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Functional Dissociation of the Posterior and Anterior Insula in

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emotions, the mid-insula plays a role in encoding contextual integration (Craig, 2002, 2009), and the anterior insula (AI) plays a role in encoding introspective awareness of emotion and bodily states (Critchley et al., 2004; Paulus and Stein, 2006). This hypothesis provides a new perspective for understanding how a complicated, high-level mentality or emotionality is built or developed from more basic feelings.

A neuroimaging study investigating the relationship between

moral transgressions and the emotional arousal associated with each moral transgression event. The severity of the moral transgressions was rated using a six-point Likert scale from 1 ("not serious at all") to 6 ("extremely serious"), and five emotions, i.e., disgust, anger, surprise, sadness and disappointment, were rated using a seven-point Likert scales from 0 ("no feeling at all") to 6 ("extremely strong"). The 60 sentences had a similar length and complexity and were divided into three equal groups according to the severity and emotional arousal ratings. No statistically significant differences were observed in the severity of the moral transgressions, moral disgust or other types of emotional arousal among the three groups of materials (**Supplementary Table S1**). In the fMRI experiment, each group of materials was assigned to only one of the three experimental conditions that used the "stranger," "mother," or "best friend" as the behavioral agent (**Table 1**). The assignment of a given agent to a given group of materials were counter-balanced across the participants.

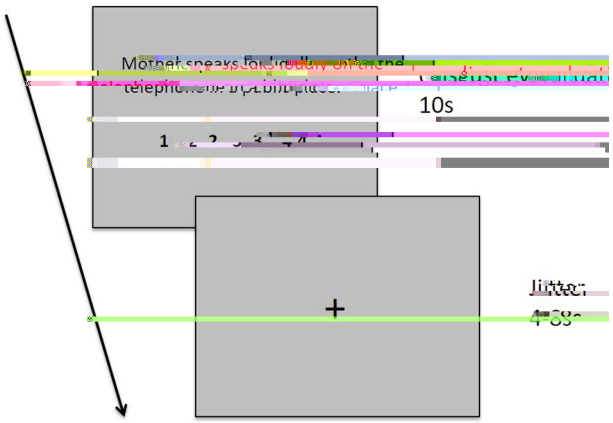

FIGURE 1 | Task design. Each sentence was displayed for 10 s, followed by the presentation of a fixation cross for 4–8 s. A fixation cross was presented for 30 s at the start and end of the task and for 16 s during each trial.

Task Design and Procedures

During the experimental fMRI scan session, the participants were asked to read and evaluate 60 sentences describing different moral transgression events with different behavioral agents (mother, stranger, and best friend) one-by-one. Each sentence was presented for 10 s, followed by a cross fixation phase of a varied duration ranging from 4 to 8 s. During the 10-s sentence presentation stage, the participants were instructed to read and comprehend the situation described by the sentence and evaluate their degree of disgust using a four-point Likert scale from 1 ("not disgusting at all") to 4 ("extremely disgusting"). The participants were required to indicate their evaluation by pressing one of four buttons using their index, middle, fourth, or little finger of their right hand. The degree of disgust and the numbers 1, 2, 3, and 4 were presented below the sentence (see **Figure 1** for a detailed description of each sentence).

To prevent the participants from frequently switching between the behavioral agent of the moral transgressions, 20 sentences in each condition were separated into two sub-groups with 10 sentences in each sub-group, and the 10 sentences in each sub-group were presented successively in one block. Therefore, two blocks of each of the three experimental conditions involved the mother, stranger and best friend as the behavioral agent, and the participants completed a total of six blocks. The sequences of the block presentations were counter-balanced across the participants with the restriction that two blocks in the same condition could never be presented successively. During the periods between the blocks, a fixation (cross-viewing) was presented for 16 s. In addition, 30-s fixation periods were presented at the beginning and end of the session.

TABLE 1 | Sample sentences from the experimental materials.

	Stranger	Best friend	Mother
Moral disgust	Stranger says dirty words in a public place	Best friend chats at a concert	Mother speaks on the telephone loudly in a public place

Image Acquisition

All MRI scans were acquired using a Siemens MAGNETOM Trio 3T MR scanner at the Imaging Center for Brain Research at Beijing Normal University. Foam padding and a plastic brace were used to minimize head movement. For the functional imaging, the whole-brain coverage of 33 axial slices was acquired using a T2-weighted echo-planar imaging sequence based on the blood oxygenation level-dependent (BOLD) contrast with the following parameters: 2000 ms repetition time (TR), 30 ms echo time (TE), 90° flip angle, 4.0-mm slice thickness, 0.6-mm gap, 64 × 64 data matrix, 200-mm field of view (FOV), and 3.1 × 3.1 × 4.0-mm voxel size. In addition, 3D structural brain scans were also acquired for each participant using a T1-weighted anatomical scan with the following parameters: 2530-ms TR, 3.39-ms TE, 7° flip angle, 256 × 256 data matrix, 256-mm FOV, 1.3 × 1.0 × 1.3-mm voxel size, and Bandwidth (BW) = 190 Hz/pixel.

Image Data Analysis

The event-related analyses of friend of friendof 2l57

BOLD signals on the regressors to identify the relationship between the hemodynamic responses and task events. Low-frequency drifts in the signal were removed using a high-pass filter with a 128-s cuto. Regressors were created by convolving a train of delta functions representing the sequence of individual events using the default SPM basis function, which consists of a synthetic hemodynamic response function (HRF) composed of two gamma functions (Friston et al., 1998). Three regressors were used for the three conditions (mother, best friend and stranger). The 6 parameters generated during the motion correction were also entered as covariates. In addition, HRF related to trials in which the participants failed to respond was also modeled separately and explicitly to partial out error-related activity. Linear contrasts of the parameter estimates were performed to identify the e ects of the three conditions and the di erence between every two conditions in each session. Then, the first level contrasts were aggregated into a second level, and one-sample *t*-tests were performed to compute the group-level statistics using a random-e ects model.

Regions of Interest (ROIs) and Psychophysiological Interaction (PPI) Analysis

To define the regions of interest (ROIs), we first conducted contrasts between the stranger condition and the mother condition. To test our hypotheses regarding the role of the AI and PI in moral disgust, ROI analyses were performed based on the templates developed by Lin and colleagues (Lin et al., 2013), which consisted of six insula regions, including the left and right AI (LAI and RAI), left and right PI (LPI and RPI), and left and right middle insula (LMI and RMI). The ROIs were defined by masking the six abovementioned insula regions on the whole brain results of a given contrast (e.g., the contrast of "mother condition minus stranger condition" and the contrast of "stranger condition minus mother condition"). The significance level was set at an uncorrected threshold of $p < 0.05$ with a cluster extent of at least 5 contiguous voxels. The LAI was activated in the mother condition minus the stranger condition. The LPI was activated in the stranger condition minus the mother condition (see **Table 2** for details). ROIs as clusters were created for the LAI and LPI. The BOLD signal changes were extracted from each ROI for the contrast between the stranger condition and the mother condition. Separate psychophysiological interaction (PPI) analyses were also performed using the LAI or LPI ROIs as seeds.

Psychophysiological interaction analyses provide a measure of functional connectivity change among di erent brain regions depending on a specific psychological context (Friston et al., 1997). This analysis was achieved using a moderator derived from the product of the activity of a source region

caused by the individuals' reluctance to think negative thoughts about their mother, and this inhibitory process might prevent individuals from further processing the sentences about their mothers.

In this study, although the processing of the materials evoked complicated feelings, emotions and cognitive processes, the feeling of moral disgust could be essentially involved in this complicated processing. Due to its well-established role in disgust, the insula could play a key role in representing moral disgust. However, in the present study, we could not completely justify that the observed insular activation did represent moral disgust rather than other feelings or thoughts. We did not find a significant correlation between the insular activation and individual subjective evaluations of disgust toward the immoral events. A possible interpretation is that the subjective evaluation of moral disgust is a holistic impression consisting of complicated cognitions, emotions, experiences, and social attitudes toward the transgression event. The element of disgust represented by the insula was not su□ciently strong to be reflected by this subjective evaluation. Further studies should adopt specific judgments that are more sensitive to detect the disgust element in moral judgment and verify the role of the insula in disgust representation.

In summary, in this study, we doubly dissociated two insular components in the processing of moral transgression events, and the component located in the posterior region was more activated in the stranger condition, while the other component located in the anterior region was more activated in the mother condition. Given that both the PI and AI were positively activated in the mother and stranger conditions (the signal change in the AI and PI regions was positive in both conditions), we propose that these two components may have been generally involved in both conditions regardless of the behavioral agent

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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