = −10, and z = 10. Periods of reducing emotion using cognitive reappraisal were associated with greater activation of right dorsolateral prefrontal cortex (BA 9), compared to periods of maintaining emotion (effect significant at $p < .01$). Bar graph shows % signal change during Reduce and Maintain trials (each relative to baseline activation); error bars represent the standard errors.

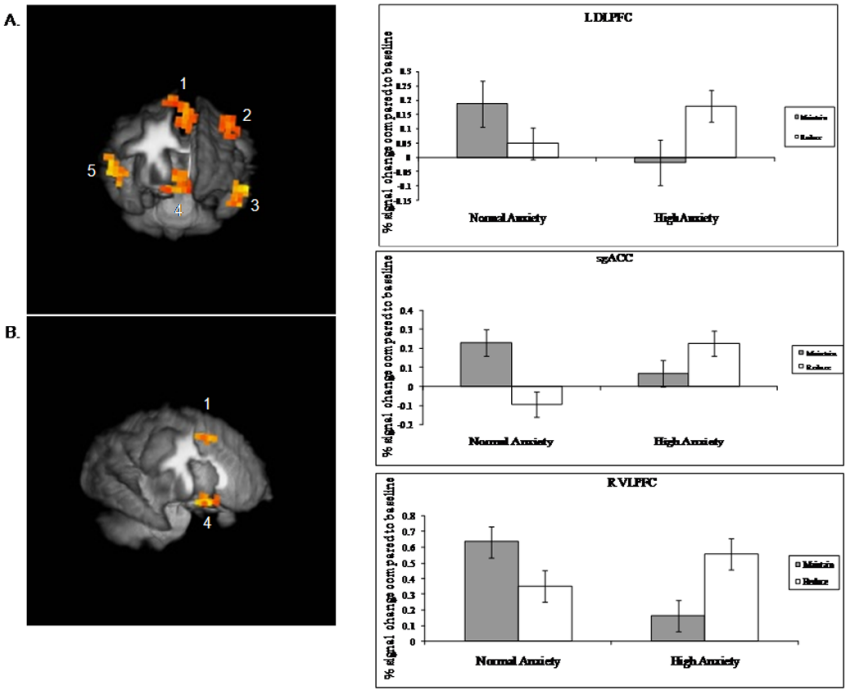

Figure 4.

Between-group effects on the emotion regulation task. The cuts in both images were made at x = −5, y = 10, and z = −10. Activation shown results from comparing the anxious (n = 13) and control (n = 13) groups on the Reduce-Maintain contrast. Warm colors indicate regions that were more active in anxious subjects during periods of reducing emotions. 4A: Anxious subjects showed greater activation in dorsomedial prefrontal cortex/dorsal anterior cingulate cortex (BA 6/32; labeled “1” in figure), left dorsolateral prefrontal cortex (BA 8/9; labeled “2” in figure), left ventrolateral prefrontal cortex (BA 47; labeled “3” in figure), , subgenual anterior cingulate cortex (BA 25; labeled “4” in figure), and right ventrolateral prefrontal cortex (BA 44/45; labeled “5” in figure). 4B: Additional view of medial PFC activation. Bar graphs show % signal change during Reduce and Maintain trials (each relative to baseline activation) for the anxious and control groups (left DLPFC, sgACC, and right VLPFC are presented as examples). Error bars indicate standard errors.

Table 1

Significant Clusters From the Reduce-Maintain Contrast (All Participants; N = 26)

			Talairach coordinates of cluster center		
Brain region	***t* score**	**Cluster size (mm^3)**	**X**	**Y**	**Z**
Reduce > Maintain					
Right inferior frontal gyrus (BA 9)	3.73	2368	44	9	33
Maintain > Reduce					
No significant clusters					

Note. The *t* statistic is derived from averaging the BOLD response for voxels in the identified cluster for each subject, and then comparing mean BOLD response during Reduce and Maintain periods. Effect is significant at $p < .01$. BA = Brodmann area.

Table 2

Significant Clusters From the Task (Reduce-Maintain) by Group (High Anxiety-Normal Anxiety) Contrast

			Talairach coordinates of cluster center		
Brain region	***t* score**	**Cluster size (mm^3)**	**X**	**Y**	**Z**
High Anxiety > Normal Anxiety					
R/L Cuneus (BA 18)	4.68	4032	4	−75	6
R/L Medial Frontal Gyrus (BA 6/32)	4.19	3136	3	15	47
L Middle Frontal Gyrus (BA 8/9)	4.00	2368	−31	14	41
R/L Anterior Cingulate (BA 25)	5.15	2240	1	14	−3
L Middle Frontal Gyrus (BA 47)	6.06	1728	−41	42	−6
R Inferior Prefrontal Gyrus (BA 44/45)	4.74	1472	50	12	3
Normal Anxiety > High Anxiety					
No significant clusters					

Note. The clusters listed in the table were identified as described in sections 2.5 and 2.6. The follow up *t* tests were conducted by averaging the BOLD response for voxels in the identified cluster for each subject, and then comparing mean Reduce-Maintain BOLD response for the HA and NA groups. Effects are significant at $p < .01$. BA = Brodmann area; R = right; L = left; R/L = bilateral.