# Modulation of electroencephalograph activity by manual acupuncture stimulation in healthy subjects: An autoregressive spectral analysis*

Yi Guo-Sheng(伊国胜)[a], Wang Jiang(王 江)[a]†, Deng Bin(邓 斌)[a], Wei Xi-Le(魏熙乐)[a], and Han Chun-Xiao(韩春晓)[b]

[a] *School of Electrical and Automation Engineering, Tianjin University, Tianjin* 300072, *China*
[b] *Tianjin Key Laboratory of Information Sensing & Intelligent Control, Tianjin University of Technology and Education, Tianjin* 300222, *China*

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To investigate whether and how manual acupuncture (MA) modulates brain activities, we design an experiment where acupuncture at acupoint ST36 of the right leg is used to obtain electroencephalograph (EEG) signals in healthy subjects. We adopt the autoregressive (AR) Burg method to estimate the power spectrum of EEG signals and analyze the relative powers in delta (0 Hz–4 Hz), theta (4 Hz–8 Hz), alpha (8 Hz–13 Hz), and beta (13 Hz–30 Hz) bands. Our results show that MA at ST36 can significantly increase the EEG slow wave relative power (delta band) and reduce the fast wave relative powers (alpha and beta bands), while there are no statistical differences in theta band relative power between different acupuncture states. In order to quantify the ratio of slow to fast wave EEG activity, we compute the power ratio index. It is found that the MA can significantly increase the power ratio index, especially in frontal and central lobes. All the results highlight the modulation of brain activities with MA and may provide potential help for the clinical use of acupuncture. The proposed quantitative method of acupuncture signals may be further used to make MA more standardized.

**Keywords:** manual acupuncture, electroencephalograph, autoregressive Burg method, relative power

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## 1. Introduction

Acupuncture, a therapeutic technique originating from ancient China, has been widely used in traditional Chinese medicine. As an important modality of alternative and complementary medicine methodology, acupuncture has been gradually recognized by the world over two thousand years and through clinical trials.[1] A number of randomized clinical trials have shown that acupuncture can be effective in the treatment of neurological conditions and pain, especially in the treatment of sleep disorders, chronic pain syndromes, and anxiety disorders, as proposed by World Health Organization[2] and the United States National Institutes of Health (NIH) Consensus.[3] Of various types of acupuncture, manual acupuncture (MA) is one of the most popular treatments in traditional Chinese medical acupuncture, which has many advantages, such as appropriate stimulus intensity, flexible operation skill, no side-effects, and so on.[4] In recent years, the controversial issues surrounding acupuncture are whether it works and how it works. Previous clinical and experimental studies have indicated that most acupuncture effects are mediated via the brain.[5,6] Therefore, aside from proving the efficacy of acupuncture in the treatment of several medical problems, more and more basic neuroscience research has focused on the modulations of brain activities during acupuncture stimulation. However, the mechanism of the interactions between acupuncture and brain activities is still largely unknown. Furthermore, this technique has mostly been developed on an empirical basis, and not using a solid rational-driven methodology.

Modern imaging techniques, such as electroencephalogram (EEG), positron emission tomography (PET) and functional magnetic resonance imaging (fMRI), can provide an non-invasive method to clinically investigate brain activity.[1,7,8] As a neurophysiological technique, EEG has high temporal resolution and low spatial resolution compared with other techniques,[7,9] which can simultaneously record multi-channel neural electrical signals in different brain areas at the macroscopic level. It can not only characterize the cerebral cortex electrical information directly, but also reflect the brain functions and the states of the whole body. Now it has been widely introduced to investigate brain activities.[7,10–14]

By characterizing EEG signals, various effects of acupuncture stimulation have been proposed. Earlier experimental studies have shown that acupuncture stimulation can reduce gamma waves and increase alpha and theta waves in rabbits,[15] which are similar to those observed during sleep.

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†Corresponding author. E-mail: jiangwang@tju.edu.cn

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Although no consistent results are reported in later studies, acupuncture also induced various EEG changes in humans. Tanaka *et al.*[16] found that all spectral band power increased by acupuncture, while other studies indicated no changes in EEG[17,18] or reduction in EEG activity induced by acupuncture.[19] Furthermore, Chen *et al.*[20] investigated the effects of high and low frequency acupuncture stimulation on the EEG power spectrum. It is found that different frequency acupunctures can induce different effects on EEG theta power. But there are no consistent results and it is difficult to integrate these findings into a unified framework. What is more, the central effects of acupuncture on brain activity still remain unclear, especially in humans.

Therefore, the aim of the present study is to investigate (i) whether acupuncture stimulation at a traditional acupoint affects EEG and (ii) how acupuncture modulates certain EEG band activities. To achieve these goals, we adopt a power spectral method to analyze the EEG signals.

Power spectral analysis is a well-established standard method of analyzing the EEG signals, which makes it possible to perform extensive calculations on the raw EEG signals in real time. The power spectrum can reflect not only the 'frequency content' of the signal but also the distribution of signal power over frequency. Recently, a number of spectral estimation techniques have been developed for EEG signal processing. Among them, the autoregressive (AR) method is the most frequently used parametric method, since there is a well-established topic for the estimation of the parameters in the AR signal models which are found by solving linear equations of the system. It can reduce the spectral losses and give a good frequency resolution compared with classical spectrum estimation method, such as fast Fourier transform (FFT).[21] Also, the AR method needs shorter duration data records than FFT.[22] The Burg method is used to estimate the parameters of the AR model, which ensures a stable AR model and is computationally efficient.[23] The AR Burg method has been widely used to estimate the power spectrum of EEG signals for migraine, epileptic, and alcoholic subjects.[23,24] It has been shown that the AR power spectral density estimates are very close to the true values. Therefore, we use the AR Burg method to estimate the power spectrum of EEG signals evoked by MA to investigate the effects of acupuncture on brain activity.

The rest of the paper is organized as follows. First, the experimental process and analysis methods are explained. Then, the results and corresponding data analysis are presented. Finally, we discuss our results by comparing previous ones and give our conclusions. The results and approach in our study could be useful to lay a foundation for revealing how acupuncture affects brain activity, and may provide a new perspective for traditional Chinese acupuncture analysis.

## 2. Material and methods

### 2.1. Subjects

Experiments are performed in 15 right-handed healthy adult volunteers (age: 23–27 years; eight males and seven females), as approved by the local ethics committee. All the volunteers have no personal history of neurologic or psychiatric disorders, and are not abusing alcohol or illicit drugs. Every subject is free from neurological active medication, or any other factor that may affect EEG activities. They are acupuncture-naive and had no or little experience with acupuncture. All the subjects have been provided with informed consent with the adequate understanding of the purpose and procedure of the study and their informed written consent is obtained according to the declaration of Helsinki.

### 2.2. EEG recording procedure

In our experiment, we acupuncture at the acupoint ST36 (Zusanli) on the right leg, shown in Fig. 1(a). ST36 is an important meridian on Stomach Meridians, which has many therapeutically beneficial effects, such as keeping calm and sedation, treating sleep disorders, controlling stress, handling dizziness insomnia, and gastrointestinal diseases, treating neurological disorders, and so on. This meridian works effectively and has been widely applied in traditional Chinese medical acupuncture experiments.[25] The acupuncture is administered manually by a single licensed acupuncturist with in clinical experience of over 25 years. The stainless steel needles used for ST36 are 0.2 mm in diameter and 40 mm in length.

EEG signal recording is completed by using a Neuroscan system using 20 Ag–AgCl electrodes, which are placed according to the 10–20 international systems as shown in Fig. 1(b). Reference electrodes are placed between A1 and A2, using the earlobe as the reference ground. The cut-off frequencies of the EEG amplifiers are set to 0.5 Hz and 100 Hz. We record 20-channel EEG signals at a 256-Hz sampling rate from 20 scalp electrodes, which are channels Fp1, Fp2, F7, F3, Fz, F4, F8, T3, C3, Cz, C4, T4, T5, P3, Pz, P4, T6, O1, Oz, and O2, respectively.

During the whole procedure, all operations are conducted in a quiet room where the volunteers lie in a soft bed and are awake. There is no obvious electromyographic signal interruption during the whole process, in order to obtain as many artifacts-free EEG data as possible. Before acupuncture, all the subjects have 5 minutes to relax. The subjects are instructed to keep their eyes closed and to relax during the whole acupuncture procedure. An acupuncture needle is inserted at acupoint ST36 of right leg before the EEG recording system is started.

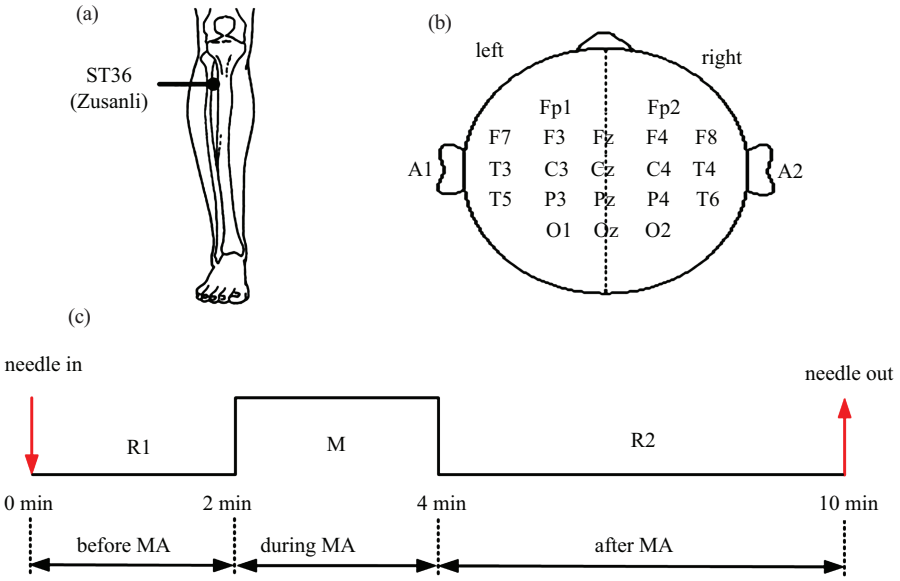



**Fig. 1.** (color online) Acupuncture experiment. (a) Manual acupuncture is administered to the ST36 acupoint on the right side; (b) electrode position on the brain; (c) acupuncture process. "MA" stands for manual acupuncture.

The acupuncture experiment is designed as follows (Fig. 1(c)). The whole process lasts about 10 min for each volunteer. First, the needle is kept in resting state for 2 min (R1), then the subjects are acupunctured by using the twirling method for 2 min (M). The twisting is within a range of 90°–180° and at a rate of about 100 times/min. After acupuncture, subjects are kept in resting state for about 6 min (R2). As the acupuncture frequency 90 times/min–150 times/min is the most widely used in traditional Chinese medical clinical treatment, we choose 100 times/min in our study.

We record EEG signals continuously during the whole acupuncture procedure. Then, we extract three EEG epochs from the recorded EEG data for each subject to represent three states of the experiments, which are 2 min before MA, during MA and 2 min after MA. Then, we intercept the intermediate 100-s data from each EEG epoch, which are free of signs of muscle artifact, eye movements, and other artifacts. Thus, $3 \times 100$-s EEG segments are obtained for each subject. The EEG datasets are preprocessed by a 0.5 Hz∼48 Hz band-pass digital filter, to further eliminate artifacts induced by residual electromyography (EMG) activity and noise, owing to the electrical mains.

### 2.3. Data analysis

In our study, EEG signals are analyzed by using spectral analysis method. The power spectrum of each multi-channel EEG segment is computed by the AR Burg method, which is a powerful tool used for signal modeling. The AR method is a model-based (parametric) method, which is based on modeling the data sequence $x(n)$ as the output of a linear system characterized by a rational structure. In the model based method, there are two steps to estimate the power spectrum. First, estimate the parameters of the model-based method from a given data sequence $x(n)$, $0 \leq n \leq N-1$. Then, compute the power spectral density (PSD) estimate from these estimates. As the AR parameters can be estimated easily by solving linear equations, the AR method is the most frequently used parametric method. In the AR method, data can be modelled as output of a causal, all-pole, discrete filter whose input is white noise. The AR method of order $p$ can be expressed as

$$x(n) = -\sum_{k=1}^{p} a_k x(n-k) + w(n), \tag{1}$$

where $a_k$ are the AR coefficients and $w(n)$ is the white noise of variance equal to $\sigma^2$. The AR($p$) model can be characterized by the AR parameters $\{a_1, a_2, \ldots, a_p, \sigma^2\}$. The PSD is

$$P_{\mathrm{AR}}(f) = \frac{\sigma^2}{\left|1 + \sum_{k=1}^{p} a_k \mathrm{e}^{-\mathrm{j}2\pi fk}\right|^2}. \tag{2}$$

In our study, the AR coefficients are estimated by the Burg method, which is a recursive algorithm.[26] The Burg method is based on minimizing forward and backward prediction errors and estimates the reflection coefficient. From the estimates of the AR parameters by the Burg algorithm, the PSD estimation is obtained as [23,26,27]

$$\hat{P}_{\mathrm{BURG}}(f) = \frac{\hat{e}_p}{\left|1 + \sum_{k=1}^{p} \hat{a}_p(k) \mathrm{e}^{-\mathrm{j}2\pi fk}\right|}, \tag{3}$$

where $\hat{e}_p$ is the total least squares error; $p$ is the model order of AR method and determined by using Akaike information criterion (AIC).[28] In our study, the model order is taken to be $p = 10$.

The EEG contains different specific frequency components, which carry the discriminative information. Therefore, we investigate the relative powers in delta band (0 Hz–4 Hz), theta band (4 Hz–8 Hz), alpha band (8 Hz–13 Hz), and beta band (13 Hz–30 Hz), respectively. The relative power in each sub-band can be obtained by dividing the power in each sub-band by the total power. The features in sub-bands are particularly important for classifying different brain states.

Statistical analysis is performed using the $t$-test, and $P < 0.05$ is considered to be significant.

## 3. Results

We use the AR Burg method to estimate the power spectra of the recorded multi-channel EEG signals in different experimental states. Then, we compute the relative powers in delta, theta, alpha, and beta bands for each subject respectively. Figure 2 shows the relative powers of EEG delta, theta, alpha, and beta bands averaged over 15 subjects at different experimental states. From the results in Fig. 2, one can easily find that acupuncture at ST36 can obviously affect EEG activities. It can be observed that the power increases relatively in delta band during MA compared with before MA, and further increases after MA. Contrary to delta band, the relative powers in alpha and beta bands decrease during MA, and continue to decrease after MA. For the relative power in theta band, there is no obvious difference during MA except for a decrease in frontal lobe compared with before MA. But after MA, most brain areas show a decrease of relative power in theta band compared with before MA.

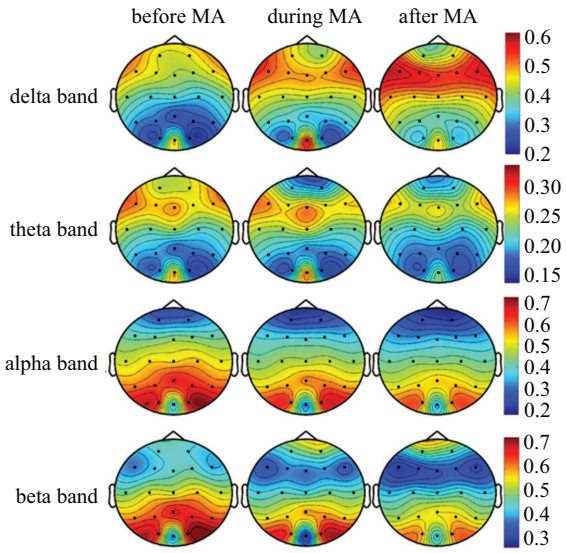



**Fig. 2.** (color online) Topographic maps of relative powers in EEG delta, theta, alpha, and beta bands in different acupuncture states.

From Fig. 2, it can be found the sub-band relative powers are different for different brain areas acupunctured. In order to quantify the relative powers in different brain areas, we compute the sub-band relative powers in frontal, temporal, central, parietal, and occipital lobes, separately. The results of the sub-band relative powers in these five brain areas are shown in Fig. 3.

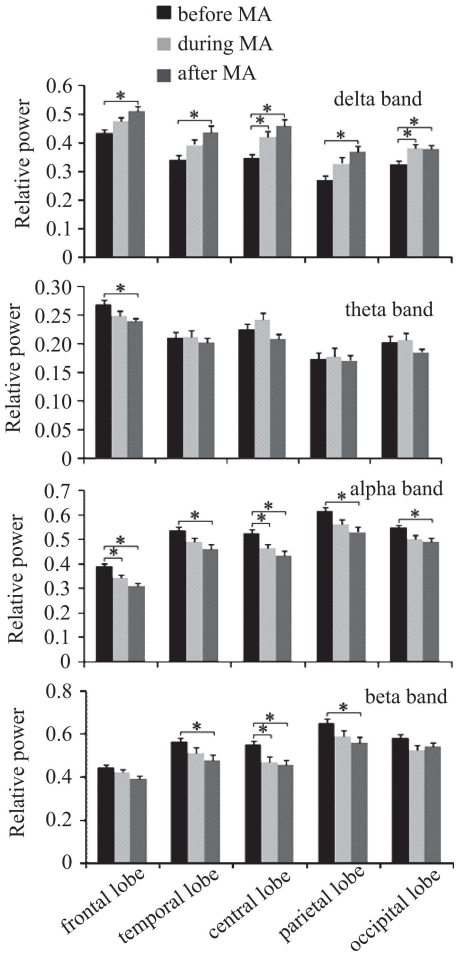



**Fig. 3.** Statistical analysis of the relative powers in delta, theta, alpha, and beta bands of EEG rhythms in different acupuncture states in five brain areas, where the symbol $*$ indicates that the difference between two acupuncture states is significant ($P < 0.05$).

For the delta band, the relative powers in five brain areas are all increased during MA. After MA, and they continue to increase in frontal, temporal, central, and parietal lobes. In occipital lobe, the delta relative power decreases slightly compared with the state during MA, but still higher than that before MA. The increases of relative power of delta band in five brain areas after MA are all significantly different from before MA. However, the differences between during MA and before MA are significant only for central and occipital lobes. For the theta band, the relative powers in temporal, central, parietal, and occipital lobes increase during MA, while decrease in frontal lobe compared with before MA. After MA, the relative powers in five brain areas are all lower than before MA, and the decrease in frontal area is significant. For the alpha band, the relative powers in five brain areas are all lower during MA than before MA, which will further decrease after MA. The

differences in states between after MA and before MA are all significant in five brain areas, while they are significant only in frontal and central lobes during MA. With regards to beta band, the relative powers in five brain areas decrease during MA and continue to decrease after MA, except for the occipital lobe. In the occipital lobe, the relative power of the beta band increases slightly after MA compared with during MA, but still lower than before MA. A significant difference appears in the central lobe during MA and in the temporal, central, parietal lobes after MA.

Figure 4 gives the sub-band relative powers of the whole brain region for different acupuncture states. Compared with the scenario before MA, an increase in relative power of delta band is observed during MA, which further increases after MA. And the increases during and after MA are both significantly different from before MA. The relative power of theta band decreases separately during and after MA. But the decreases are very small and there are no statistical differences between different acupuncture states. The relative powers in alpha and beta bands are all reduced during MA, which continues to decrease after MA. Moreover, the decreases for the relative power of the alpha band during and after MA, and the beta band after MA are significantly different from before MA.

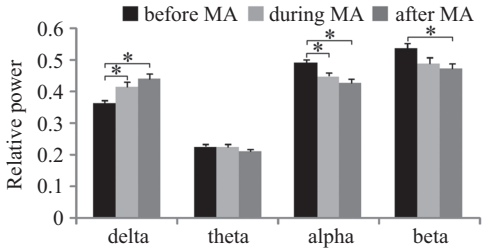



**Fig. 4.** Statistical analyzes of the relative powers in delta, theta, alpha, and beta bands of EEG rhythms at different acupuncture states over the 20 electrodes, where the symbol $*$ indicates that the difference between two acupuncture states is significant ($P < 0.05$).

From the above discussion, it can be concluded that MA can increase the relative power of EEG slow wave (delta band) and reduce the relative power of fast wave (alpha and beta bands). In order to quantify this relative change ratio, we introduce the ratio of slow to fast wave EEG activity. It refers to the power ratio index (PRI), which is calculated as the ratio of the relative power in the delta and theta bands (slow wave activity) to the relative power in the alpha and beta bands (fast wave activity).[29] Figure 5(a) gives the brain topographic maps describing the PRIs at different acupuncture states. It can be seen during MA that the PRIs are higher than before MA in the whole brain area, which are even higher in most brain areas after MA. The PRIs in different brain areas at three acupuncture states are quantified in Fig. 5(b). We can observe that the PRIs in five brain areas are all increased during MA. In frontal, temporal, and central lobes, the PRIs continue to increase after MA. However, in parietal and occipital lobes, the PRIs after MA are lower than during MA, but still higher than before MA. Figure 5(c) shows the PRIs in the whole brain areas for different experimental states. Although there is slight difference in state between during MA and after MA, the significant increases of PRI can be observed during and after MA compared with before MA. Thus, MA at ST36 can increase the PRI, which is consistent with the above results.

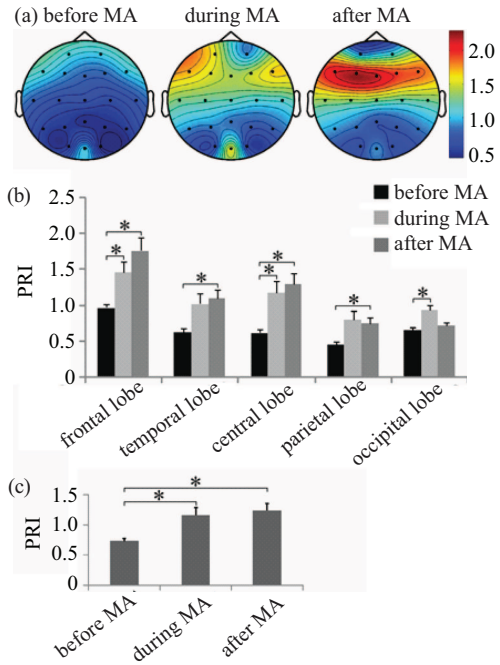



**Fig. 5.** (color online) (a) Topographic maps of PRI at different acupuncture states; (b) statistical analyzes of the PRIs in three experimental states in five brain areas; (c) statistical analyzes of the PRIs over the 20 electrodes for different acupuncture states, where the symbol $*$ indicates that the difference between two acupuncture states is significant ($P < 0.05$).

## 4. Discussion

By computing the EEG power spectrum (using AR Burg method), we systematically investigate the effects of traditional Chinese MA on the activities of different brain rhythms, such as delta, theta, alpha, and beta rhythms. It is found that the acupuncture at ST36 can induce obvious changes in different EEG rhythms in healthy subjects. The central effects of acupuncture stimulus are different from the results of Refs. [17] and [18], which show there are no changes brought about by acupuncture in the EEG signals. The modulations of brain activities with MA provide further support for previous hypothesis that the brain plays a key role in acupuncture research.[7,30–32] Our results also highlight the previous proposal that modulation of cortical electrical activities may be an important mechanism by which acupuncture exerts its complex multisystem effects.[7,31,32]

As a form of complementary and alternative medicine, acupuncture has been widely used in traditional Chinese medicine as a valuable treatment in various diseases. For instance, acupuncture on acupoint ST36 has many therapeuti-

cally beneficial effects, such as treating sleep disorders, keeping calm and sedation, controlling stress, handling dizziness, treating neurological disorders, and so on. However, how acupuncture generates so many beneficial effects is still not well understood. Although there is a lot of research and many theories as regards how acupuncture works, the mode of interactions and real effects are still controversial.[16,18,20] Moreover, most studies focus on the effects of electroacupuncture, available literature on EEG and MA is very rare. In addition, due to a lack of multiple EEG recordings, most MA studies have inadequate control, poor stimulus design, and incomplete statistical analysis, which precludes more conclusive and convincible results from being drawn. Of interest is a recent report about the MA effects on the complexity of EEG signals[7] and the effects of MA on brain information exchange complexity by using the mutual information Lempel–Ziv complexity method.[30] However, how an MA stimulus modulates the activities of different brain rhythms are still unclear. Thus, our current study can contribute to rectify these inadequacies and fill these gaps.

In a physiological sense, EEG power is a measure that reflects the capacity or performance of cortical information processing.[33] It has been found that the delta band power density can reflect sleep intensity,[34] and beta band power is positively correlated with regional cerebral blood flow in regions including the brainstem, pons, midbrain, and thalamus.[35] In addition, Mukai *et al.*[35] and Uchida *et al.*[36] have reported that the beta band power density could reflect the level of a central arousal mechanism. Therefore, the increase in delta band power and decrease in beta band power in the present study suggests that MA at ST36 can reduce the central arousal level, which may be a sleep improvement mechanism. Moreover, it has been shown that the increased alpha band activity could imply the activity of a sleep-disturbing mechanism.[35] So the decrease in relative power of alpha band by MA can also be regarded as an interpretation that acupuncture at ST36 can treat sleep disturbance and further improve sleep quality. All of these conclusions are coincident with the theory of traditional Chinese medical acupuncture.

## 5. Conclusion

In this paper, we adopt the AR Burg method to estimate the power spectrum of the scalp EEG signals evoked by acupuncture at ST36 and compute the relative powers in EEG delta, theta, alpha, and beta bands. Our results show that MA at ST36 can increase the relative power of EEG slow wave (delta band) and decrease the relative power of fast wave (alpha and beta bands). In order to quantify this relative change ratio, we compute the PRI which is the ratio of slow wave EEG activity to fast wave EEG activity. We observe that the MA can significantly increase the PRI, especially in frontal and central lobes. All of these results demonstrate that acupuncture at ST36 can modulate the activities of different brain rhythms in healthy subjects.

Moreover, by investigating the relative powers in different EEG rhythms, we provide a plausible interpretation of why acupuncture at ST36 could improve the quality of sleep. The quantitative method of acupuncture signals can be further used to make MA more standardized, which could facilitate the acceptance and integration of these therapeutic modalities into the practice of modern medicine. Our results could be regarded as a further support for the modulation of brain activities with MA, which may provide potential helps in clinical treatment of acupuncture.

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