

Neural substrates of self-referential processing in Chinese Buddhists

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Our recent work showed that self-trait judgment is associated with increased activity in the ventral medial prefrontal cortex

that self-referential processing was associated with increased activity in the VMPFC in non-religious participants but in the DMPFC in Christians. As the VMPFC and DMPFC engage, respectively, in encoding stimulus, self-relevance and evaluation of self-referential stimuli (Northoff *e al.*, 2006), we proposed that Christians' self-referential processing is characterized by weakened coding of stimulus self-relatedness but enhanced evaluative processes. This is consistent with Christian spiritual request for self-transcendence (i.e. to deny oneself in order to live a spiritual life as dictated by Jesus; Ching, 1984; McDaniel, 1987). The transcultural neuroimaging findings illustrate how cultures influence the neural basis of self-related processing and reinforce the emergence of cultural neuroscience (Chiao and Ambadi, 2007; Han and Northoff, 2008).

The current work further investigated neural consequences of Buddhist belief and practice on self-referential processing. One of the most famous claims of Buddhism is that the self does not exist (Ishigami-Iagolnitzer, 1997; Albahari, 2006). According to the teaching of Buddha, one of the three characteristics of existence is selflessness (Kornfield, 1977) and the idea of self is an imaginary false belief that has no corresponding reality (Ching, 1984). The aim of Buddhist practice is to get rid of one's mindset of any sense of 'me' or 'mine' (Albahari, 2006). The doctrine of No-self produces great influence on Buddhists' life style. However, to date, we know little about the potential consequence of Buddhist belief and practice on neural underpinnings of self-referential processing. The current research examined whether Buddhist doctrine of No-self results in modulation of MPFC activity in a way similar to that observed in Christians (Han *e al.*, 2008). Moreover, we assessed whether asking Buddhists to conduct self-reflective thinking induces neural activity related to conflict monitoring since the task itself conflicts with the doctrine of No-self.

We scanned Chinese Buddhists in trait judgment tasks associated with the self and a public person (the former Chinese premier Zhu Rongji). Region-of-interest (ROI) analysis was first conducted to examine the engagement of VMPFC and DMPFC in self-referential processing. Previous studies roughly divided the MPFC into the VMPFC for coordinate ≤ 10 mm and the DMPFC for coordinate > 10 mm (Ochsner *e al.*, 2005; D'Argembeau *e al.*, 2008). The VMPFC and DMPFC in the current work were defined using a priori anatomically defined regions based on an entirely independent data set (Han *e al.*, 2008). Similar to our previous studies (Zhu *e al.*, 2007; Han *e al.*, 2008), the contrast between self-judgment and Zhu Rongji judgment identified neural substrates of self-referential processing in Buddhists. The contrast of Sakyamuni and Jesus judgments was also conducted to examine whether specific neurocognitive processes are involved in trait judgments of the leader of one's own religion or a different religion. Finally, since relative to the self-judgment, trait judgments of others engage enhanced episodic memory retrieval of behavioral

evidence (Klein *e al.*, 2002) that is associated with increased functional connectivity between MPFC and posterior parietal cortex (PPC)/precuneus (Lou *e al.*, 2004; Babiloni *e al.*, 2006), we conducted functional connectivity analysis to examine neural processes that may distinguish between trait judgments of the self and others in Chinese Buddhists. Such analysis has identified reduced functional connectivity between MPFC and PPC/precuneus during self-trait judgment compared with trait judgment of others in non-religious and Christian subjects (Ge *e al.*, in press), which is in accord with the idea that self-trait judgment engages increased employment of semantic trait summary but decreased memory retrieval of behavioral episodes (Klein *e al.*, 1992, 2002, 2008; Klein and Loftus, 1993).

METHODS

Subjects

Fourteen self-identified Chinese Buddhists (seven males, seven females, 21–31 years of age, mean 25.4 ± 2.46) participated in this study as paid volunteers. The participants had been attached to local faith communities for 1–7 years (mean 2.5 ± 2.0) when they participated in this study. Eleven participants reported to attend the community activity at least once a week. Twelve participants reported to cultivate themselves according to Mahayana (one of the major schools of Buddhism) doctrine everyday. Ten participants reported to read sutra everyday. The participants were asked to rate the importance they placed on religion and their attitude toward Buddha, based on a 5-point scale (0 = not important or do not believe at all, 4 = very important or strongly believe), resulting a mean rating score of 3.56 ± 0.43 . All participants had no neurological or psychiatric history. All participants had college education, were right-handed and had normal or corrected-to-normal vision. Informed consent was obtained from all participants prior to scanning. This study was approved by a local ethics committee.

Stimuli and procedure

Participants were first imaged while performing trait judgment tasks. The stimuli were presented through an LCD projector onto a rear-projection screen mounted above subjects' heads. The screen was viewed with an angled mirror positioned on the head coil. There were three functional scans. Each scan consisted of five sessions during which participants, respectively, conducted the following judgment tasks: (i) self-judgment: does this adjective describe you?; (ii) Jesus judgment: does this adjective describe Jesus?; (iii) Sakyamuni judgment: does this adjective describe Sakyamuni?; (iv) other judgment: does this adjective describe Zhu Rongji (the former Chinese premier)?; and (v) font judgment: is the word presented written in bold faced character? The questions and traits were in Chinese. Subjects made judgments after the presentation of each trait adjective by pressing one of the two buttons with the left or right thumb. The assignment of 'yes' response to the left

or right buttons was counterbalanced across subjects. Participants did not press any button if they were not sure about the judgment task. Percentage of 'yes' and 'no' responses was recorded. The judgment tasks were intervened by null sessions during which subjects were presented with two rows of asterisks (*) replacing the words in the judgment tasks. The participants were asked to passively view the symbols in the null sessions. The order of the judgment tasks was random.

Each session began with the presentation of a 'cue' (either 'Self', 'Jesus', 'Sakyamuni', 'Zhu Rongji' or 'Font') on the screen for 4 s. The words were black on a white background. A trait adjective then appeared above the cue word with a duration of 2 s. There was a 1 s interstimulus interval before the next trait adjective was presented. Each of the Chinese character in trait adjectives and cue words was 2.4×2.4 cm (width \times height) and 0.8×0.8 cm, subtending a visual angle of $1.5^\circ \times 1.5^\circ$ and $0.5^\circ \times 0.5^\circ$ at a viewing distance of 90 cm. Sixteen trait adjectives were presented in each session of the functional scans. Thus, each session of the judgment tasks lasted for 52 s. The large and small symbols used in the null condition were 1.1×1.1 cm ($0.7 \times 0.7^\circ$) and 0.5×0.5 cm ($0.3 \times 0.3^\circ$). Each null session lasted for 20 s including a 4 s instruction that asked subjects to view the screen passively.

A total of 240 unique adjectives were selected from established personality trait adjective pools (Liu, 1990). The adjectives were classified into 15 lists of 16 words. Each Chinese adjective consisted of two characters. One-half of the adjectives were positive and the other half negative. Fifteen lists of words were pseudo-randomly selected for the judgment tasks, while the remaining 15 lists of words were used in the latter recognition memory test as new words. After the functional and anatomy scans, participants took 1 h break and then were given a 'surprise' recognition memory test. All the trait adjectives used in the judgment tasks were mixed with another 240 new trait adjectives and were presented in a random order. Participants were asked to identify old or new items by pressing one of the two buttons. If a trait adjective was identified as a 'new' word, it disappeared after subjects' response. If a trait adjective was identified as an 'old' word, participants were further asked to make an R/K judgment by indicating whether 'remembering' (R) the item or simply 'knowing' (K) the item. A 'remembering' item was defined as one for which subjects can consciously recollect specific details of the item that appeared in the earlier list. A 'knowing' item was defined as one that is not accompanied by recollective experience but has a feeling of knowing or familiarity to the participants. During the recognition memory test, participants were required to respond to every item without a time limit.

MRI data acquisition

Brain imaging was performed on a 3 T Siemens Trio MR scanner with a standard birdcage head coil at Beijing MRI Center for Brain Research. Pieces of foam were used

to minimize head movement. A T2* weighted gradient-echo planar imaging sequence (TR =

Table 1 Results of behavioral performances [mean (s.d.)]

Group	Self	Sakyamuni	Jesus	Zhu Rongji	Font
Behavioral data during scanning					
'Yes' responses (%)	0.466 (0.029)	0.426 (0.016)	0.412 (0.019)	0.435 (0.015)	
RTs (ms)	1093 (34)	932 (22)	950 (25)	1044 (48)	751 (22)
Behavioral data of memory test					
Total recognition score	0.57 (0.135)	0.45 (0.144)	0.40 (0.179)	0.41 (0.122)	0.10 (0.113)
Score of remember	0.61 (0.113)	0.47 (0.147)	0.35 (0.223)	0.40 (0.158)	0.13 (0.127)

areas as ROIs. The whole brain analysis is more conservative and is a kind of *post hoc* analysis. Thus, both ROI and whole brain analyses were reported.

A psychophysiological interaction (PPI) analysis (Friston *et al.*, 1997) was performed in order to identify brain regions that showed significantly different covariation (i.e. functional connectivity) with DMPFC activity between trait judgments of the self and others. The coordinates of the peak voxel from the random effect analysis were used to serve as a landmark for the individual seed voxels. An ROI of a sphere with a radius of 5 mm in the DMPFC was defined around the peak voxel. The time series of each ROI was then extracted, and the PPI regressor was calculated as the element-by-element product of the mean-corrected activity of this ROI and a vector coding for the differential task effect of trait judgment of others – trait judgment of the self. The PPI regressors reflected the interaction between psychological variable (trait judgments of others – self) and the activation time course of DMPFC. The individual contrast images reflecting the effects of the PPI between DMPFC and other brain areas were subsequently subjected to one-sample *t*-tests. The results of the group analysis identified brain regions of which the activity systematically showed increased correlation with MPFC activity during trait judgment of others. The threshold at the cluster level was set to $P < 0.05$ (corrected for multiple comparisons) for the identification of brain areas that showed significantly different functional connectivity with the seed ROIs.

RESULTS

Behavioral performance

Response accuracy was high in the font-judgment task (98.3%). Participants failed to make any responses on <3% of the trials in trait judgment tasks. Table 1 shows the proportion of 'yes' responses and reaction times in the judgment tasks during the scanning procedure. The proportion of 'yes' responses did not differ between different trait judgment tasks [$F(3,39) = 1.78$, $P = 0.167$]. Participants responded faster to font judgment than to trait judgment tasks [$F(4,52) = 33.19$, $P < 0.001$] and slower to self-judgment than to Jesus and Sakyamuni judgments [$F(1,13) = 6.393$ and 4.838 , both P 's < 0.001].

Table 1 also shows corrected recognition scores (i.e. the proportion of hits minus false alarms) in the recognition

memory test. The corrected recognition scores were higher for trait words associated with deep semantic processing (self-, Jesus-, Sakyamuni-, Zhu Rongji judgments) than for those associated with font judgment [$F(4,52) = 41.62$, $P < 0.001$]. In addition, the corrected recognition scores were higher in self-judgment than Jesus-, Sakyamuni- and Zhu Rongji judgment conditions (all P 's < 0.01) but did not differ among Jesus-, Sakyamuni- and Zhu Rongji judgments (P 's > 0.05). Similar results were observed for the scores of remembering, which were higher for trait words associated with self-judgment than those linked to Jesus-, Sakyamuni- and Zhu Rongji judgments [$F(4,52) = 25.61$, $P < 0.001$]. The scores of remembering did not differ between Jesus-, Sakyamuni- and Zhu Rongji judgments (P 's > 0.05). Thus, the results of memory test revealed a self-reference superiority effect in memory.

fMRI data

The ROI analysis first calculated signal changes in the VMPFC and DMPFC associated with trait judgments to examine whether self-judgment can be dissociated from Zhu Rongji judgment in these brain areas. Paired *t*-tests confirmed that signal changes in the VMPFC did not differ significantly between self- and Zhu Rongji judgments [$F(1,13) = 0.115$, $P = 0.911$]. However, signal changes in the DMPFC were significantly larger when participants made self- than Zhu Rongji judgments [$F(1,13) = 2.117$, $P = 0.05$, Figure 1].

A whole brain SPM analysis was conducted to compare self- with font judgments to identify neural activities linked to the semantic processing associated with trait judgments. This identified increased activations in the superior MPFC, the left superior and inferior frontal cortex, the left middle temporal cortex and the right cerebellum (Table 2). To identify the neural structures involved in self-referential processing in Buddhists, we calculated the contrast between self- and Zhu Rongji judgment, which showed increased activity in the DMPFC/rostral ACC ($P < 0.05$, corrected for multiple comparisons) (Figure 2 and Table 2). The contrast of self- Zhu Rongji judgment also showed greater activation in the midcingulate and left frontal/anterior insular cortex. The contrast of self- and Sakyamuni judgment was also associated with increased activations in the DMPFC/rostral ACC and left frontal/anterior insular cortex. To examine if

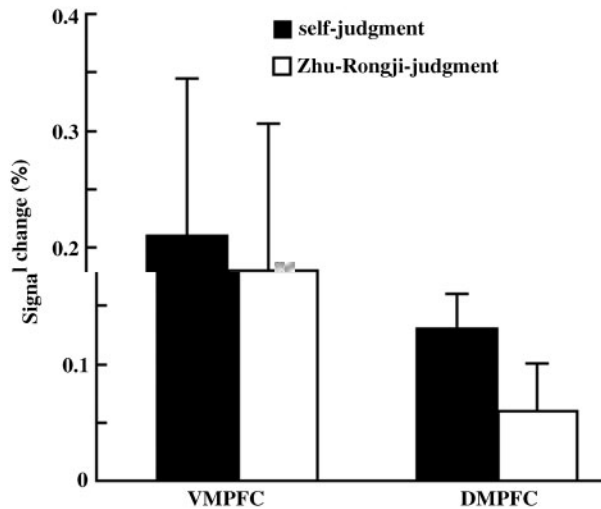


Fig. 1 The results of ROI analysis. Percent signal changes at the VMPFC and DMPFC were plotted separately in the self- and Zhu Rongji judgment conditions.

Table 2 Brain activations in association with contrasts between different judgment tasks

Condition/region	Voxel no.	Z-value	X	Y	Z
Self- vs. Font judgment					
Superior MPFC	2560	5.29	-10	34	50
Left inferior frontal cortex	1155	4.78	-42	26	-4
Left superior frontal cortex	318	4.49	-38	8	46
Left middle temporal gyrus	1120	4.86	-58	0	-12
Right cerebellum	237	3.89	32	-76	-32
Self- vs. Zhu Rongji judgment					
MPFC/rostral ACC	1079	4.72	8	34	28
Midcingulate	291	5.34	-4	-18	42
Left frontal/insular cortex	218	4.33	-34	10	-6
Self- vs. Sakyamuni judgment					
MPFC/rostral ACC	1839	5.45	-4	40	16
Left frontal/insular cortex	291	4.16	-34	8	-6
Sakyamuni vs. Jesus judgment					
Right visual cortex	132	4.43	18	-98	6

Voxels no., number of voxels in a cluster.

there is any neural activity specific to Sakyamuni judgment, we calculated the contrast of Sakyamuni- Jesus judgment and found increased activity in the right visual cortex (Table 2). The reverse contrast, however, failed to show any significant activation.

To examine the relations between the neural activities in the brain areas involved in self-referential processing, we conducted correlation analysis of the contrast values of self- Zhu Rongji judgments in the ROIs (spheres with 10 mm diameter) centered at the peak voxel of DMPFC/ rostral ACC, midcingulate and left frontal/anterior insular cortex. We found that DMPFC/ACC activity in association with self-judgment positively correlated with the activity in the left frontal/insular cortex linked to self-judgment ($r=0.753$, $P=0.002$; Figure 3), suggesting that the greater



Fig. 2 Increased activations shown in the contrast of self- vs Zhu Rongji judgments and self- vs Sakyamuni judgments. Scale bars indicate t-values.

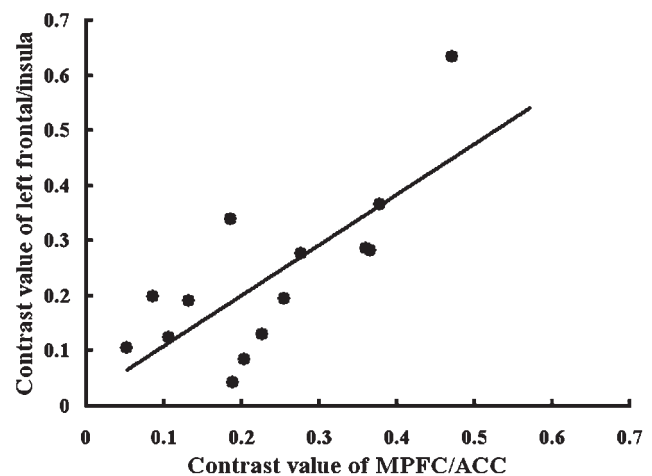


Fig. 3 Correlation between the activity in the MPFC/ACC and the left frontal/insular cortex.

MPFC/ACC activity, the greater the left frontal/insular activity during self-trait judgment. No reliable correlation was observed between neural activities in any other two brain areas.

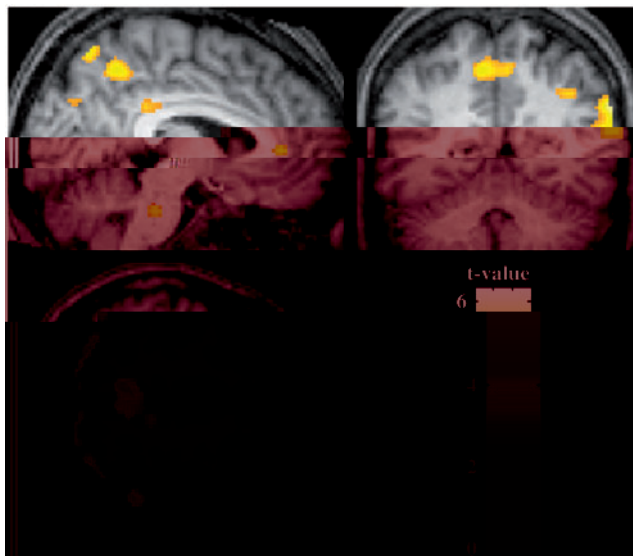


Fig. 4 Brain areas showing increased covariation with the DMPFC during Zhu Rongji judgment relative to self-judgment. Scale bars indicate t-values.

Finally, we conducted PPI analysis to evaluate whether self-trait judgment involves decreased process of episodic memory retrieval and thus induced weakened functional connectivity between the MPFC and the PPC. Since the contrast of self- other trait judgment tasks identified only the DMPFC activation in Buddhist participants, we used the DMPFC as the seed for the PPI analysis. The PPI analysis confirmed that relative to Zhu Rongji judgment, self-judgment resulted in decreased functional connectivity between the DMPFC and medial/bilateral PPC (medial parietal cortex: $-8/-54/52$, $k=659$, $Z=3.37$; right parietal cortex: $68/-40/32$, $k=608$, $Z=4.02$; left parietal cortex: $-20/-82/46$, $k=601$, $Z=4.16$), as illustrated in Figure 4. Similarly, relative to Jesus judgment, self-judgment induced decreased functional connectivity between the DMPFC and PPC (left parietal cortex: $-36/-80/24$, $k=325$, $Z=3.43$). However, similar PPI analysis that compared variation of functional connectivity between Sakyamuni and self-judgment did not show any significant results.

DISCUSSION

This study explored potential influence of Buddhist belief and practice on neural substrates of self-referential processing in trait judgment tasks. Buddhist participants remembered trait words associated with the self better than those associated with others, similar to the observation in Chinese non-religious and Christian participants (Zhu and Zhang, 2002; Zhu *e al.*, 2007; Han *e al.*, 2008). The superior memory for self-referenced trait adjectives suggests that specific neural substrates were involved in the promotion of elaboration and organization of information related to the self during encoding in Buddhists regardless of practicing the doctrine of No-self.

Indeed, our fMRI data revealed specific patterns of neural underpinnings of self-referential processing in Buddhists. First, our fMRI results confirmed that the VMPFC, which has been demonstrated to engage in self-trait judgment in non-religious Chinese (Zhu *e al.*, 2007; Han *e al.*, 2008) and to mediate coding of stimulus self-relevance (Moran *e al.*, 2006; Northoff *e al.*, 2006), failed to differentiate between self- and Zhu Rongji-judgments in Chinese Buddhists. However, DMPFC activity that mediates reappraisal and evaluation of self-related information (Gusnard *e al.*, 2001; Zysset *e al.*, 2002; Northoff *e al.*, 2006) was involved in self-referential processing in Buddhists as DMPFC activity increased significantly to self-judgment compared with Zhu Rongji judgment. The peak voxel of DMPFC cluster (coordinate = 34) fell in the DMPFC defined in the previous studies (Ochsner *e al.*, 2005; D'Argembeau *e al.*, 2008). Such pattern of MPFC activity linked to self-referential processing differs from that observed in non-religious Chinese but is similar to that observed in Chinese Christians (Han *e al.*, 2008). Similar patterns of MPFC activity in Buddhists and Christians are in agreement with the spiritual request for self-transcendence and the doctrine of No-self that begins with the recognition of one's own 'nothingness' (Ching, 1984). A potential psychological consequence of the doctrine of No-self or self-transcendence is to weaken the subjective entity that affords self-relevance of stimuli. As a result, trait judgment of the self in Christians and Buddhists is not characterized by encoding self-relatedness in the VMPFC as that observed in non-religious subjects. Given the functional role of the DMPFC in reappraisal and evaluation of self-related information (Gusnard *e al.*, 2001; Zysset *e al.*, 2002; Northoff *e al.*, 2006), it may be speculated that to compensate for the weakened encoding of self-relatedness, self-trait judgment depends more upon the evaluative processes mediated by the DMPFC in Christians and Buddhists compared with non-religious subjects. Moreover, as the DMPFC engages in inference of others' mental states such as belief or intention (Gallagher *e al.*, 2000; Grèzes *e al.*, 2004), it may be speculated that Buddhists may think about the self from a third person perspective because of elimination of the boundary between self and others. However, although using a strategy different from that used by non-religious subjects, both Christian and Buddhist Chinese showed behavioral self-referential effect during memory retrieval, suggesting that different strategies used for encoding trait words (e.g. enhanced encoding of self-relatedness or enhanced evaluative process of self-related stimuli) may similarly benefit memory retrieval of self-related information.

Interestingly, Buddhists engaged the midcingulate and the left insular cortex during self-trait judgment, which has not been observed in non-religious and Christian Chinese (Zhu *e al.*, 2007; Han *e al.*, 2008). Previous work reported increased activation in the posterior cingulate cortex (PCC) and precuneus in association with self-referential processing

in Westerners (Johnson *e al.*, 2002; Kelley *e al.*, 2002; Moran *e al.*, 2006; D'Argembeau *e al.*, 2008) but not in Chinese (Zhu *e al.*, 2007; Han *e al.*, 2008). The midcingulate activation observed here is anterior to the PCC/precuneus activation observed in the previous work. The midcingulate cortex contributes to multiple emotional and cognitive processes. For example, noxious stimuli inducing dread (Bern *e al.*, 2006) and painful feelings (Singer *e al.*, 2004) result in increased activation in the midcingulate cortex. In the Stroop conflict task, midcingulate activity increased to stimuli when color and semantic meaning of words are incongruent than when they are congruent, suggesting a functional overlap between pain and Stroop effects in the midcingulate (Derbyshire *e al.*, 1998). It appears that the midcingulate is involved in resolving both cognitive and affective conflicts. In our study, self-judgment apparently clashes with Buddhists' belief of No-self that denies the existence of selfhood. The midcingulate activation thus possibly underpins the process of monitoring the conflict between the self-judgment task and Buddhist doctrine of No-self. Interestingly, although self-judgment also generated increased activations in the DMPFC/rostral ACC and left frontal/anterior insular cortex relative to Sakyamuni judgment, the contrast of self- Sakyamuni judgment did not show increased midcingulate activity, possibly because trait judgments of both self and Sakyamuni induced comparable conflicts with the doctrine of No-self.

The left frontal/insular activity related to self-trait judgment has not been reported in the previous studies of Westerners (Johnson *e al.*, 2002; Kelley *e al.*, 2002; Moran *e al.*, 2006; D'Argembeau *e al.*, 2008; Northoff *e al.*, 2006) or non-religious and Christian Chinese (Zhu *e al.*, 2007; Han *e al.*, 2008). However, the insular cortex has been indicated in varieties of neural processes related to the self. Insular activation is associated with judgment of the timing of one's own heartbeats (Critchley *e al.*, 2004) and awareness of causing an action (Farrer and Frith, 2002). Craig (2009) proposed that the insula and adjoining frontal operculum contain an ultimate representation of the sentient self. The unique frontal/insular activity observed here implies that reflective thinking of one's own personal traits in Buddhists may be characterized by enhanced awareness of one's own existence in addition to the engagement of conflict monitoring, which can be assessed in future work that examine the correlation between subjective ratings of the sentient self and the variation of VMPFC and DMPFC activity in Buddhists. The awareness of one's own existence and feelings may coordinate with the evaluative processes of self-related information during self-trait judgment as MPFC/ACC activity positively correlated with the left frontal/insular activity.

We also found reduced functional connectivity between the MPFC and the PPC during self-trait judgment relative to trait judgment of others (Zhu Rongji) in Buddhists, similar

to our previous observations in non-religious and Christian Chinese (Ge *e al.*, under review). According to Klein *e al.* (1992, 2002), self-trait judgment is achieved by accessing a database of summary traits in semantic memory that is abstracted from multiple experiences with one's own trait-relevant behaviors. In contrast, trait judgment of others requires the retrieval of behaviors from episodic memory when there are no sufficient experiences to form a trait summary about the others. The results of functional connectivity analysis suggest

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