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Mortality salience enhances racial in-group bias in empathic neural responses to others' suffering

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ABSTRACT

Behavioral research suggests that mortality salience (MS) leads to increased in-group identification and in-group favoritism in prosocial behavior. What remains unknown is whether and how MS influences brain activity that mediates emotional resonance with in-group and out-group members and is associated with in-group favoritism in helping behavior. The current work investigated MS effects on empathic neural responses to racial in-group and out-group members' suffering. Experiments 1 and 2 respectively recorded event related potentials (ERPs) and blood oxygen level dependent signals to pain/neutral expressions of Asian and Caucasian faces from Chinese adults who had been primed with MS or negative affect (NA). Experiment 1 found that an early frontal/central activity (P2) was more strongly modulated by pain vs. neutral expressions of Asian than Caucasian faces, but this effect was not affected by MS vs. NA priming. However, MS relative to NA priming enhanced racial in-group bias in long-latency neural response to pain expressions over the central/parietal regions (P3). Experiment 2 found that MS vs. NA priming increased racial in-group bias in empathic neural responses to pain expression in the anterior and mid-cingulate cortex. Our findings indicate that reminding mortality enhances brain activity that differentiates between racial in-group and out-group members' emotional states and suggest a neural basis of in-group favoritism under mortality threat.

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Introduction

Being aware of mortality induces anxiety in humans. However, humans have developed different schemes to buffer the fear of death or mortality terror. The Terror Management Theory (TMT, Greenberg et al., 1986; Pyszczynski and Greenberg, 1999) proposed that human beings use both proximal strategies and distal strategies to defense mortality terror. Proximal defenses employ mental strategies to suppress death-related thoughts or to push the worry of death into the remote future. Distal defenses take advantage of cultural worldview and self-esteem that function to buffer the anxiety associated with reminders of death (or mortality salience (MS)) by providing a meaningful, orderly concept system of reality. In addition, according to the TMT, the identification of oneself with a social group has an important anxiety-buffering function because group identification provides a source of worldviews (Harmon-Jones et al., 1996) and offers the prospect of death transcendence if individuals' self-concept can be merged into a lasting collective identity (Castano et al., 2002). The proposition of enhanced group identification and in-group favoritism arising from MS is supported by behavioral findings. For example, after being reminded of death or MS priming, individuals exhibited a greater tendency to confirm the opinions of their in-group or to increase identification with their in-group (Hohman and Hogg, 2015; Renkema et al., 2008; Routledge et al., 2013). In addition, American participants who received MS priming relative to those in the control condition contributed more money to charities supporting American projects but didn't increase donations to international projects (Jonas et al, 2002). While the behavioral findings indicate that MS profoundly influ-

While the behavioral findings indicate that MS profoundly influences group identification and in-group favoritism in helping behavior, the intermediate cognitive and neural mechanisms have been poorly understood. A recent research suggests that MS increases the perceived continuity of a social group and strengthens the perception of group entitativity, which, in turn, enhances in-group identification (Herrera and Sani, 2013). However, there is still a gap between in-group identification and in-group bias in helping behavior. It is widely accepted that empathy – the ability to understand and share others' emotional sates – is a proximate mechanism underlying helping or prosocial behavior (Batson et al., 1987; Batson, 2011; de Waal, 2008). Neuroimaging research has uncovered that the brain activity in response to others' suffering can predict individuals' intention to help others (e.g., Hein et al.,







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2010; Ma et al., 2011; Mathur et al., 2010; Luo et al., 2015). Therefore, it is likely that MS may modulate the cognitive and neural underpinnings of empathy so as to influence in-group favoritism in social behavior. The current research tested this hypothesis by examining racial in-group bias in empathic neural responses to racial in-group and out-group members' suffering.

Behavioral research has revealed that, when being asked to make judicial decisions, white university students reported greater feelings of empathy for a white than a black defendant and assigned more lenient punishments to the white defendant (Johnson et al., 2002). White participants also exhibited pro-white empathy bias to patients' pain expressions and showed remarkable pro-white bias in pain treatment (Drwecki et al., 2011). These behavioral observations provide evidence for racial in-group bias in empathy and racial in-group favoritism in motivation to help. The related neural underpinnings have been examined by recent functional magnetic resonance imaging (fMRI) studies of racial in-group bias in empathic neural responses. Xu et al. (2009) showed the first evidence that, in both Asian and Caucasian participants, the blood oxygen level dependent (BOLD) signal in the cingulate cortex was greater in response to perceived painful stimuli applied to racial in-group than to out-group members. The racial in-group bias in empathic responses was also evident in the anterior insula (Azevedo et al., 2013; Sheng et al., 2014), the medial prefrontal cortex (Mathur et al., 2010), and the sensorimotor cortex (Avenanti et al., 2010). Event related potential (ERP) studies also reported evidence for racial in-group bias in empathic neural responses (Sessa et al., 2014; Sheng and Han, 2012; Sheng et al., 2013). Relative to neutral expressions, pain expressions increased neural responses at 128-188 ms (P2 component) after stimulus onset over the frontal/central brain regions when participants categorized faces in terms of race (Sheng and Han, 2012). Moreover, the modulation of P2 amplitudes by pain vs. neutral expressions was more salient for racial in-group than racial out-group faces. When participants identified each observed individual's painful feelings or performed pain judgments on face stimuli, a long-latency component (i.e., P3) was observed over the central/parietal regions, of which, however, the amplitude to pain versus neural expressions did not show racial in-group bias (Contreras-Huerta et al., 2014; Sheng and Han, 2012).

The ERP and BOLD indices of differential neural responses to racial in-group and out-group individuals' suffering provide us useful measures of whether and how MS priming modulates the neural substrates underlying racial in-group bias in empathy. The MS effects on neural correlates of racial in-group bias may be manifested in increasing empathic neural responses to racial in-group member's suffering, decreasing empathic neural responses to racial out-group member's suffering, or both. ERP research has divided empathic neural responses into an early automatic component at 140-380 ms after sensory stimulation over the frontal/central regions and a late top-down controlled component after 380 ms over the central/parietal regions (Fan and Han, 2008; Han et al., 2008). In addition, painful expressions selectively modulated the early activity at 110-360 ms over fronto-central and centro-parietal regions, whereas painful contexts selectively modulated the late activity at 400–840 ms over the same regions (Sessa et al., 2014). It is thus interesting to examine whether MS effects on neural correlates of racial in-group bias occur either in the early automatic or the late top-down controlled process of empathy. In addition, because the activity in the cingulate cortex to others' pain showed racial in-group bias (Xu et al., 2009; Sheng et al., 2014) and MS priming relative to negative affect (NA) priming decreased the cingulate activity during perception of painful stimulations applied to others (Luo et al., 2014), we predicted that the MS effect on racial in-group bias in empathic neural responses mainly occurs in the cingulate cortex.

We conducted two experiments to test these predictions. Experiment 1 recorded ERPs to pain and neutral expressions of Asian and Caucasian faces from two groups of Chinese adults who had been primed with MS or NA, respectively. We examined whether MS compared to NA priming enhances the racial in-group bias in empathic neural responses and such effects, if any, occur during the early automatic or the late controlled processes of empathy. These were tested using the task of pain judgment on each face because ERP results in this task did not show racial in-group bias in our previous research (Sheng and Han, 2012). This allowed us to examine whether MS compared to NA priming would augment the racial in-group bias in empathic neural responses. Experiment 2 employed a similar design but recorded BOLD signals, using fMRI, in response to pain or neutral expressions of Asian and Caucasian faces from two groups of Chinese adults who had been primed with MS and NA, respectively. The results of Experiments 1 and 2 together allowed us to examine the neural underpinnings with both high temporal and spatial resolutions that mediate MS effects on racial in-group bias in empathy for others' suffering.

Materials and methods

Participants

Experiment 1 recruited 32 Chinese college students as paid volunteers. There were 16 participants in each group who received MS or NA priming. MS group consisted of 12 males and 4 females (mean age \pm SD = 22.38 \pm 2.70 yrs). NA group consisted of 10 males and 6 females (23.25 \pm 2.46 yrs). There was no significant difference in gender distribution ($\chi^2(1) = 0.58$, p > 0.1) and age (t(30) = -0.823, p > 0.1) between MS and NA groups. Experiment 2 recruited 40 Chinese college students as paid volunteers. There was 20 participants in each group who received MS or NA priming. MS group consisted of 11 males and 9 females (mean age \pm SD = 23.15 \pm 2.35 yrs); NA group consisted of 8 males and 12 females (mean age \pm SD = 21.95 \pm 2.09 yrs). There was no significant difference in gender distribution ($\chi^2(1) =$ 0.902, p > 0.05) and age (t (38) = 1.708, p > 0.05) between MS and NA groups. All participants were right-handed, had normal or corrected-to-normal vision, and reported no neurological history. Informed consents were obtained from all participants. This study was approved by a local ethics committee. The sample sizes in Experiments 1 and 2 were determined based on the previous studies (Sheng and Han, 2012; Xu et al., 2009; Sheng et al., 2014) that showed robust ERP and fMRI evidence of racial in-group bias in empathic neural responses using the same stimuli and procedure.

Stimuli and procedure

Stimuli used during EEG recording were adopted from our previous research (Sheng and Han, 2012) and consisted of 64 digital photographs of faces from 16 Chinese models (8 males) and 16 Caucasian models (8 males). Each model contributed two photographs, one with neutral and one with pain expressions. Asian and Caucasian faces were matched in perceptual features (e.g., luminance), emotional intensity and social features (e.g., attractiveness) (Sheng and Han, 2012).

Materials used for MS and NA priming were adopted from our previous work (Luo et al, 2014) and consisted of 28 statements for each priming procedure. Each statement was displayed for 7 s on a computer monitor and participants had to judge whether he/she agreed with the statement by pressing of two keys. Statements used for MS priming were related to death (e.g. "I won't feel terrible even if I would die lonely", "My body would rot after death"). Statements used for NA priming were not related to death but referred to negative emotions such as fear (e.g. "I am not frightened about life at all") and anxiety (e.g., "The coming exam makes me uneasy"). After the priming procedure, participants were asked to perform 40 mathematical calculations in 5 min, which served as a delay between priming and critical dependent measures so that death-related thoughts faded away from consciousness. Participants had to judge whether each calculation would give an odd or even number by mental arithmetic and press a corresponding button. Each calculation lasted for 7 s and two consecutive calculations were

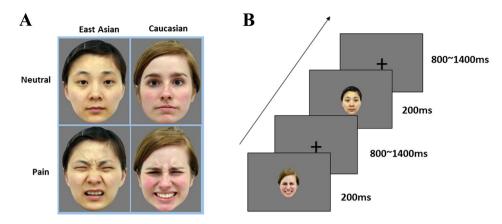


Fig. 1. (A) Illustration of Asian and Caucasian faces with pain or neutral expressions used in Experiment 1. (B) Illustration of the experimental procedure in Experiment 1.

intervened with 0.5 s. The priming and calculation tasks were finished before EEG recording or fMRI scanning.

During the electroencephalography (EEG) recordings in Experiment 1, each photograph was presented in the center of a gray background on a 21-inch color monitor, subtending a visual angle of $3.8^{\circ} \times 4.7^{\circ}$ at a viewing distance of 120 cm. Each trial consisted of a face stimulus with a duration of 200 ms, which was followed by a fixation cross with a duration varying randomly between 800 and 1400 ms (Fig. 1). Each participant finished 4 blocks of 128 trials (each of the 64 photographs was presented twice in a random order in each block) during which they performed judgments on facial expression (pain vs. neutral) of each stimulus.

During fMRI scanning in Experiment 2, stimuli were presented through an LCD projector onto a rear projection screen, which were viewed with an angled mirror positioned on the head-coil. Each photo was presented at the center of a gray background, subtending a visual angle of $4.0^\circ \times 5.0^\circ$ at a viewing distance of 100 cm. On each trial an Asian or Caucasian face with pain or neutral expression was presented with a duration of 2 s, which was followed by a cross fixation with a duration of 2, 4, 6, or 8 s. In an event-related design participants were instructed to identify facial expression of each face (pain vs. neutral) by a button press using the right index and middle fingers. In order to increase perceptual duration of each stimulus, participants were instructed to respond after the stimuli had disappeared. Each participant conducted 4 functional scans. Each functional scan started with a 4 s prompt screen with an instruction followed by 32 trials. The 64 pictures of faces were presented in a random order in every two functional scans.

To assess participants' feelings of closeness to death and fear of death during the priming procedure, after EEG recording in Experiment 1 and fMRI scanning in Experiment 2, participants were asked to rate feelings about the priming task (e.g. How close do you feel to death after reading all the sentences and making your judgments?, How unpleasant do you feel after reading all the sentences and making your judgments?, How fearful do you feel about death after reading all the sentences and making your judgments?). A Likert-type scale was used for all ratings where 0 indicated no effect and 10 indicated maximal effect (e.g. extremely close, extremely unpleasant, or extremely fearful). Individual's negative affect related to death was assessed using the Death Depression Scale (Templer et al., 1990). Individuals' trait empathic ability was measured using the Interpersonal Reactivity Index (IRI, Davis, 1983). The 28-item IRI is a self-report measure consisting of four 7-item subscales, each item was answered on a 5-point Likert scale ranging from "doesn't describe me well" to "describe me very well". The Perspective-Taking (PT) scale assesses the tendency to spontaneously adopt the psychological point of view of others. The Fantasy (FS) scale taps respondents' tendencies to transpose themselves imaginatively into the feelings and actions of fictitious characters in books, movies, and plays. The Empathic Concern (EC) scale assesses "other-oriented" feelings of sympathy and concern for unfortunate others. The Personal Distress (PD) scale measures "self-oriented" feelings of personal anxiety.

ERP data recording and analysis

The NeuroScan system was used for EEG acquisition in Experiment 1. EEG was continuously recorded from 62 scalp electrodes that were mounted on an elastic cap in accordance with the extended 10-20 system and were referenced to the left and right mastoid electrodes that were physically linked. The electrode impedance was kept less than 5 k Ω . Eye blinks and vertical eye movements were monitored with electrodes located above and below the left eye. The horizontal electro-oculogram was recorded from electrodes placed 1.5 cm lateral to the left and right external canthi. The EEG was amplified (band pass 0.1-100 Hz) and digitized at a sampling rate of 250 Hz. The ERPs in each condition were averaged separately off-line with an epoch beginning 200 ms before stimulus onset and continuing for 1200 ms. Trials contaminated by eye blinks, eye moments, muscle potentials exceeding \pm 50 µV at any electrode, or response errors were excluded from the average. 14.8% and 16.9% of the trials were excluded due to artifacts in MS and NA groups, respectively. The baseline for ERP measurements was the mean voltage of a 200 ms pre-stimulus interval and the latency was measured relative to the stimulus onset. Data analyses were conducted at electrodes selected from frontal/central (Fz, FCz, F1, F2, F3, F4, FC1, FC2 FC3, FC4), central/parietal (Cz, CPz, C1, C2, C3, C4, CP1, CP2, CP3, CP4), and parietal (Pz, P1, P2, P3, P4) regions. Reaction times (RTs) shorter than 100 ms and longer than 800 ms were excluded from analyses. Behavioral performances and ERPs were subjected to repeated-measure analyses of variance (ANOVAs) with Expression (pain vs. neutral) and Race (Asian vs. Caucasian) as within-subjects variables and Group (MS priming vs. NA priming) as a between-subjects variable.

Both voltage topography and the standardized Low Resolution Brain Electromagnetic Tomography (sLORETA) (Pascual-Marqui, 2002) were used to estimate potential sources of empathic neural responses. sLORETA is a linear method of computing statistical maps from EEG data that reveals locations of the underlying source processes and does not require a priori hypotheses regarding the field distribution of the active sources. We performed the analysis using sLORETA to assess the 3D current source of neural activity that differentiated between ERPs to pain and neutral expressions of Asian vs. Caucasian faces. A boundary element model was first created with about 5000 nodes from a realistic head model. Statistical nonparametric mapping was calculated in a specific time window to estimate the source that differentiated ERPs to pain and neutral expressions. The log of the F ratio of averages was used and considered with a 0.95 level of significance.

fMRI data acquisition and analysis

Imaging data that covered the whole brain were acquired using a 3-T GE Signa MR750 scanner (GE Healthcare; Waukesha, WI) with a standard head coil. Head motion was minimized using foam padding. Anatomical images were obtained using a standard 3D T1-weighted sequence ($512 \times 512 \times 180$ matrix with $0.47 \times 0.47 \times 1.0$ mm³ spatial resolution, TR = 8.204 ms, TE = 3.22 ms, flip angle = 12°). Functional images were acquired using T2-weighted, gradient-echo echo-planar imaging (EPI) sequences sensitive to BOLD contrast ($64 \times 64 \times 32$ matrix with $3.75 \times 3.75 \times 5$ mm³ spatial resolution, repetition time (TR) = 2000 ms, echo time (TE) = 30 ms, flip angle = 90° , field of view (FOV) = 24×24 cm).

The fMRI data were analyzed using SPM8 (the Wellcome Trust Centre for Neuroimaging, London, United Kingdom). The functional images were corrected for differences in acquisition time between slices for each whole-brain volume and realigned within and across runs to correct for head movement. Six movement parameters (translation: x, y, z and rotation: pitch, roll, yaw) were included in the statistical model. The anatomical image was co-registered with the mean functional image produced during the process of realignment. All images were normalized to a $3 \times 3 \times 3$ mm³ Montreal Neurological Institute (MNI) template. Functional images were spatially smoothed using a Gaussian filter with the full-width/half-maximum parameter (FWHM) set to 8 mm. Whole brain statistical parametric mapping analyses were conducted to examine any brain area that showed increased activity to pain vs. neutral expressions or modulations of the activity to pain vs. neutral expression by MS/NA priming. Effects at each voxel were estimated and regionally specific effects were compared using linear contrasts in individual participants using a fixed effect analysis. The contrast value of pain vs. neutral expressions was calculated to define pain specific neutral activations. ANOVAs of the contrast value with Race (Asian vs. Caucasian) as a within-subjects variable and Group (MS priming vs. NA priming) as a between-subjects variable were conducted to examine the priming effect on racial in-group bias in empathic neural responses. Racial in-group bias in empathic neural responses was also examined separately for the MS and NA groups by calculating the interaction of Expression (pain vs. neutral) and Race (Asian vs. Caucasian). Random effect analyses were conducted across each participant group based on statistical parameter maps from each individual participant to allow population inference. Significant activations were identified using a threshold of p < 0.05 corrected for multiple comparisons based on a combined voxelwise and cluster-size threshold (p < 0.05, k = 32) derived by Monte Carlo simulation based upon the whole brain gray matter search volume and an estimate of the data set spatial correlation based upon the GLM residual images.

Results

Experiment 1

Behavioral results

Behavioral performances during EEG recording are shown in Table 1. Response accuracies were high (87.3%) and ANOVAs of responses accuracy did not show any significant effect. ANOVAs of RTs showed significant main effects of Race (F(1,31) = 15.81, p < 0.001, $\eta^2 = 0.345$) and

Table 1

Behavioral performances during EEG recording (mean \pm SD) in Experiment 1.

	Group	Caucasian face		Asian face	
		Neutral	Pain	Neutral	Pain
RT (ms)	MS	525 ± 63	507 ± 67	510 ± 67	507 ± 64
	NA	500 ± 67	478 ± 54	491 ± 64	472 ± 59
Accuracy (%)	MS	88.4 ± 6.5	88.2 ± 9.5	90.2 ± 7.8	88.3 ± 7.5
	NA	88.0 ± 6.2	87.7 ± 9.1	89.9 ± 7.6	87.0 ± 8.4

Table	2
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Rating scores (mean \pm SD) in Experiment 1.

		MS group	NA group
IRI			
	Perspective taking	16.81 ± 5.84	18.07 ± 3.33
	Fantasy	19.13 ± 4.94	17.67 ± 4.91
	Empathic	18.88 ± 3.65	19.40 ± 2.47
	concern		
	Personal distress	15.25 ± 4.71	14.27 ± 3.77
Death depi	ression	50.56 ± 17.18	52.81 ± 13.11

ps > 0.1.

Expression (F(1,31) = 15.06, p < 0.001, η^2 = 0.334), as RTs were longer to Caucasian than to Asian faces and longer to neutral than to painful faces, similar to the previous results (Sheng and Han, 2012). The effects of Race and Expression did not differ between MS and NA groups (F(1, 31) = 0.017 and 1.691, ps > 0.05). Independent sample t-test did not find significant differences in IRI rating scores and death depression rating scores between MS and NA groups (ps > 0.1, see Table 2). Independent sample t-test confirmed higher rating scores of closeness to death in MS group than in NA groups (5.22 ± 3.37 vs. 2.22 ± 2.94, t(30) = 2.64, p < 0.05). MS and NA groups did not differ in rating scores of fear of death (2.53 ± 2.20 vs. 1.06 ± 2.23, t(30) = 1.88, p > 0.1) and unpleasantness (4.16 ± 3.43 vs. 4.13 ± 2.46, t(30) = 0.03, p > 0.1).

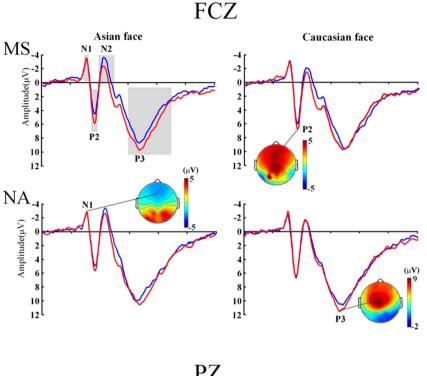
ERP results

The ERPs to faces were characterized by a negative wave at 84– 116 ms (N1) and a positive deflection at 128–188 ms (P2) over the frontal and central regions. These were followed by a negative wave at 200– 300 ms (N2) over the frontal region and a long-latency positivity at 400–700 ms (P3) over the central and parietal regions (Fig. 2). Face stimuli also elicited a posterior positivity at 88–148 ms (P1) and negativity at 140–180 ms (N170) over the occipital and temporal regions.

The ANOVAs of the N1 amplitudes at 84–116 ms did not show any significant effect (ps > 0.05). The ANOVAs of the P2 amplitude at 128–188 ms over the frontal/central electrodes showed a significant main effect of Race (F(1,31) = 22.68–48.52, ps < 0.001, see Table S1 for details of the statistical analyses) as Caucasian faces elicited greater amplitude than Asian faces. There was a significant main effect of Expression (F(1,31) = 6.5–12.41, ps < 0.05), indicating larger P2 amplitude in response to pain than neutral expressions. In addition, there was a significant interaction of Race × Expression (F(1,31) = 4.23–5.91, ps < 0.05) as the modulation of the P2 amplitude was stronger for Asian than for Caucasian faces, indicating racial in-group bias in P2 amplitude to others' pain. However, neither the main effect of Group nor its interaction with other factors was significant (ps > 0.5), suggesting similar in-group bias in P2 responses in individuals who received MS and NA priming.

The ANOVAs of the N2 component at 200–300 ms showed a significant main effect of Race (F(1,31) = 16.35-26.79, ps < 0.001, see Table S2 for details) due to that Asian compared to Caucasian faces elicited larger N2 amplitude. There was also a significant main effect of Expression (F(1,31) = 11.62-34.13, ps < 0.005) as pain relative to neutral expressions induced a positive shift of the N2 amplitude. However, the interaction of Race × Expression did not reach significance (F(1,47) = 0.70-2.64, ps > 0.1). The effect of Group and its interaction with other variables on the N2 amplitude was not significant (ps > 0.05).

The ANOVAs of the P3 amplitude at 400–700 ms over the central/parietal electrodes showed a significant main effect of Race (F(1,31) = 7.47–25.29, ps < 0.05, see Table S3 for details) as Caucasian compared to Asian faces elicited larger P3 amplitude. Interestingly, there was a significant three-way interaction of Race × Expression × Group (F(1,31) = 4.18–10.42, ps < 0.05). Separate analyses revealed a significant interaction of Race × Expression (F(1,15) = 7.13–23.08, ps < 0.05) for MS group because the modulation of the P3 amplitude was stronger for





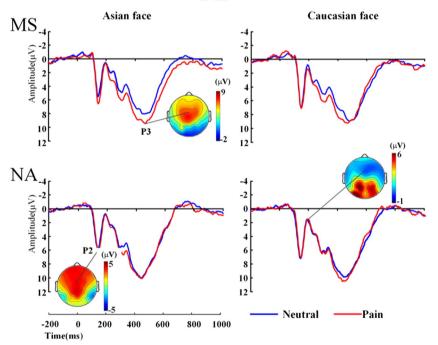


Fig. 2. Illustration of ERPs at electrodes FCZ and PZ in response to pain and neutral expressions of Asian and Caucasian faces in Experiment 1. ERPs are plotted separately for MS and NA groups. The gray areas indicated the time windows for calculating P2, N2 and P3 amplitudes. Voltage topographies illustrate the scalp distribution of the maximum amplitude of each ERP component.

Asian than Caucasian faces (Fig. 3A). However, the interaction of Race \times Expression was not significant for NA group (F(1,15) = 0.00-0.85, ps > 0.5). Therefore, relative to NA group, MS showed stronger racial in-group bias in P3 amplitudes to perceived pain in others and, as illustrated in Fig. 3, these effects occurred due to that MS priming compared to NA priming increased the P3 amplitude to pain expression of Asian faces. We further conducted sLORETA to estimate the sources of the racial in-group bias in empathic neural responses in the P3 time

window in MS group. This revealed two potential sources in the midcingulate cortex and the left anterior insula (Figs. 3B and C).

To estimate the relationship between trait empathy ability and racial in-group bias in empathic neural responses, we first subtracted the differential amplitude to pain vs. neutral expressions of Caucasian faces from the differential amplitude to pain vs. neutral expressions of Asian faces. We then examined the correlation of this measure and IRI scores. This analysis revealed a significant negative correlation between racial

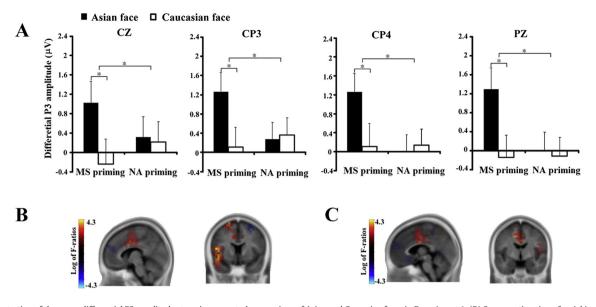


Fig. 3. (A) Illustration of the mean differential P3 amplitudes to pain vs. neutral expressions of Asian and Caucasian faces in Experiment 1. (B) Source estimation of racial in-group bias in empathic neural responses at 436 ms after stimulus onset suggested two sources in the mid-cingulate (peak MNI coordinates: -7/10/35) and left anterior cortex (peak MNI coordinates: -45/5/-2). (C) Source estimation of racial in-group bias in empathic neural responses at 456 ms after stimulus onset suggested a source in the mid-cingulate (peak MNI coordinates: -3/-5/30).

in-group bias in the P3 amplitude and the ability of perspective taking in MS group (r = -.36 to -0.42, ps < 0.05, Fig. 4, see Table S4 for details) but not in NA group (ps > 0.05), suggesting that individuals with better perspective taking ability showed weaker racial in-group bias in P3 amplitude after MS priming.

Experiment 2

Behavioral results

Behavioral performances during fMRI scanning are shown in Table 3. Response accuracies were high (>95.9%). RTs were slow because participants were asked to respond after the offset of stimuli. ANOVAs of RTs showed a significant interaction of Expression × Group (F(1,39) = 6.083, p < 0.05). Separate analyses revealed a significant effect of Expression in NA group (F(1,39) = 4.907, p < 0.05) but not in MS group (F(1,39) = 1.238, p > 0.1). RTs were slightly longer for neutral than pain expressions in the NA group. Independent sample t-test did not find significant differences in IRI rating scores and death depression rating scores between MS and NA groups (ps > 0.1, Table 4). Independent sample t-test confirmed higher rating scores of closeness to death (5.25 ± 2.94 vs. 2.10 ± 2.22 , t(38) = 3.826, p < 0.001) in MS group than in NA group. MS and NA groups did not differ in rating scores of fear of death (3.175 ± 2.806 vs. 2.075 ± 2.386 , t(38) = 1.336, p > 0.1) and unpleasantness (3.625 ± 2.635 vs. 3.475 ± 2.441 , t(38) = 0.187, p > 0.1).

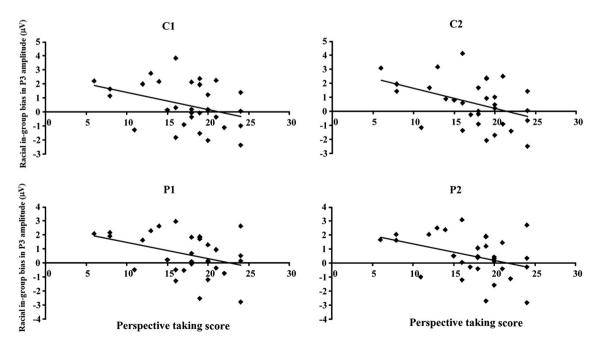


Fig. 4. Illustration of the correlation between self-reported perspective taking score and the racial in-group bias in the P3 amplitude at electrodes C1–C2 and P1–P2 in MS group in Experiment 1.

Table 3
Behavioral performances during fMRI scanning (mean $+$ SD) in Experiment 2.

	Group	Asian face		Caucasian face	
		Neutral	Pain	Neutral	Pain
RT (ms)	MS	2552 ± 350	2582 ± 339	2568 ± 337	2576 ± 348
	NA	2731 ± 249	2691 ± 318	2769 ± 320	2687 ± 269
Accuracy (%)	MS	98.1 ± 4.5	97.8 ± 6.3	96.7 ± 7.1	97.8 ± 4.7
	NA	97.7 ± 4.5	98.6 ± 2.6	98.0 ± 2.9	95.9 ± 6.7

fMRI results

We first examined the priming effect on racial in-group bias in empathic neural responses by calculating a whole-brain ANOVA of the contrast of pain vs. neutral expression with Race (Asian vs. Caucasian) as a within-subjects variable and Group (MS priming vs. NA priming) as a between-subjects variable. This analysis revealed significant activations in the anterior and mid-cingulate cortex (-6/20/28, k = 80, Z = 3.11) and the right precentral gyrus (54/2)37, k = 38, Z = 3.75, Fig. 5A). We then conducted whole-brain interaction analyses of the contrast of (pain-neutral)_{Asian faces} vs. (pain-neutral)_{Caucasian faces} in MS and NA groups, respectively. This contrast revealed significant activations in the dorsal portion of the anterior cingulate cortex that extended into the mid-cingulate cortex (-6/20/40, k = 38, z = 3.33) and in the right precentral gyrus (57/-1/34, k = 78, Z = 3.56, Fig. 5B) in MS group. NA group, however, showed a significant activation in the right inferior parietal cortex (36/-73/43, k = 55, Z = 3.35). These results suggest that, relative to NA priming, MS priming significantly enhanced the racial bias in empathic neural response in the cingulate cortex and the right precentral gyrus.

To further examine how MS vs. NA priming modulated empathic neural response to pain versus neutral expressions of Asian and Caucasian faces, we conducted separate whole-brain interaction analyses of the contrast of (pain-neutral)_{MS} priming vs. (pain-neutral)_{NA} priming for Asian and Caucasian faces. The results indicated that MS vs. NA priming significantly increased activity in the right posterior temporal cortex (57/-61/4, k = 103, Z = 4.00) and the mid-cingulate cortex (3/32/25, K = 28, Z = 3.47, Fig. 5C) in response to pain expression of Asian faces. In contrast, MS vs. NA priming increased the activity in the right posterior temporal cortex (60/-58/4, k = 139, Z = 3.96) but decreased the activity in the right precentral gyrus (57/-4/37, k = 84, Z = 3.45) in response to pain vs. neutral expression of Caucasian faces.

Finally, to examine the main effect of facial expression, we calculated the contrast of pain vs. neutral expressions by combining Asian and Caucasian faces. This analysis revealed significant activations in the bilateral occipital and temporal cortices (right hemisphere: 51/-64/-8, k = 1018, Z = 5.06; left hemisphere: -24/-94/-5, k = 801, Z = 4.65, Fig. 5D).

Table 4
Rating scores (mean \pm SD) in Experiment 2.

		MS group	NA group
IRI			
	Perspective taking	17.55 ± 4.26	18.65 ± 3.47
	Fantasy	16.35 ± 5.07	17.65 ± 3.54
	Empathic concern	18.10 ± 4.25	19.25 ± 3.39
	Personal distress	14.50 ± 4.32	15.15 ± 2.56
Death depression		51.40 ± 13.95	57.75 ± 14.64

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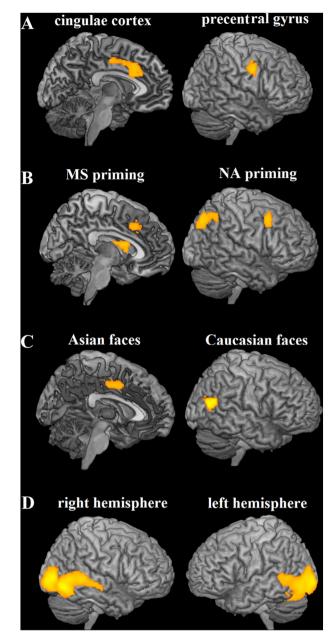


Fig. 5. Illustration of fMRI results in Experiment 2. (A) Significant interactions of MS/NA priming × Asian/Caucasian faces on the contrast of pain vs. neutral expressions were identified in the cingulate and right precentral gyrus. (B) An enhanced activation to pain (vs. neutral) expressions of Asian compared to Caucasian faces was identified in the cingulate cortex after MS priming. The same contrast only showed a significant activation in the right inferior parietal cortex after NA priming. (C) MS compared to NA priming induced a cingulate activation to pain (vs. neutral) expressions of Asian faces. (D) The main effect of facial expression (pain vs. neutral) independent of race and priming was evident in the bilateral occipital and temporal cortices.

Discussion

The current work tested the hypothesis that reminding mortality modulates racial in-group bias in empathy for others' suffering. Specifically, we predicted that reminding people of death relative to deathunrelated negative affect such as anxiety increases racial in-group bias in empathic neural responses to perceived pain in others. This prediction was verified by measuring brain activity using both ERP and fMRI that allowed us to examine the priming effect on neural responses to pain vs. neutral expressions with both high temporal and spatial resolution. Our manipulation check showed that, relative to NA priming, MS priming induced stronger feelings of closeness to death in our participants, indicating enhancement of death awareness in MS compared to NA groups.

In Experiment 1 where participants were asked to respond as fast as possible, their RTs were longer to Caucasian than to Asian faces during pain judgments. This result is consistent with the posit that, relative to same-race faces, other-race faces are perceived as more psychologically similar to each other (Valentine and Endo, 1992; Vizioli et al., 2010) and with less reference to an individual's personal situation (Kinder and Sears, 1981; Sheng and Han, 2012). RTs were faster to pain compared to neutral expressions possibly due to greater perceptual salience of or task-related top-down attention to pain expression. In addition, this effect was stronger for Caucasian than Asian faces under the MS priming condition. The differential RTs to Caucasian and Asian faces were mainly manifested in that MS priming compared to NA priming specifically speeded responses to neutral expression of Asian relative to Caucasian faces (see Table 1), suggesting non-specific effects on perceived salience of neutral expression of Asian faces.

Our ERP results in Experiment 1 showed evidence that MS compared to NA priming significantly modulated the racial in-group bias in the P3 amplitudes at 400-700 ms in responses to others' suffering. Specifically, the empathic neural responses in the P3 time window did not show racial in-group bias in the NA group, whereas the MS group displayed significant racial in-group bias in the P3 amplitude in response to others' pain. The effect of MS priming was particularly prominent in increasing the P3 amplitude to pain (vs. neutral) expression of racial in-group faces. The absence of racial in-group bias in the P3 amplitude in the NA condition replicated the results of our previous work (Sheng and Han, 2012), which showed that racial in-group bias in the P3 amplitudes was evident when participants performed judgments on race identification but not on pain vs. neutral expressions. It was assumed that the pain judgment task demanded focus on each individual' personal feeling and thus reduced racial in-group bias in empathy by increasing neural responses to racial out-group individuals' pain (Sheng and Han, 2012). The P3 results indicate that MS compared to NA priming facilitated empathy for racial in-group members' suffering and, as a consequence, increases racial in-group bias in empathy even when participants were asked to focus on each individual's pain feeling.

Pain relative to neutral expressions significantly increased the P2 amplitudes over the frontal/central electrodes. The N2 amplitude was also positively shifted by pain compared to neutral expressions. The modulations of the P2 amplitudes by pain vs. neutral expressions were more prominent for Asian than Caucasian faces. These results replicate the previous ERP findings of modulations of brain activity by pain expression (Sheng and Han, 2012; Sheng et al., 2013; Huang and Han, 2014; Sessa et al., 2014), but were different from Sheng and Han's (2012) observation that the P2 and the N2 amplitude to pain expression did not illustrate racial in-group bias during judgments on pain vs. neutral expressions. This is not surprising because participants in Sheng and Han (2012) had not received any priming before EEG recording whereas participants in the current study received MS or NA priming that seemed to produce a similar effect in enhancing racial in-group bias in empathic neural responses. Most relevant to the hypothesis tested in the current work, we showed that the racial in-group bias in the P2 amplitudes did not differ significantly between MS and NA groups, suggesting the absence of MS effects on racial in-group bias in empathic neural responses in the P2 time window.

Experiment 2 reinforced the results of Experiment 1 by showing fMRI evidence that MS relative to NA priming magnified racial ingroup bias in empathic neural responses. Specifically, MS compared to NA priming increased racial in-group bias in BOLD signals in the anterior and mid-cingulate cortex in response to perceived pain expressions. Moreover, our post hoc analyses revealed that MS vs. NA priming mainly increased the cingulate activity associated with pain expression of racial in-group faces but produced little effect on the activity in the same brain region in response to pain expressions of racial out-group faces. Thus the modulation of racial in-group bias in neural responses to pain expression by MS vs. NA priming occurred in the same brain region that showed racial in-group bias in neural responses to perceived painful stimuli applied to others (Xu et al., 2009) and racial in-group bias in neural responses modulated by task demands (Sheng et al., 2014). The effect of MS priming on BOLD signals associated with racial ingroup bias in empathy is in line with the effect of MS priming on racial in-group bias in the P3 amplitude in Experiment 1 and our ERP source estimation in Experiment 1 is consistent with the fMRI results in Experiment 2. MS compared to NA priming increased the activity in the right posterior temporal cortex related to pain expression of Caucasian faces. Similar MS effects were observed in our previous work where MS relative to NA priming increased the activity in the posterior temporal cortex in responses to others' pain (Luo et al., 2014). The superior temporal sulcus and adjacent occipital gyrus respond to changes in different facial expressions such as fear and anger (Andersen et al., 2001; Harris et al., 2012; De Winter et al., 2015) and are associated with early perceptual processing of facial expressions. Thus our fMRI results identified that the most salient MS effect on racial in-group bias in empathy occurred in the cingulate cortex but not in the brain regions involved in perceptual processing of facial expressions. Taken together, our ERP and fMRI findings demonstrate that reminding people of mortality enhances racial in-group bias in empathy in the cingulate cortex and this effect was produced by increasing empathic neural responses to racial ingroup members' pain.

Our brain imaging results have several implications. First, racial ingroup bias in empathy occurred during the early stage of neural responses to perceived pain expression regardless of MS or NA priming applied to participants. Therefore, the racial in-group bias at the early stage of empathy was not affected by negative mood elicited by reminding either mortality or negative daily-life events. The previous ERP research has shown that the early empathic neural responses in the P2 and N2 time windows were associated with emotional sharing (Fan and Han, 2008) and the empathic neural responses to racial ingroup members' pain in these time windows were not modulated by task demands that emphasized top-down attention to emotional cues in visual stimuli. The previous and current ERP results illustrate a robust effect of racial intergroup relationships between an observer and a target on empathy (Sessa et al., 2014; Sheng and Han, 2012; Sheng et al., 2013). The current finding that racial in-group bias in early empathic neural responses in the P2/N2 time windows was independent of individuals' mood states further suggests that social intergroup relationships dominate task demands and affective states in modulation of early emotional sharing during empathy for pain.

Second, as empathic neural responses in the P3 time window have been associated with cognitive evaluation of others' pain (Fan and Han, 2008), the current ERP results suggest that MS compared to NA priming can increase racial in-group bias in empathy by enhancing evaluation of racial in-group members' pain. Thus MS priming enhanced group identification and affiliation in terms of racial relationships, which mainly facilitated late cognitive components of empathy for racial in-group members' pain. However, this effect was reduced in those with better perspective taking ability, who intended not to differentiate between racial in-group and out-group members' suffering. Our fMRI results in Experiment 2 further reinforced the proposal that MS compared to NA priming modulates the cognitive components of empathic processes of racial in-group individuals' pain. Ample fMRI studies of empathy have identified a core neural network consisting of the anterior/mid-cingulate, supplementary motor areas, and bilateral anterior insula (Fan et al., 2011; Lamm et al., 2011). Among these brain regions the anterior insula and anterior cingulate are frequently engaged in the affective-perceptual forms of empathy whereas the midcingulate cortex is frequently activated in cognitive-evaluative forms of empathy (Fan et al., 2011). Therefore, our findings of MS effects on the racial in-group bias in the mid-cingulate activity complemented the MS effects on the racial in-group bias in the P3 amplitude and

these brain imaging results jointly demonstrate a key role of MS priming in modulation of the cognitive component of empathy for racial ingroup members' suffering.

Interestingly, MS compared to NA priming activated the right precentral gyrus during perceiving pain vs. neutral expression of Asian faces but decreased the activity in the right precentral gyrus in response to pain vs. neutral expression of Caucasian faces. The precentral gyrus showed enhanced activity not only during motor execution but also during attention to action (Binkofski et al., 2002) and motor preparation (Kawashima et al., 1994; Simon et al., 2002). This brain region was likewise engaged during imagining motor acts and the activity in the region was associated with accuracy of imagery task performances (Hanakawa et al., 2003). Therefore, besides promoting helping behavior toward racial in-group members (e.g., Johnson et al., 2002; Drwecki et al., 2011), MS vs. NA priming might also facilitate motor preparation that helps to link enhanced empathy with helping behavior. The mid-cingulate cortex may also contribute to this function because this brain region was recruited when participants performed target detection followed the presentation of unpleasant or neutral pictures (Pereira et al., 2010).

Finally, our brain imaging findings help to get a deeper understanding of the mechanism by which MS influences in-group favoritism in social behavior. MS priming led to more altruistic behavior toward racial in-group members (Jonas et al, 2002) or stronger identification of in-group members' opinions (Renkema et al., 2008). These can be explained by assuming that empathy mediates the MS effects on behavior or attitude toward in-group members because empathy is a proximate mechanism of prosocial behavior (Batson, 2011). Our ERP and fMRI results suggest that the increased in-group favoritism following MS priming may be augmented by improving cognitive evaluation of in-group members' painful feelings rather than by increasing emotional sharing with in-group members. Empathy is an evolved function that has been observed in both human and nonhuman social mammals (Preston and de Waal, 2002). The perception-action-model of empathy assumes that, in comparison to the automatic and emotional aspects of empathic representation, cognitive empathic processing appears to be differentially available across species and demonstrated more complexity and flexibility (Preston and de Waal, 2002). Witnessing another person experiencing pain may have different meanings depending on social contexts. Viewing an in-group member's pain may cause empathy, sympathy and caregiving, whereas viewing an out-group member in pain may signal alarm and fear that are related to personal safety. In agreement with the perception-action-model of empathy (Preston and de Waal, 2002) and TMT (Greenberg et al., 1986; Pyszczynski and Greenberg, 1999), our brain results provide neuroimaging evidence that cognitive empathic processes may be more susceptible to the thoughts of death in humans. Recent brain imaging findings indicated that death-related thoughts decreased the activity in the anterior insula (Han et al., 2010; Klackl et al., 2014; Shi and Han, 2013), which is linked mainly to the affective-perceptual forms of empathy (Fan et al., 2011). Together, these brain imaging findings illustrate the dynamic social function of empathy and unveil the underlying neural mechanisms.

A limitation of our EEG recording should be noted. Our EEG signals were recorded in reference to the linked mastoids. This method for EEG acquiring might induce distortion of ERPs if the impedances between the two mastoid electrodes are mismatched relative to one another (Pivik et al., 1993) and give rise to false lateralization of ERP amplitudes. Our ERP analyses focused on the comparison between neural activities in response to pain versus neutral expressions, between empathic neural responses to same-race and other-race faces and between racial in-group bias in empathic neural responses in MS and NA groups. These analyses compared ERP amplitudes recorded at the same electrode over the left or right hemispheres and reduced the influences of the linked-mastoid reference to a minimum degree. We did not find any effect of hemisphere lateralization on ERP amplitudes that might be contaminated by the linked-mastoid reference. Future research should consider the choice of the reference electrode if

investigating hemisphere lateralization of racial in-group bias in empathic neural responses.

In conclusion, our ERP and fMRI findings lend support to the hypothesis that MS enhances in-group bias in empathy for others' pain, though the MS effect on empathy was evident mainly during the late stage of empathic neural responses and in the anterior and mid-cingulate cortex. The MS effect on in-group bias in empathic neural responses suggests an intermediate neural mechanism of variation of in-group favoritism under mortality threat. However, our study focused on MS effects on in-group bias in empathy when intergroup relationship was defined by race. It remains unknown whether the MS effect on ingroup bias in empathy can go beyond race-based in-group/out-group relationships. Future research should seek direct evidence that ingroup bias in empathy mediates MS priming and changes of in-group favoritism in helping behavior.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx. doi.org/10.1016/j.neuroimage.2015.06.023.

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