Dissociating Bottom-Up and Top-Down Mechanisms in the Cortico-Limbic System during Emotion Processing

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The cortico-limbic system is critically involved in emotional responses and resulting adaptive behaviors. Within this circuit, complementary regions are believed to be involved in either the appraisal or the regulation of affective state. However, the respective contribution of these bottom-up and top-down mechanisms during emotion processing remains to be clarified. We used a new functional magnetic resonance imaging (fMRI) paradigm varying 3 parameters: emotional valence, emotional congruency, and allocation of attention, to distinguish the functional variation in activity and connectivity between amygdala, anterior cingulate cortex (ACC), and dorsolateral prefrontal cortex (DLPFC). Bottom-up appraisal of negative compared with positive stimuli led to a greater amygdala response and stronger functional interaction between amygdala and both dorsal ACC and DLPFC. Top-down resolution of emotional conflict was associated with increased activity within ACC and higher functional connectivity between this structure, and both the amygdala and DLPFC. Finally, increased top-down attentional control caused greater engagement of the DLPFC, accompanied by increased connectivity between DLPFC and dorsal ACC. This novel task provides an efficient tool for exploring bottom-up and top-down processes underlying emotion and may be particularly helpful for investigating the neurofunctional underpinnings of psychiatric disorders.

Keywords: cortico-limbic system, emotion, fMRI, psychophysiological interaction

Introduction

Past research on emotion has led to models in which affective cues are processed through 2 complementary brain systems: a ventral system involved in rapid appraisal of emotional stimuli and generation of affective responses, and a dorsal system responsible for the regulation of the emotional states (Phillips et al. 2003, 2008; Ochsner and Gross 2005; Phan et al. 2005; Goldin et al. 2008). These models define the cortico-limbic circuit, which supports humans' vital adaptive functioning in their environment. When studying this circuit, psychological researchers have been particularly interested in the interaction between the ventral part of the circuit, mainly the amygdala, and the dorsal system, which includes a variety of prefrontal regions. Studies support the view that the amygdala, through bottom-up mechanisms, facilitates perceptual processing by directing or biasing attention (Ledoux 2000; Davis and Whalen 2001; Phelps 2006). Concurrently, a growing literature indicates that cognitive processes such as distraction or reappraisal can regulate emotional responses through top-down mechanisms (Ochsner and Gross 2005; McRae et al. 2010; Kanske et al. 2011; Ochsner et al. 2012). The manner by which these 2 systems interact has become increasingly central to models of psychopathology. Indeed, most psychiatric disorders are believed to be defined by some level of dysfunction in this circuit, such as defective modulation of the dorsal part of the system or enhanced engagement of the ventral part (Mohanty et al. 2005; Bishop et al. 2007; Johnstone et al. 2007; Fales et al. 2008; Almeida et al. 2009).

Theoretical frameworks of emotional processing emphasize the reciprocal relationship between the dorsolateral prefrontal cortex (DLPFC), a major component of the high-order associative cortex engaged in attentional and complex cognitive operations (MacDonald 2000; Miller and Cohen 2001; Blasi et al. 2007), and the amygdala, an extensively studied structure that plays a central role in automatic affective processing (Ledoux 2000; Anderson et al. 2003; Ohman 2005; Adolphs 2008). However, by probing only “bottom-up” or “top-down” mechanisms, it is difficult to distinguish the respective contribution of appraisal and control systems during emotion processing. Furthermore, there are few direct projections between these regions (Barbas 2000; Ray and Zald 2012). Thus, to substantiate any influence of DLPFC on amygdala (and vice versa), alternative models require intermediate regions should be incorporated.

Anatomical studies have repeatedly highlighted the strategic position and function of the anterior cingulate cortex (ACC). This medial frontal region is extensively connected to both the lateral prefrontal cortex (including the dorsolateral region) and the amygdala (Bush et al. 2000; Ongür and Price 2000; Paus et al. 2001). Convergent evidence from cytoarchitectural, lesion, connectivity and functional neuroimaging studies supports that ACC is functionally segregated into 2 subregions: a dorsal evaluative or cognitive-processing region and a ventral emotion-related region (Vogt et al. 1992; Devinsky et al. 1995; Bush et al. 2000; Steele and Lawrie 2004). The dorsal part of the ACC is thought to exercise cognitive control over emotional processes (Phillips et al. 2003) and to be involved in the explicit judgment of emotional stimuli (Kalisch 2009). Thus, dorsal ACC has been linked to conscious emotional evaluation (i.e., explicit, sophisticated context-dependent analysis) particularly for conscious appraisal of fear-related information (Mechias
et al. 2010; Etkin et al. 2011). In contrast, the ventral part of the ACC has been described as important for the induction of emotional response (Phan et al. 2002), such as the generation of sad mood (Mayberg et al. 1999) and depression (Mayberg et al. 2005). Previous work has highlighted the key role played by the ventral ACC in automatic emotion regulation (i.e., unconscious, implicit, unintended process) inducing change in any aspect of an individual’s emotional response (Phelps et al. 2004; Delgado et al. 2008; Phillips et al. 2008), as well as its dysfunction in depression (Mayberg et al. 1997, 1999). The functional heterogeneity of the ACC is echoed in the pattern of functional and structural connectivity. The dorsal aspect of ACC exhibits strong interactions with regions typically implicated in higher order cognitive functions (e.g., attentional processes and working memory), such as dorsolateral and posterior inferior prefrontal cortices, whereas the ventral aspect shows preferential connectivity with limbic and paralimbic regions (e.g., amygdala, hippocampus, and ventromedial prefrontal cortex) implicated in affective processing (Amaral et al. 1992; van Hoesen et al. 1993; Ghashghaei et al. 2007; Margulies et al. 2007; Beckmann et al. 2009). The dorsal and ventral divisions of the anterior cingulate are also interconnected (Musil and Olson 1988a, 1988b; van Hoesen et al. 1993); a negative functional relationship has been evidenced between these subregions at rest (Margulies et al. 2007).

Thus, a particularly significant neural loop includes the amygdala and its projection to the ventral ACC, which itself interacts with a more dorsal part of the ACC that in turn allows downregulation of the amygdala (Pezawas et al. 2005). Next to this first basic emotion circuit, a second track would engage the ACC in attentional or cognitive control through its large projections to the DLPFC (Bush et al. 2000; Botvinick et al. 2004; Kerns et al. 2004; Ridderinkhof et al. 2004). For instance, higher attentional load engages the DLPFC (Blasi et al. 2007), whereas executive control in the case of conflict resolution activates the ACC and increases the interplay between the ACC and the DLPFC (Van Veen et al. 2001; Kerns et al. 2004; Carter and Van Veen 2007; Fan et al. 2008). Interestingly, this cognitive circuit, composed of the ACC–DLPFC interaction, is also recruited in emotion regulation when mechanisms of a more cognitive nature are employed (Ochsner and Gross 2005; Banks et al. 2007; Phillips et al. 2008; Kalisch et al. 2011). Delgado et al. demonstrated that when elaborate emotion regulation strategies are used, the dorsal ACC–DLPFC “cognitive” circuit is engaged, and it mediates its regulatory action through recruitment of the ventral ACC–amygdala circuit involved in automatic control of emotion (Delgado et al. 2008).

Here, we conceived a task, the variable attention and congruency task (VAT), with the intention of dissociating the various components of this cortico-limbic circuit, that is, the dorsal cognitive circuit (ACC–DLPFC) intertwined with the ventral automatic circuit (ACC–amygdala). For this, the task varied according to 3 parameters that differentially indexed appraisal and regulation processes: emotional valence (positive or negative), emotional congruency (same or opposite emotional content in the stimuli), and allocation of attention (low or high attentional load).

We predicted that negative relative to positive valence conditions would result in faster reaction time and better accuracy (Ohman et al. 2001; Vuilleumier 2005; Pessoa 2009). Conversely, we expected to find lower behavioral performance in the incongruent versus congruent conditions (Etkin et al. 2006; Egner et al. 2008; Kanske and Kotz 2011) and in the high-attentional load versus low-attentional load conditions (Blasi et al. 2007). From the neuroimaging perspective, based on the literature cited earlier, we hypothesized that the variation of each parameter would differentially engage the amygdala, ACC, and DLPFC. Specifically, we expected that bottom-up appraisal of negative compared with positive stimuli would cause higher engagement of the amygdala, in keeping with its role in negative/ fear-related stimuli processing (LeDoux 2000) and that top-down resolution of emotional conflict would lead to increased activity within ACC, given the well-established implication of this region in conflict monitoring (Bush et al. 2000; Etkin et al. 2006). Finally, in agreement with the engagement of DLPFC in the allocation of attentional resources (Blasi et al. 2007), we anticipated that increased top-down attentional demands would be associated with stronger activation of DLPFC. Furthermore, we conducted psychophysiological interaction (PPI) analyses to better understand the functional connectivity between these cerebral regions. We speculated that the effects of the emotional valence, congruency, and attentional demands specifically alter the functional interactions between the amygdala, ACC, and DLPFC.

Materials and Methods

Participants

Thirty-three healthy volunteers (11 females, 20–47 years old, mean age 32.2 years, standard deviation [SD] = 7.3) completed the study. Participants were recruited through advertising in the local community of Marseille. Average years of education completed were 12.9 (SD = 1.9). IQ estimates were obtained for each participant based on the Wechsler Adult Intelligence Scale (WAIS III) (Wechsler 1997). The average IQ was 114.8 (SD = 16.4). The sample was 61% Caucasian, 27% North African, and 12% African. Before entering the study, subjects underwent a medical interview and examination. The non-patient version of the Structured Clinical Interview for DSM-IV (SCID; First et al. 2002) was used to ensure the absence of psychiatric disorder or psychiatric history. Clinicians were trained to use the SCID in accordance with the SCID User’ Guide (SCID; First et al. 2002) before the study commenced. Participants had no current or past serious medical or neurological condition; they were not taking any psychotropic drugs at the time of the study and had no contraindication for MRI.

All participants were right-handed according to the Edinburgh Handedness Inventory (Oldfield 1971) and had normal or corrected-to-normal vision. This study was conducted in accordance with the principles of the declaration of Helsinki. Approval was obtained from the local ethics committee (Comité consultatif de protection des personnes dans la recherche biomédicale, Marseille). Each participant was registered with the French national file and gave informed written consent before entering the study. Subjects received financial compensation for their participation. Data from 7 participants were removed because of excessive head motion, anomalies detected on the anatomical scans, or visible artifacts in the functional images (i.e., truncated limbic lobes on 1 subject’s functional data). Thus, data from 26 participants (9 females; 32.7 ± 7.7 years old; average years of education = 12.8 ± 1.8) were included in the final analysis.

Stimuli and Procedure

The experimental task presented images composed of 2 parts (Fig. 1). The central part of the image displayed photographs of faces, drawn from NimStim Face stimulus set (Tottenham et al. 2009), expressing positive emotion (joy) or negative emotion (fear, disgust, or anger). The peripheral part, on which the face images were superimposed, represented scenes extracted from IAPS files (Lang et al. 2008) with a pleasant or unpleasant emotional content. Subjects had to focus on the part...
of the image framed in green (either the face or the scene) and determine its emotional content (pleasant vs. unpleasant) by pressing the corresponding key on a response keypad placed in their right hand.

The task (VAAT) consisted of 3 × 2 conditions varying according to emotional valence (positive or negative), emotional congruency (same or different emotional content in the face and the scene), and attentional load (attention focused on the face [low attention] or on the scene [high attention]).

The VAAT had a mixed event-related/block design, comprising 4 sessions of 6 min 8 s each. The sessions were divided in 16 blocks that each lasted 20.4 s. The blocks began by an instruction panel (displayed during 1400 ms) specifying upon which part of the image the subject had to focus during the block, followed by 4 experimental trials, each lasting 3000 ms, during which time subjects provided their response. The valence parameter varied from trial to trial whereas the emotional congruency and attention parameters varied from block to block. The task (VAAT) consisted of 3 × 2 conditions varying according to (A) the emotional valence (positive [1] or negative [2]), (B) the emotional congruency (same [1] or different [2]) emotional content in the face and the scene, and (C) the allocation of attention (attention focused on the face [i.e., low demanding] [1] or on the scene [i.e., high demanding] [2]).

Each stimulus could therefore be classified along these 3 dimensions; picture C2 for instance is a negative valence, emotionally incongruent, high attention item. Subjects were asked to selectively attend to the part of the image framed in green (either the face or the scene) and to determine its emotional content (pleasant vs. unpleasant).

Figure 1. Experimental paradigm. The task presented images composed of 2 parts, a central part composed of a face and a peripheral part (on which the face images were superposed) representing scenes. Both parts of the picture had an emotional content (positive or negative) that could be similar or conflicting with that of the other part. The paradigm consisted of 3 × 2 conditions varying according to (A) the emotional valence (positive [1] or negative [2]), (B) the emotional congruency (same [1] or different [2]) emotional content in the face and the scene), and (C) the allocation of attention (attention focused on the face [i.e., low demanding] [1] or on the scene [i.e., high demanding] [2]).

Behavioral Data Analysis
Behavioral data consisted of reaction times and accuracy rate. For the analysis of reaction time, only correct trials for which subjects answered within the 3-s window were used. For the analysis of accuracy scores, only trials for which subjects answered within the 3-s window were considered. Three one-way within-subjects ANOVAs were conducted separately to evaluate the main effect of emotional valence, emotional congruency, and attentional load on subjects’ performance (reaction time and accuracy). The threshold for statistical significance was fixed at $P < 0.05$. Behavioral data were analyzed in SPSS (v18.0).

fMRI Data Analysis
Pre-Processing
Prior to analysis, the quality of the functional images was assessed using tsdifana (http://imaging.mrc-cbu.cam.ac.uk/imaging/DataDiagnostics). Functional images were subjected to spike artifact detection. The quantitative quality indicators (signal-to-noise ratio, scaled variance, scaled mean voxel, slice by slice variance…) were examined to ensure the stability of the signal over time and the lack of abrupt variation between successive slices.

All data were analyzed using SPM8 software (Wellcome department of Cognitive Neurobiology, University College London; http://www.fil.ion.ucl.ac.uk/spm/software/spm8). The first 4 volumes of each session, corresponding to signal stabilization, were excluded from the analysis. The remaining scans were corrected for differences in slice acquisition time. To reduce the effect of head motion, whole images were realigned to the mean scan of each session. Realignment plots were examined to ensure the absence of excessive movements during

MRI Acquisition
Data were acquired on a 3-T MEDSPEC 30/80 AVANCE imager (Bruker). After an initial localizing scan, functional data were acquired using a T2*-weighted gradient-echoplanar imaging sequence (TR = 3000 ms; TE = 30 ms; FOV = 19.2 × 19.2; 64 × 64 matrix; flip angle 84.8°; voxel size 3 × 3 × 3 mm³). Four functional runs of 45 interleaved axial slices were acquired along the anterior–posterior commissure plane with a continuous slice thickness of 3 mm. Following the functional magnetic resonance imaging (fMRI) scans, high-resolution anatomical images were acquired for the purpose of anatomical identification with a sagittal T1-weighted MP-RAGE sequence (TR = 9.4 ms; TE = 4.42 ms; TI = 800 ms; 256 × 256 × 180 matrix; flip angle 30°, voxel size 1 × 1 × 1 mm³).
the scan. Data were discarded from further analysis if movements in any axis were superior to 3 mm and/or 2°.

The structural scan was co-registered to the functional images, and all images were transformed into a standardized coordinate system corresponding to the Montreal Neurological Institute (MNI) space. The normalized images were spatially smoothed with an isotropic Gaussian kernel (full width at half maximum of 6 mm). Finally, each preprocessing step was check using the Check Registration function implemented in SPM.

Image Analyses
The pre-processed functional images from each participant were analyzed in an event-related approach using a General Linear Model to estimate blood oxygen level-dependent (BOLD) signal changes due to the experimental conditions. An analytic design matrix was constructed, which modeled the onsets and durations of each condition (positive valence, negative valence, congruent, incongruent, attention to the scene [high], and attention to the face [low]). All factors were convolved with a canonical hemodynamic response function. The 6 movement parameters were included in the analysis as regressors of no interest to model residual effects due to head motion. A 128-s high-pass filter was applied to the data to remove low-frequency noise. For each participant, first-level contrast images were calculated to estimate BOLD signal changes due to variation in emotional valence (negative vs. positive valence conditions), emotional congruency (incongruent vs. congruent conditions), and attentional level (attention to the scene [high] vs. attention to the face [low]). The first-level contrast images were then entered into a second-level one-sample t-test with a random effects statistical model to examine the main effects of the task at the group level. We first performed exploratory whole-brain analyses with a threshold at P < 0.001 (uncorrected for multiple comparisons) and a 10-voxel spatial extent to ensure activation of our a priori regions of interest in response to variations in the task parameters. The advantages of combining a voxel-based threshold with a minimum cluster size have been described elsewhere (Forman et al. 1995).

Then, we used a region of interest approach focusing on areas previously implicated in emotion processing: the amygdala, ACC, and DLPFC. The amygdala and DLPFC ROIs were anatomically defined using the Automated Anatomical Labeling software implemented in the WFU PickAtlas (Maldjian et al. 2003). For the ACC, we defined the boundaries between ventral and dorsal ACC based on the functional mapping of the ACC delineated in the work of Margulies et al. (2007). We created masks of the ventral and dorsal ACC using MarsBar toolbox (http://marsbar.sourceforge.net/). Statistical threshold was at P < 0.001 voxel-wise (uncorrected for multiple comparisons) and a 10-voxel spatial extent (Forman et al. 1995). The reported activations survived correction for multiple comparisons (family-wise error [FWE]) at the voxel level.

Functional Connectivity Analyses
Psychophysiological interaction (PPI) analyses were used to assess context-dependent variations in functional connectivity between our regions of interest. PPI is a regression-based method for functional connectivity that examines changes in the regression slope of activation between a seed region and the rest of the brain under the influence of different experimental contexts (Friston et al. 1997).

Three separate PPI analyses were conducted for each participant, using 3 regions of interest as a seed region to examine the variations in their functional interactions according to either the negative > positive contrast (for the amygdala), incongruent > congruent contrast (for the ACC), and high attention > low attention contrast (for the DLPFC). For each subject, the seed regions were determined using subject-specific local maxima in our regions of interest that were within 15 mm (10 mm for the amygdala) of the group maxima and within the same anatomical mask, as defined by the PickAtlas toolbox (Maldjian et al. 2003). For each participant, the first eigenvariate time series of the BOLD signal, adjusted for the effects of interest, was extracted from a 5-mm (3 mm for the amygdala) sphere around the seed coordinates. A time series was calculated for each participant with the first eigenvariate from the time series of all voxels within the sphere. The PPI regressor was calculated as the product of the time series of the seed region (physiological factor) and the vector coding for the task factor (psychological factor). The individual contrast images were then entered into a second-level analysis using a one-sample t-test to assess regions showing significantly different connectivity to the seed region at the group level. Given the subtle nature of brain activity during emotion processing, and the use of a priori defined regions, this interaction was assessed at a combined statistical threshold of P < 0.005 voxel-wise, consistent with earlier studies on emotional processing (Foland et al. 2008; Monk et al. 2008; Townsend et al. 2013). Clusters larger than or equal to 10 voxels are reported. The threshold applied was more severe than that used in PPI reference articles (Friston et al. 1997; Penny et al. 2003) in order to minimize false-positive risk.

Results
Behavioral Data
The 26 subjects included in the final fMRI analysis performed the task successfully. Participants’ accuracy was high, with a mean value of 93% (SD = 9.8), suggesting that subjects had understood the task’s instructions and had been attentive during the experiment.

One-way within-subjects ANOVAs revealed a significant main effect of valence (F1,855 = 22.7, P < 0.001) and attention (F1,855 = 489, P < 0.001) on reaction time. Post hoc t-test indicated that subjects were significantly slower in responding to negative (M = 137.1; SD = 305) compared with positive stimuli (M = 132.2; SD = 279) (t855 = 4.8, P < 0.001) as well as high-attention (M = 147.8; SD = 319) compared with low-attention stimuli (M = 121.6; SD = 290) (t855 = 22.1, P < 0.001). There was no main effect of congruency (Fig. 2).

Concerning participants’ accuracy, there were significant main effects of valence (F1,855 = 24.9, P < 0.001), congruency (F1,855 = 11, P = 0.001), and attention (F1,855 = 18.4, P < 0.001). Post hoc t-test showed that subjects were less accurate in performing the task in the presence of negative (M = 91.4; SD = 15) compared with positive stimuli (M = 94.5; SD = 12) (t855 = 5, P < 0.001), of emotional conflict (M = 92; SD = 14) compared with emotional congruency (M = 94; SD = 12) (t855 = 3.3, P < 0.001), and in high attention (M = 91.6; SD = 15) compared with low attention condition (M = 94.4; SD = 13) (t855 = 4.3, P < 0.001) (Fig. 3).

fMRI Data
Regional Brain Activation
We first examined BOLD signal changes within the corticocerebrolimbic circuit (amygdala, ACC, and DLPFC) according to the 3 parameters of the task. We assessed the emotional valence effect by contrasting negative valence trials to positive trials. Activity within the right amygdala was significantly greater in response to negative than to positive valence pictures (x, y, z = 24, −2, −18; T = 5.29; P < 0.001). Moreover, negative valence trials were accompanied by higher activation of the bilateral DLPFC (right: x, y, z = 54, 30, 22; T = 7.5; P < 0.001; and left: x, y, z = −50, 20, 12; T = 4.08; P < 0.001) (Table 1 and Fig. 4). To determine the emotional congruency effect, incongruent conditions (scene and superposed face of opposite emotional valence) were contrasted to congruent conditions (scene and superposed face of same valence). Within our a priori regions of interest, emotional conflict (i.e., the incongruent condition) was associated with activation of the ACC (x, y, z = 8, 46, 28; T = 4.98; P < 0.001). Activity also increased in the left amygdala in incongruent versus congruent trials (x, y, z =
−28, −8, −12; T = 4.96; P < 0.001) (Table 1 and Fig. 4B). Finally, to examine the effect of attention, we compared trials in which subjects had to concentrate on the scene to trials in which they had to evaluate the face. As expected, higher attentional demands (focusing on the scene rather than the face) led to higher engagement of the bilateral DLPFC (left: x, y, z = −54, 18, 16; T = 6.77; P < 0.001; and right: x, y, z = 56, 24, 24; T = 5.78; P < 0.001) and, to a lesser extent, the dorsal part of ACC (x, y, z = 8, 48, 18; T = 3.93; P < 0.001) (Table 1 and Fig. 4C).

Results obtained from whole-brain analyses are displayed in supplemental material (Supplementary Table 1). These analyses indicated that negative valence trials compared with positive trials, in addition to recruiting the amygdala and bilateral DLPFC, was also associated with increased activity notably within the inferior and middle temporal gyrus, superior parietal

![Figure 2. Behavioral results. (A) Mean reaction time (and standard error) plotted as a function of congruency, attention, and valence effects. (B) Mean reaction time and SD (in parentheses) for the main conditions of the task.](image)

![Figure 3. Behavioral results. (A) Mean accuracy (and standard error) plotted as a function of congruency, attention, and valence effects. (B) Mean percentage of correct answers and SD (in parentheses) for the main conditions of the task.](image)

Table 1
<table>
<thead>
<tr>
<th>Contrast</th>
<th>Anatomical region</th>
<th>H</th>
<th>BA</th>
<th>MNI coordinates (x, y, z)</th>
<th>Cluster size (in voxels)</th>
<th>T</th>
</tr>
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<tr>
<td>Negative &gt; positive</td>
<td>Amygdala</td>
<td>R</td>
<td>24</td>
<td>−2, −18</td>
<td>26</td>
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<td></td>
<td>DLPFC</td>
<td>R</td>
<td>54, 30, 22</td>
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<td>7.50</td>
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<tr>
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<td>R</td>
<td>32</td>
<td>8, 46, 28</td>
<td>46</td>
<td>4.98</td>
</tr>
<tr>
<td></td>
<td>Amygdala</td>
<td>L</td>
<td>−28, −8, −12</td>
<td>16</td>
<td>4.96</td>
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<tr>
<td>High &gt; low attention</td>
<td>DLPFC</td>
<td>L</td>
<td>−54, 18, 16</td>
<td>546</td>
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<td></td>
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<td>56, 24, 24</td>
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<tr>
<td></td>
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<td>R</td>
<td>32</td>
<td>4, 28, 38</td>
<td>18</td>
<td>4.97</td>
</tr>
</tbody>
</table>

Note: Activation clusters for the respective contrasts. Voxel-level threshold, P < 0.001 (uncorrected); 10 voxel spatial extent. The reported activations survived correction for multiple comparisons (FWE) at the voxel level (P < 0.05). H, hemisphere; BA, Brodmann area; cluster size in voxels; T, T-values.
gyrus, and superior frontal/precentral gyrus. In addition to ACC and left amygdala, emotional conflict caused greater activity principally within the superior frontal gyrus, right thalamus, middle temporal gyrus, and calcarine gyrus. Other regions recruited by higher relative to low attentional demands included fusiform/lingual gyrus, rectus gyrus, opercular/ventrolateral prefrontal gyrus, and medial superior frontal gyrus.

Results from the inverse contrasts (i.e., positive vs. negative, congruent vs. incongruent, low vs. high attentional load) indicated no significant BOLD signal changes within our regions of interest.

**Functional Connectivity: PPI**

PPI analyses revealed that in response to negative valence compared with positive valence stimuli, the right amygdala exhibited stronger functional coupling with both DLPFC (right: \(x, y, z = 44, 36, 16; T = 4.28; P < 0.005\); and left: \(x, y, z = -36, 18, 28; T = 4.17; P < 0.005\)) and the dorsal ACC (\(x, y, z = 8, 26, 36; T = 3.97; P < 0.005\)) but decreased functional coupling with ventral ACC (\(x, y, z = -4, 44, 8; T = 4.02\); and \(x, y, z = -4, 34, 12; T = 3.72\) (Table 2 and Fig. 5i). During incongruent versus congruent trials, dorsal ACC displayed increased functional connectivity with both the right amygdala (\(x, y, z = 30, 0, -22; T = 3.20\);

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**Table 2**

<table>
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<th>Contrast/seed region</th>
<th>Connectivity</th>
<th>Anatomical region</th>
<th>H</th>
<th>BA</th>
<th>MNI coordinates ((x, y, z))</th>
<th>Cluster size</th>
<th>(T)</th>
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<tr>
<td>Negative &gt; positive (amygdala)</td>
<td>Increased</td>
<td>DLPFC</td>
<td>R</td>
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<td>44, 36, 16</td>
<td>38</td>
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<tr>
<td>DLPFC</td>
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<td>-36, 18, 28</td>
<td>21</td>
<td>4.17</td>
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<td>-48, 22, 0</td>
<td>23</td>
<td>3.58</td>
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<tr>
<td>Dorsal ACC</td>
<td>R</td>
<td>32</td>
<td>8, 26, 36</td>
<td>20</td>
<td>3.97</td>
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<tr>
<td>Decreased</td>
<td>Ventral ACC</td>
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<td>-4, 46, -4</td>
<td>21</td>
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<td>Increased</td>
<td>DLPFC</td>
<td>R</td>
<td>46</td>
<td>44, 24, 20</td>
<td>16</td>
<td>3.44</td>
</tr>
<tr>
<td>Amygdala</td>
<td>R</td>
<td>32</td>
<td>0, -22</td>
<td>13</td>
<td>3.20</td>
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<tr>
<td>Decreased</td>
<td>Ventral ACC</td>
<td>L</td>
<td>32</td>
<td>0, 52, 4</td>
<td>90</td>
<td>4.06</td>
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<td>2, 44, 8</td>
<td>10</td>
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<td>High &gt; low attention (DLPFC)</td>
<td>Increased</td>
<td>Dorsal ACC</td>
<td>R</td>
<td>32</td>
<td>6, 18, 44</td>
<td>55</td>
<td>4.39</td>
</tr>
<tr>
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<td>Ventral ACC</td>
<td>R</td>
<td>32</td>
<td>16, 42, 10</td>
<td>22</td>
<td>3.28</td>
<td></td>
</tr>
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</table>

Note: Activation clusters for the respective contrasts. Voxel-level threshold, \(P < 0.005\) (uncorrected); 10 voxel spatial extent; H, hemisphere; BA, Brodmann area; cluster size in voxels; \(T\), \(T\)-values.

**Figure 4.** Main effects of the task: (A) an activation overlay for activity associated with the emotional valence effect (negative trials minus positive trials) in the right amygdala (MNI \(x = 24, y = -2, z = -18; 26\) voxels), at \(P < 0.001\) with a cluster size of \(>10\) voxels displayed on a single subject’s anatomical slices; (B) an activation overlay for activity associated with emotional conflict (incongruent trials minus congruent trials) in the right ACC (MNI \(x = 8, y = 46, z = 28; 48\) voxels) at \(P < 0.001\) with a cluster size of \(>10\) voxels) displayed on a single subject’s anatomical slices; (C) an activation overlay for activity associated with higher attentional load (scene trials minus face trials) in the right DLPFC (MNI \(x = 54, y = 24, z = 24; 241\) voxels), at \(P < 0.001\) with a cluster size of \(>10\) voxels displayed on a single subject’s anatomical slices.
and the right DLPFC ($x, y, z = 46, 24, 20; T = 3.44; P < 0.005$) but decreased functional coupling with ventral ACC ($x, y, z = 0, 52, 4; T = 4.06; P < 0.005$) (Table 2 and Fig. 5I).

Finally, as a result of increased attentional demands, increased functional connectivity was noted between the DLPFC and the dorsal ACC ($x, y, z = 6, 18, 44; T = 4.39; P < 0.005$) but decreased connectivity with ventral ACC ($x, y, z = 16, 42, 10; T = 3.28; P < 0.005$) (Table 2 and Fig. 5II).

### Discussion

The present study explored the respective contribution of appraisal (bottom-up) and regulation (top-down) processes during emotion processing. By varying 3 parameters, emotional valence, emotional congruency and allocation of attention, we were able to discriminate specific functional variations in BOLD activity and connectivity of the amygdala, ACC, and DLPFC. For emotional valence effects, we observed higher
amygdala activation for negative images than for positive ones. This effect of valence was also associated with strengthened functional interaction between amygdala and both the dorsal ACC and DLPFC as well as with reduced functional connectivity between amygdala and the ventral ACC. Examination of congruency effects showed that emotional conflict (i.e., the incongruent condition) was associated with enhanced activation within the dorsal ACC. Furthermore, PPI analyses revealed higher functional connectivity between the dorsal ACC and both the DLPFC and the amygdala as well as lower functional coupling between ACC subregions. Finally, concerning the attentional effects, we observed higher DLPFC engagement, as well as enhanced coupling between the DLPFC and the dorsal ACC, but lower coupling between DLPFC and ventral ACC when switching from conditions of low to high attentional load.

Valence Effect

Behavioral data indicated that negative stimuli were associated with slower reaction time and poorer accuracy than positive stimuli. This challenges the traditional view, which posits that negative or threat-related emotions are signals receiving privileged access to attention in order to allow for quick reaction to the potential threat (Ohman et al. 2001; Vuilleumier et al. 2001; Vuilleumier 2005). However, even though threat-related items benefit from enhanced sensory treatment, they also divert processing resources away from other cognitive operations, leading eventually to impaired behavioral performances (Pessoa 2009).

Accordingly, numerous studies have found people to be faster and more accurate in identifying positive faces than negative faces (Leppänen and Hietanen 2004; Juth et al. 2005; Tottenham et al. 2009). Suggested explanations for the behavioral benefit of processing positive faces include facilitated expression categorization and speeded selection among response alternatives (Calvo and Nummenmaa 2008; Calvo and Beltrán 2013). Of importance, the advantage observed for positive valence faces has been shown to extend to non-facial picture stimuli as well (Maljkovic and Martini 2005; Becker 2012).

As anticipated, we observed a greater amygdala response to negative images than to positive ones. Even though amygdala is known to respond to negative stimuli (LeDoux 2003; Zald 2003; Adolphs 2008), some studies have argued that positively valenced stimuli could also induce amygdala activity (Yang et al. 2002; Fitzgerald et al. 2006; Sergerie et al. 2008; Ball et al. 2009). However, the few works that have directly examined contrasts of unpleasant versus pleasant stimuli reported greater amygdala activation in these contrasts (Morris et al. 1998; Whalen et al. 1998; Paradiso et al. 1999; Rauch et al. 2000; Gorno-Tempini et al. 2001; Pessoa et al. 2002), which our present results confirm.

The right-lateralized activation of the amygdala observed here may appear at odds with findings of several meta-analyses indicating more frequent activity in the left than in the right amygdala for negative emotional cues (Wager et al. 2003; Sergerie et al. 2008; McMenamin and Marsolek 2013). Nonetheless, the robust activation of the right amygdala observed in our study may be explained by the task design, because left amygdala has been observed only in block designs as opposed to event-related designs (Sergerie et al. 2008; McMenamin and Marsolek 2013). Moreover, it has been suggested that the type of emotional material processed differs between the right and left amygdala. Specifically, left amygdala would be involved in the processing of semantic material (e.g., sentences or words), whereas the right amygdala would be preferentially involved in processing of non-semantic information (e.g., faces, pictures) (Markowitsch 1998; Phelps et al. 2001), as in the present task. Note, however, that our study did not intend and was not designed to test hemispheric asymmetry. In fact, left amygdala activation was observed in our study when the threshold was raised to $P<0.01$ uncorrected. Thus, it is possible that the absence of left amygdala here was merely due to lack of statistical power. We also found higher response to negative stimuli in bilateral DLPFC. As discussed earlier, this could be the consequence of bottom-up processes, that is, enhanced attentional resources allocated to negative stimuli.

Concurrently, we found that negative compared with positive valence enhanced functional interaction between right amygdala and both DLPFC and dorsal ACC but reduced functional coupling between amygdala and ventral ACC. As previously mentioned, both DLPFC and dorsal ACC are known for their implication in executive control. Once again, increased functional connectivity between amygdala and these prefrontal regions is in line with bottom-up influences of the amygdala on attentional control regions; in other words, it could reveal the natural bias towards emotion-laden information. In addition, the dorsal “cognitive” circuit recruitment would lead to reduction in functional interaction within the ventral “affective” circuit, in agreement with the negative relationship between these circuits (Ochsner and Gross 2005; Pezawas et al. 2005; Margulies et al. 2007; Phillips et al. 2008).

Emotional Congruency Effect

As expected, emotionally incongruent trials resulted in slower reaction times than congruent trials, replicating results of previous studies (Egner et al. 2008; Wittfoth et al. 2010; Kanske and Kotz 2011). Simultaneously, emotional incongruency was associated with activity in the ACC. These data support the view that ACC plays a prominent role in monitoring conflict between affective cues (Etkin et al. 2006; Egner et al. 2008; Ochsner et al. 2009; Wittfoth et al. 2010). The anatomical location of this activity falls within the dorsal or supragenual division of the ACC (although we also found a cluster in the more ventral part of the ACC, this activation survived cluster-wise but not voxel-wise FWE correction for multiple comparisons). This result is consistent with the role of dorsal (Etkin et al. 2006; Haas et al. 2006; Egner et al. 2008; Ochsner et al. 2009; Chiew and Braver 2011; Kanske and Kotz 2011; Wittfoth et al. 2010) and the ventral ACC (Bush et al. 2000; MacDonald 2000; Egner and Hirsch 2005; van Veen and Carter 2005; Etkin et al. 2006; Mohanty et al. 2007) in emotional conflict. The distinctive roles of these 2 subregions could lie in the fact that dorsal ACC has a broader role in conflict detection whereas ventral ACC is specifically associated with resolution of conflict (Etkin et al. 2006; Egner et al. 2008). We also found a modest activation in the left amygdala, consistent with the affective reaction that may occur when conflict is generated (Etkin et al. 2006; Egner et al. 2008).

PPIn analyses showed higher connectivity between dorsal ACC and both the amygdala and the DLPFC during emotional conflict processing but reduced functional coupling with ventral ACC. These results underline the well-established ventral/dorsal functional dichotomy within ACC (Bush et al. 2000; Mohanty et al. 2007) and suggest that the negative coupling between
dorsal and ventral ACC seen at rest (Margulies et al. 2007) might extend to emotional processing tasks. Besides, given the large anatomical efferent projections from dorsal ACC to the amygdala in the primate brain (Ghashghaei et al. 2007; Cho et al. 2013) and the functional evidence in rodents (Rosenkranz and Grace 2002; Maren and Quirk 2004) and humans (Delgado et al. 2008) indicating that dorsal ACC modulates amygdala activity by inhibition, we can speculate that the increased dorsal ACC–amygdala coupling seen here reflects top-down negative modulation of dorsal ACC on amygdala activity. Finally, PPI findings revealed enhanced functional coupling within the dorsal ACC–DLPFC circuit. This result is consistent with the notion that conflict monitoring relies on the interplay between ACC and the lateral PFC (LPFC) (MacDonald 2000; Kerns et al. 2004). Hence, in the presence of conflict, ACC would recruit the LPFC, a region involved in cognitive control (MacDonald 2000; Miller and Cohen 2001). In turn, PFC engagement would reduce the detrimental effect of conflict through behavioral adjustment (Miller and Cohen 2001; Kerns et al. 2004; Egner and Hirsch 2005; Egner et al. 2008).

**Attention Effect**

As expected, the contrast of high versus low attentional demand revealed a robust activation in the DLPFC. We hypothesized that focusing on scenes, compared with faces, would require higher attribution of attentional resources because the inherent properties of IAPS scenes make their processing more difficult and effortful. IAPS pictures are complex scenes containing various features. To grasp the significance of the scenes, subjects must bind the different elements, a process requiring increased attentional resources (Treisman 1999). Besides, faces are composed of common features (nose, mouth etc...) making their complexion predictable and relatively invariant perceptually, whereas IAPS pictures depict scenes with rich and varied contexts rendering them more unique (Britton et al. 2006). In contrast to scenes, facial expressions, because of their immediate, prepotent, and unequivocal message, constitute directly accessible information that necessitates less attentional resources (Hariri et al. 2002). Indeed, studies using emotional masked faces to activate regions involved in affective processing have demonstrated that emotional appraisal due to facial expression can occur automatically and unconsciously (Morris et al. 1996; Whalen et al. 1998; Liddell et al. 2005). Finally, and most critically, our theoretical assumption was supported empirically by poorer performance (longer RTs and lower accuracy) in the high than low attentional load conditions.

The DLPFC is considered a key region in top-down modulation of stimulus processing (Miller and Cohen 2001; Corbetta and Shulman 2002; Kerns et al. 2004). Together with parietal cortex and the dorsal part of ACC, DLPFC has been implicated in attentional control (Carter 1998; Botvinick et al. 1999; MacDonald 2000; Miller and Cohen 2001; Corbetta and Shulman 2002; Fan et al. 2003; Kerns et al. 2004; Langenecker et al. 2004). DLPFC is particularly known to play a prominent role in the allocation of attentional resources (MacDonald 2000; Blasi et al. 2007), which can be used to bias processing in favor of task-relevant stimuli. Thus, although emotional faces and scenes are processed through partly overlapping networks (Britton et al. 2006), our findings suggest that IAPS scenes recruit frontal regions involved in attentional control to a greater extent.

Functional connectivity analyses indicated that DLPFC showed stronger interaction with dorsal ACC but weaker functional coupling with ventral ACC after increased attentional demand. Thus, when greater cognitive effort is needed, DLPFC-dorsal ACC functional coupling is reinforced, in keeping with the role that both regions play in cognitive control (Kerns et al. 2004; Blasi et al. 2007). Simultaneously, the reduced functional interaction observed between the dorsal/cognitive circuit and ventral ACC is consistent with the strengthened dichotomy between executive regions and default mode regions, such as medial prefrontal regions, which occurs as the attentional demand of the task increases (Fox et al. 2005; Whitfield-Gabrieli et al. 2009).

This study has some limitations. First, emotion processing is a complex mechanism that includes various components and involves numerous brain regions; it cannot be reduced solely to appraisal and regulation processes. However, we chose to restrict the scope of this study to 2 particular aspects of emotion processing and to focus on a consistent and unanimously acknowledged neuronal circuit. Second, the gender proportion was not balanced in our population. This may be a concern, given the influence that gender could have on emotional processing (Hamann et al. 2004; Cahill 2006; Whittle et al. 2011). However, we repeated the analysis with sex as covariate, and this did not modify our findings. Nonetheless, it could be of interest to apply this task in larger and more balanced samples to investigate whether males and females exhibit different patterns of emotion processing. Another potential issue is the utilization of 3 emotions in the negative valenced stimuli, which could have increased variability in the result. This concern echoes the longstanding controversy in the field of emotion research concerning whether emotions are better conceptualized in terms of discrete categories, such as fear and anger (Ekman 1992), or underlying dimensions, such as arousal and valence (Barrett 1998). Although there is physiological and neuroimaging evidence in support of both theories (Lang and Bradley 2010; Stephens et al. 2010; Vytal and Hamann 2010; Lindquist et al. 2012), we adopted the dimensional approach because the standardized set of IAPS pictures, used here, was originally rated in terms of valence and arousal (Lang et al. 2008). Besides, although neuroimaging studies have identified consistent neural correlates associated with basic emotions, they failed to establish clearly discriminable neuronal patterns because individual brain regions often contributed to multiple emotions (Hamann 2012; Lindquist et al. 2012). Nevertheless, we repeated the analyses to compare the brain activity induced by the 3 negative emotions. For our region of interest, results indicated no difference depending on the negative emotion category. Finally, the interpretation of our connectivity analyses must be considered within the context of the inherent limitations of functional connectivity measures. PPI connectivity analyses rely on statistical correlations and thus cannot indicate the directionality of regional influence. Nevertheless, we believe that theory and previous research lay a reasonable foundation for the model presented.

**Conclusion**

We propose here a novel fMRI paradigm, the VAAT, designed to dissociate bottom-up and top-down mechanisms in emotion processing. By independently manipulating the 3 parameters...
of our task, namely emotional valence, emotional congruency, and attentional load, in a factorial design, we dissociated bottom-up (valence) from top-down (congruency and attention) subnetworks within the cortico-limbic circuit. The amygdala, and its connections to the dorsal “cognitive” circuit (DLPFC and dorsal ACC), was engaged by bottom-up emotional processing while the connectivity within the ventral “affective” circuit diminished. On the other hand, dorsal ACC, and its connections to DLPFC and amygdala, was preferentially recruited by top-down resolution of emotional conflict whereas the functional coupling between the dorsal and ventral parts of ACC decreased. Finally, DLPFC, and its connections to dorsal ACC, was engaged by top-down attentional control whereas its connectivity with ventral ACC was reduced.

Given the central role of emotional dysregulation in many, if not all, psychiatric illnesses, this task may allow for better characterization of the neural correlates underlying these disorders.

Supplementary Material
Supplementary material can be found at: http://www.cercor.oxfordjournals.org/

Funding
This work was supported by research grants from Bristol-Myers Squibb Company and Otsuka Pharmaceutical Company, the multi-agency thematic institute (ITMO), and Pierre Hauriez foundation.

References
Becker MW. 2012. Negative emotional photographs are identified more slowly than positive photographs. Atten Percept Psychophys. 74:1241–1251.

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