Acupuncture (from the Latin acus, “needle,” and pungere, “to prick”) is a technique of inserting and manipulating fine needles at specific points (acupoints) on the body. Many neuroimaging studies have revealed that acupuncture stimulation modulates the central nervous system in humans [1,3,11]. Subsequent functional magnetic resonance imaging (fMRI) investigations regarding several acupoints have demonstrated a correlation between acupuncture stimulation and corresponding cortical response, suggesting that acupuncture at disease-implicated acupoints modulates the pain network, including the hypothalamus and limbic system [5,16,21]. However, many of the studies were not performed with appropriate control groups, so whether changes were genuinely acupuncture-specific or a general phenomenon that could be elicited as a placebo response or by non-specific effects of mechanical stimulation has not been clearly demonstrated.

To provide an effective and credible placebo (defined as a physiologically inert procedure), the control must be convincing and should mimic, apart from the physiological effect, all aspects of the real active treatment. As a newly developed placebo sham needle is pushed against the skin, it causes a pricking sensation, but as increased pressure is applied, the shaft of the needle disappears into the handle, mimicking a ‘stage dagger’ [17,19]. Although this ‘placebo’ needle has been used in clinical trials, few studies have examined the specific effects of verum acupuncture compared to non-penetrating placebo acupuncture. Interfering factors, such as pain or emotion during acupuncture, are considered to contribute, at least in part, to the pattern of brain activity in the field of neuroimaging studies. To provide specific patterns implicated with the therapeutic effect of acupuncture, it is necessary to subtract fMRI brain images evoked by stimulating with sham acupuncture from those evoked by stimulating with verum acupuncture.

The present study was thus performed using fMRI to examine changes in blood oxygen level-dependent (BOLD) responses with verum acupuncture at acupoint LR2, as compared with the response to sham acupuncture.
This study was carried out in 10 healthy, right-handed subjects (6 men, 4 women; age range 20–34 years). After screening, those with psychiatric or neurological disorders were excluded. They were told that they would experience acupuncture stimulation, but they were not told whether it would be verum or sham acupuncture. All participants received a detailed explanation of the study and written, informed consent was provided. This study was conducted in accordance with the guidelines of the human subjects committee of Kyunghee University, Seoul, Republic of Korea.

Acupuncture was performed by an experienced Korean medical doctor. Each subject received verum or sham acupuncture in random order at acupoint LR2 between the first and second toes on the dorsum of the foot (Fig. 1). The verum acupuncture needle (sterile, disposable, stainless steel, 40 mm long, and 0.25 mm in diameter) was inserted to a depth of 0.8 cm perpendicular to the skin surface. The needle was rotated manually, clockwise and counterclockwise, once per second (1 Hz) for 60 s (ON phase). The needle was then withdrawn for 120 s (OFF phase). The sham procedure was performed using a blunt, telescopic, non-penetrating Park Sham needle (DongBang Acupuncture Inc., Boryeoung, Korea) in an attempt to exclude the placebo phenomenon[17].

After each session, subjects were questioned using the short-form of the needle sensation questionnaire, leading to ratings of both deqi and acute pain. The deqi scale consists of sore, numb, heavy, and distended sensations, and the acute pain scale consists of hurting and aching sensations. Participants rated the intensity of each sensation using a 6-point Likert scale. They also answered the question “Did you think the acupuncture stimulation you just received was real or sham?”

Subjects were asked to lie in a supine position on the scanner bed with their eyes closed. fMRI scans were conducted over two 9-min periods; one scan involved verum acupuncture and the other scan involved sham acupuncture. The total scan time for each session was 9 min (Fig. 1). Scanning commenced with a rest period (2 min) and was followed by three identical active blocks of needle stimulation (1 min), separated by a rest period (2 min).

All MRI experiments were performed using a 3 T MR scanner (ISOL Tech, Daejeon, Republic of Korea). It is well known that BOLD contrast depends upon the total amount of deoxygenated hemoglobin present in a brain region, which in turn depends upon the balance between oxygen consumption and oxygen supply. These differential signals could enable measurement of functional changes in brain activity. BOLD functional imaging was carried out using a gradient echo planar imaging (EPI) T2*-weighted sequence (TR 3000 ms, TE 25 ms, flip angle 90°, acquisition matrix 64 × 64, FOV 220 mm, slice thick 4 mm without gap, voxel size 3.43 mm × 3.43 mm × 4 mm). Image collection was preceded by four dummy scans to allow for equilibration of the MRI signal. For anatomical images, a 3D gradient-echo T1-weighted sequence (TR 2800 ms, TE 16 ms, flip angle 60°, FOV 192 × 220, slice thickness 5 mm) was acquired after the functional scans.

The fMRI data were processed using the Statistical Parametric Mapping software (SPM5, http://fil.ion.ac.uk). For motion correction, all functional images were realigned to the first volume of the first scan session. All subjects satisfied our excessive motion threshold of less than 2 mm spatial displacement in any direction. Subsequently, each image volume was normalized to the Montreal Neurological Institute (MNI) space based on Talairach coordinates, and then smoothed spatially using a 5-mm full-width at half-maximum (FWHM) isotropic Gaussian kernel to decrease spatial noise. Low-frequency noise was removed with a high-pass filter, which was applied with default values to the fMRI time series at each voxel. We constructed a design matrix for each subject that included regressors representing the blocks of acupuncture application separately for the verum and sham conditions. Contrast maps were generated between the verum and sham acupuncture regressors for each subject, and the resulting individual contrast maps were used in random-effects group analyses such as one sample t-tests and multiple regression analyses. A statistical threshold of $p < 0.005$ (uncorrected), combined with spatial extent threshold (>10 contiguous voxels), was used to detect any significant signal difference between the verum and sham conditions.

![Fig. 1. Diagram of verum and sham acupuncture and location of acupoint LR2 are presented in the left panel. The sham acupuncture was performed using a new, blunt, telescopic, non-penetrating Park Sham needle. The total scan time for each scan session was 9 min in the right panel. Scanning commenced with a rest period (R1) and was followed by three identical active blocks of needle stimulation (S1, S2, S3), separated by a rest period (R2, R3). For each participant, two fMRI scans were performed using the above block design, one with three consecutive verum acupuncture stimulations and one with three consecutive sham acupuncture stimulations. The order of the two sessions was randomly allocated.](image-url)
In examining the psychophysical assessment between verum and sham acupuncture stimulation, the mean deqi score of the verum acupuncture was significantly higher than that of the sham acupuncture (Mann–Whitney U-test, 2.33 ± 0.56 versus 0.73 ± 0.27; p < 0.001). The mean acute pain score of the verum acupuncture, however, was slightly higher than that of the sham acupuncture, but not significantly (2.75 ± 0.59 versus 1.37 ± 0.34; p > 0.065). The post-scan interview revealed that the question about acupuncture, but not significantly (2.75 ± 0.73 versus 0.59 ± 0.34; p > 0.139). The main contrast of verum acupuncture versus sham acupuncture (VA–SA) using a random effect analysis showed a significant effect in the left medial frontal gyrus (BA9), the caudate, the claustrum, the bilateral posterior cingulate gyrus (BA29, BA31), the bilateral fusiform gyrus (BA20, BA27), the cuneus (BA7), the precuneus gyrus (BA31), and the left declive of the cerebellum (see Fig. 2 and Table 1 for a complete list of regions activated). Contrasts were set up between verum and sham acupuncture at the same acupuncture point, LR2. The main contrast of verum acupuncture versus sham acupuncture (VA–SA) using a random effect analysis showed a significant effect in the left medial frontal gyrus (BA9), the caudate, the claustrum, the bilateral posterior cingulate gyrus (BA29, BA31), the bilateral fusiform gyrus (BA20, BA27), the cuneus (BA7), the precuneus gyrus (BA31), and the left declive of the cerebellum (see Fig. 2 and Table 1 for a complete list of regions activated). However, the contrast of sham acupuncture versus verum acupuncture (SA–VA) revealed no significant increase in activation anywhere in the brain.

The main finding of the present study was that significant differences were observed in the caudate, the claustrum, the declive of the cerebellum, the posterior cingulate gyrus, the medial frontal gyrus, the fusiform gyrus, the cuneus, and the precuneus gyrus when subtracting the BOLD signal of the placebo acupuncture from the verum acupuncture at the motor function-implicated acupoint LR2 [2,8]. Several studies have observed differential activation after acupuncture stimulation between real and minimal acupuncture [15,22]. As to how one should set a suitable control is controversial. Fang et al. [4] have recently failed to provide definitive answers on the issue of acupoint specificity by comparing a meridian point and a non-meridian point. It could be also argued that what is selected as sham point cannot be considered to be inert just because it is not known to be active now. To our knowledge, this is the first reported fMRI study to directly compare neural substrates between verum and sham acupuncture at the same acupuncture point using the well validated placebo needle.

In the present study, we demonstrated that verum acupuncture stimulation at the motor function-implicated acupoint LR2 elicited significantly higher activation than sham acupuncture, over the basal ganglia, such as the caudate and the cerebellum, which are involved primarily with the control of voluntary movement [20]. Li et al. [13] reported that stimulation of sensorimotor-implicated acupoints, but not adjacent non-acupoints, produced activation of the somatotopic sensory cortices, with co-activation of the motor cortices. Furthermore, acupuncture stimulation at GB34 has been reported to be effective in recovering motor function by modulating the cortical activities of the somatomotor area in humans [7,22]. Our findings are consistent with these previous neuroimaging studies of acupuncture that revealed the existence of an acupoint–brain correlation; acupuncture at acupoints implicated in specific functions may modulate the activity of the corresponding cerebral sites. However, it has to be further examined how the transient changes in the BOLD signal that possibly correspond to alteration in regional brain activity exert therapeutic effects on the target organ, i.e. muscle.

Common features of acupuncture points, such as ST36, LI, and GB34, include the modulation of the activity of the pain-related limbic system and limbic-related structures, including the amygdala, hippocampus, cingulate gyrus, and the prefrontal cortex [5,16,21]. Acupuncture-induced modulation of brain activity in these areas has been suggested to influence the affective and cognitive dimensions of pain processing. In this study, we also found an increased BOLD signal in the limbic and higher cognition centers of the pain matrix, such as the cingulate gyrus, fusiform gyrus, and the medial frontal gyrus, with verum acupuncture, as compared to sham acupuncture. Alterations in the limbic area by verum acupuncture in the present study support the notion that acupuncture commonly activates the affective component of the pain neuromatrix. Some might argue that acupuncture generally exhibited signal decreases in larger areas including different limbic, paralimbic and neocortical regions [4]. However, lots of studies have revealed that sham needles also produced placebo analgesia [9,10]. This suggests that fewer limbic structures responses to verum acupuncture observed in the current study could be attributable to the involvement of these ‘limbic touch’ responses to the non-penetrating placebo needle [14].

As the participants were apparently unable to discriminate verum acupuncture from sham acupuncture, the observed differential activations in the central nervous system between the two stimulations are considered to be specific physiological effects of acupuncture. In examining the psychophysical assessment, a significant difference in the deqi rating was observed, but not so in the acute pain rating between verum and sham acupuncture. The elicitation of deqi, a composite sensation that includes numbness and fullness developing at the site of acupuncture stimulation, is considered to be clinically essential to establish treatment efficacy. Kong et al. [9] demonstrated the relationship between the deqi sensation induced by acupuncture and the putative therapeutic effects of acupuncture. The limbic and paralimbic structures of the cortical and subcortical regions were found to demonstrate a concerted attenuation of signal intensity when the subjects experienced deqi [6]. Thus, activation of the limbic area in verum acupuncture may be attributable to the enhanced deqi sensation.
Fig. 2. Normalized SPM T-maps overlaid on the corresponding axial T1-weighted images showing statistically significant (p < 0.005, uncorrected with 10 continuous voxels) brain activation of verum acupuncture at LR2 in contrast to the sham acupuncture. Verum acupuncture-specific activation was seen in (a) the medial frontal gyrus, (b) the caudate, (c) the precuneus, (d) the claustrum, (e) the posterior cingulate, (f) the posterior cingulate, (g) the right fusiform gyrus, (h) the left fusiform gyrus, and (i) the declive of the cerebellum. Talairach Z coordinates for slices are indicated below each figure. The Z value thresholds were above 2.84.

Although non-penetrating placebo needle has been widely used to examine the efficacy of acupuncture in clinical trials, however, its appropriateness as a placebo control is still controversial. Since activity in unmyelinated afferents produces a ‘limbic touch’ response resulting in emotional and hormonal reactions, some have claimed that non-penetrating placebo needle might not be regarded as a proper placebo control [14]. However, lots of studies have validated non-penetrating placebo needle as the best control intervention so far because subjects were not just unable to differentiate the sensation between verum and placebo acupuncture, but elicited a minimal physiologic response [17,19]. Owing to these benefits of non-penetrating placebo needle, it has been widely used in clinical trials. Thus, it is expected that investigation of brain activation to verum acupuncture compared to non-penetrating placebo needle might be helpful to understand neurophysiological mechanisms of genuinely acupuncture-specific therapeutic effect in clinical trials.

It could be argued that activated brain responses derived from the subtraction might be due to less intense deactivation of verum acupuncture compared to placebo acupuncture. A recent study has demonstrated a differential activation pattern in cortical somatosensory areas by comparing verum or placebo acupuncture to the resting state, respectively [23]. Comparing the response of verum or placebo acupuncture to the resting state, however, interfering factors, such as pain or emotion during acupuncture might be involved in the pattern of brain activity. In order to examine the neurophysiological action of verum acupuncture, therefore, the current investigation only analyzed the differential fMRI responses to verum and placebo acupuncture at the same acupoint. Although we could not definitely verify whether the brain responses were derived from more activation or less deactivation to
verum acupuncture, our findings would be alternative approaches to reveal specific brain patterns implicated with the therapeutic effect of acupuncture. In addition, the present study design is exploratory and the sample size is limited. Larger sample studies with greater statistical power are required for the clarification of the differential effect of the genuine acupuncture.

In conclusion, we demonstrated that acupuncture not only elicited acupoint-implicated brain activation, but also modulated affective components of the pain matrix over sham needle stimulation. The present investigation on the differential patterns of brain activation between verum and sham acupuncture provides valuable data on the neurobiological bases of acupuncture.

Acknowledgements

This work was supported by the Korea Science and Engineering Foundation (KOSEF) grant funded by the Korea government (MEST) (R11-2005-014).

References