



doi:10.3969/j.issn.1673-5374.2013.09.011 [http://www.nrronline.org; http://www.sjzsyj.org]

Wu WC, Chen JQ, Zhen EC, Huang HL, Zhang P, Wang J, Ou YY, Huang Y. Association of auricular pressing and heart rate variability in pre-exam anxiety students. *Neural Regen Res.* 2013;8(9):857-862.

Association of auricular pressing and heart rate variability in pre-exam anxiety students

Wocao Wu, Junqi Chen, Erchuan Zhen, Huanlin Huang, Pei Zhang, Jiao Wang, Yingyi Ou, Yong Huang

TCM School of Southern Medical University, Guangzhou 510515, Guangdong Province, China

Abstract

A total of 30 students scoring between 12 and 20 on the Test Anxiety Scale who had been exhibiting an anxious state > 24 hours, and 30 normal control students were recruited. Indices of heart rate variability were recorded using an Actiheart electrocardiogram recorder at 10 minutes before auricular pressing, in the first half of stimulation and in the second half of stimulation. The results revealed that the standard deviation of all normal to normal intervals and the root mean square of standard deviation of normal to normal intervals were significantly increased after stimulation. The heart rate variability triangular index, very-low-frequency power, low-frequency power, and the ratio of low-frequency to high-frequency power were increased to different degrees after stimulation. Compared with normal controls, the root mean square of standard deviation of normal to normal intervals was significantly increased in anxious students following auricular pressing. These results indicated that auricular pressing can elevate heart rate variability, especially the root mean square of standard deviation of normal to normal intervals in students with pre-exam anxiety.

Wocao Wu and Junqi Chen contributed equally to this article.

Corresponding author: Yong Huang, M.D., Professor, Chief physician, Doctoral supervisor, TCM School of Southern Medical University, Guangzhou 510515, Guangdong Province, China, nhy@fimmu.com.

Received: 2012-11-20
Accepted: 2013-01-05
(N20120301009/WLM)

Key Words

neural regeneration; traditional Chinese medicine; clinical practice; pre-exam anxiety; heart rate variability; auricular pressing; auricular point therapy; cowherb seed; immediate effects; stress; autonomic nerve system disorder; mental disorder; grants-supported paper; neuroregeneration

Research Highlights

- (1) Immediate stimulation of auricular pressing elevated heart rate variability among students with pre-exam anxiety and normal controls.
- (2) Auricular pressing transiently restored balance between sympathetic and vagus nerves in students with pre-exam anxiety.
- (3) Auricular pressing was able to simultaneously excite sympathetic and vagus nerves in normal students.
- (4) Compared with normal controls, auricular pressing increased the root mean square of standard deviation of normal to normal intervals and vagus nerve excitation in students with pre-exam anxiety.
- (5) Elevation of heart rate variability appears to be a potential mechanism for auricular pressing to treat pre-exam anxiety.

INTRODUCTION

Pre-exam anxiety is a type of acute stress disorder^[1]. The disorder typically manifests as nervousness and increased heart rate, and can affect students' exam results^[2]. Severe pre-exam anxiety causes a clear physiological and psychological reaction, directly affecting examination results, and can even influence quality of life and learning ability^[3].

Presently used methods for treating pre-exam anxiety include drugs, acupuncture, auricular point therapy, and psychotherapy^[4-5]. Auricular point therapy is very popular in China, because of its significant curative effect. In addition, auricular point therapy is convenient to learn, has a low price, and no known adverse effects. Auricular pressing is one type of auricular point therapy, in which cowherb seed is fixed on the auricular point using tape, to perform compression stimulation^[6-15]. Auricular pressing has been reported to have significant curative effects in the treatment of chronic disease^[6-15].

Pre-exam anxiety is a special type of anxiety^[3], and anxiety disorders in childhood and adolescence are strongly associated with autonomic nervous system disorders^[16]. The sympathetic and vagus nerves are thought to be distributed on the auricle^[17]. As such, we determined if auricular pressing could correct the disordered state of the autonomic nervous system and restore balance between sympathetic and vagus nerve tensions to treat pre-exam anxiety.

Heart rate variability is a quantitative index of autonomic nervous system function, and can reflect the tension and dynamic balance of the sympathetic and vagus nerves^[18-20].

The present study compared the effects of auricular pressing on heart rate variability between healthy pre-exam students and students with pre-exam anxiety using an Actiheart electrocardiogram recorder.

RESULTS

Quantitative analysis of subjects

A total of 60 subjects were included, consisting of 30 students with pre-exam anxiety (anxiety group) and 30 normal control students (normal group). Two subjects from the anxiety group were excluded because data collection was interrupted. Thus, a total of 58 subjects

were included in the final analysis.

Basic data of pre-exam anxiety students and normal control students

No significant differences in gender, age, height, body mass, body mass index and heart rate were found between the two groups ($P > 0.05$). However, Test Anxiety Scale scores were significantly higher in the anxiety group than in that the control group (Table 1).

Table 1 Comparison of general data of pre-exam anxiety students and normal control students

Item	Anxiety group (n = 28)	Normal group (n = 30)	χ^2/t	P
Gender (male/female, n)	14/14	15/15	0.000	1.000
Age (year)	22.04±0.92	21.63±0.89	1.691	0.096
Height (cm)	164.71±6.46	166.20±7.37	-0.814	0.419
Body mass (kg)	52.75±6.54	55.70±8.55	-1.468	0.148
Body mass index (kg/m ²)	19.42±1.96	20.08±1.99	-1.266	0.211
Heart rate (times/min)	61.86±8.06	63.53±6.25	-0.888	0.378
Test Anxiety Scale	15.32±2.42	6.70±1.75	15.469	0.000

Results are expressed as mean ± SD. *t*-test and chi-square tests revealed no significant differences in gender, age, height, body mass, body mass index and heart rate between the two groups. High scores on Test Anxiety Scale represent increased anxiety.

Auricular pressing elevated heart rate variability in subjects

In the anxiety group, the Actiheart electrocardiogram results revealed that the standard deviation of all normal to normal intervals and the root mean square of standard deviation of normal to normal intervals were significantly increased in the first half (10 minutes) of auricular pressing stimulation compared with baseline. Standard deviation of all normal to normal intervals, logarithm of very-low-frequency power and logarithm of low-frequency power were significantly increased in the second half (10 minutes) of stimulation compared with baseline ($P < 0.05$). In the normal group, standard deviation of all normal to normal intervals, root mean square of standard deviation of normal to normal intervals, logarithm of very-low-frequency power and the ratio of low-frequency to high-frequency power were significantly increased in the first half (10 minutes) of the stimulation period compared with baseline. Standard deviation of all normal to normal intervals, logarithm of low-frequency power, logarithm of very-low-frequency power and ratio of low-frequency to high-frequency power were significantly increased in the second half (10 minutes) of stimulation compared with baseline ($P < 0.05$). Compared with the first half (10 minutes) of the stimulation period, ratio of low-frequency to high-frequency power was higher in the second half (10 minutes) of stimulation in the normal

control group ($P < 0.05$).

Compared with the normal control group in the same time period, the increase in root mean square of standard deviation of normal to normal intervals was significantly greater in the anxiety group in both the first half and second half of the stimulation period ($P < 0.05$; Table 2).

DISCUSSION

According to traditional Chinese medicine, the onset of pre-exam anxiety is associated with heart, liver and kidney function. However, in Western medicine exam anxiety is thought to be associated with an imbalance of excitation and inhibition in the cerebral cortex^[6, 21]. The current study examined six auricular points of the heart, liver, kidney, *Shenmen* (HT7), endocrine and adrenal gland to observe the influence of immediate effects on heart rate variability in pre-exam anxiety students, and to explore the mechanism of action of auricular point therapy.

A previous study reported that anxiety patients exhibited autonomic nerve dysfunction, parasympathetic hypofunction and sympathetic hyperfunction^[22]. The current results revealed that the standard deviation of all normal to normal intervals and root mean square of standard deviation of normal to normal intervals were increased in the first half (10 minutes) of auricular pressing stimulation. Previously, it has been reported that standard deviation of all normal to normal intervals reflects the tension of the autonomic nervous system,

and the root mean square of standard deviation of normal to normal intervals reflects the tension of the vagal system^[23]. The results discussed above suggest that the treatment may have enhanced the regulatory ability of the autonomic nervous system, possibly by increased vagal nerve excitability. Taken together, the current findings indicate that auricular pressing increased standard deviation of all normal to normal intervals and root mean square of standard deviation of normal to normal intervals, possibly reflecting the mechanism of action for treating pre-exam anxiety.

Standard deviation of all normal to normal intervals^[23], which reflects the tension of the autonomic nervous system; very-low-frequency power^[24], which reflects the function of the sympathetic nerve system; and low-frequency power^[24], a quantitative indicator of vagus-sympathetic balance, were increased in the second half (10 minutes) of auricular pressing. The intensity of auricular pressing was presumably relatively weak, so the sympathetic nerve and vagus nerve activity became disordered again after transient recovery (*i.e.* sympathetic hyperfunction). In contrast to the pre-exam anxiety students, standard deviation of all normal to normal intervals, root mean square of standard deviation of normal to normal intervals, logarithm of very-low-frequency power and ratio of low-frequency to high-frequency power were all elevated in normal students in the first half (10 minutes) of the stimulation period. Moreover, the sympathetic and vagus nerves were excited, and their total tension was increased. These findings indicate that autonomic nerve function was in a balanced state after stimulation.

Table 2 Comparison of immediate effects of heart rate variability after auricular pressing between anxiety and normal groups

Item	Group	Pre-stimulation	First 10 minutes of stimulation	Second 10 minutes of stimulation
Standard deviation of all normal to normal intervals (ms)	Anxiety	70.94±27.38	79.64±33.25 ^a	77.95±28.84 ^a
	Normal	60.34±23.35	65.43±24.15 ^a	68.06±23.35 ^a
Standard deviation of the averages of normal to normal intervals (ms)	Anxiety	22.59±19.39	24.32±24.82	20.91±18.10
	Normal	20.48±26.98	17.23±13.45	19.42±16.74
Root mean square of standard deviation of normal to normal intervals (ms)	Anxiety	70.86±45.65	78.55±43.40 ^{ac}	77.28±44.13 ^c
	Normal	54.49±24.04	58.86±25.03 ^a	57.43±25.65
Heart rate variability triangular index	Anxiety	15.71±5.18	16.89±4.65	16.46±4.38
	Normal	14.50±4.65	15.27±4.43	15.13±3.98
Logarithm of very-low-frequency power (ms)	Anxiety	7.07±0.84	7.29±0.93	7.54±0.65 ^a
	Normal	6.77±0.87	7.28±0.77 ^a	7.48±0.86 ^a
Logarithm of low-frequency power (ms)	Anxiety	7.09±0.87	7.27±0.88	7.41±0.73 ^a
	Normal	6.93±1.02	7.07±0.84	7.24±0.87 ^a
Logarithm of high-frequency power (ms)	Anxiety	7.13±1.04	7.28±1.08	7.28±0.96
	Normal	6.94±0.80	6.92±0.84	6.91±0.85
Ratio of low-frequency to high-frequency power	Anxiety	1.32±1.23	1.30±0.85	1.57±1.49
	Normal	1.20±0.75	1.43±0.96 ^a	1.77±1.23 ^{ab}

Results are expressed as mean ± SD. There were 28 subjects in the anxiety group and 30 subjects in the normal group. ^a $P < 0.05$, vs. pre-stimulation; ^b $P < 0.05$, vs. first 10 minutes of stimulation (analysis of variance/Friedman test); ^c $P < 0.05$, vs. normal group (analysis of variance/Mann-Whitney *U* test).

Compared with normal controls, auricular pressing affected root mean square of standard deviation of normal to normal intervals significantly more in pre-exam anxiety students. Pre-exam anxiety students have been previously reported to suffer from autonomic nerve dysfunction, parasympathetic hypofunction and sympathetic hyperfunction^[22]. Root mean square of standard deviation of normal to normal intervals was clearly increased and the vagus nerve was excited during stimulation.

The current findings revealed changes in heart rate variability indices between pre-exam anxiety students and normal control students after auricular pressing, demonstrating that immediate stimulation of auricular pressing could elevate heart rate variability, particularly increasing the root mean square of standard deviation of normal to normal intervals, which may represent a mechanism for the treatment of pre-exam anxiety. However, the present study had several limitations, including small sample size, short course of treatment, and students susceptible to outside influence. These limitations should be addressed in future studies.

SUBJECTS AND METHODS

Design

Electrophysiological, clinical case-controlled study.

Time and setting

Experiments were conducted in the Laboratory of Acupuncture and Manipulation, TCM School of Southern Medical University, China from March to May 2011.

Subjects

A total of 60 subjects were recruited from Southern Medical University, including 30 normal controls and 30 students with pre-exam anxiety. Students were recruited through posters at the University.

Inclusion criteria

Normal control group: (1) no heart diseases, such as valvular heart disease, myocarditis or arrhythmia, no history of organic disease or psychiatric history; (2) did not take drugs that can impair cardiac rhythm such as β receptor blocker or digitalis; (3) no thrombocytopenia, hemophilia or coagulation disorders; (4) did not receive acupuncture treatment within the previous 1 month; did not have fear about acupuncture; (5) have examination within 1 week from testing; (6) scoring < 12 points on the Test Anxiety Scale^[25-26].

Anxiety group: As well as the first five inclusion criteria for the normal control group, the anxiety group had a score of 12–20 points on the Test Anxiety Scale, and experienced anxiety over 24 hours before the examination, as well as meeting the diagnostic criteria of acute stress disorder^[1]: (1) suffering from a stressful life event suddenly before onset, and no other mental disorders; (2) a clear correlation between symptom occurrence and stress source; onset within several minutes or several hours; (3) clinically exhibiting psychomotor excitement, *i.e.* fearful emotions and other autonomic nervous system symptoms; (4) short course of symptoms, typically between several hours to 1 week, with good prognosis.

Exclusion criteria

(1) Secondary anxiety following hyperthyroidism, hypertension and coronary atherosclerotic heart disease; (2) analeptic drug overdose, sedatives or anti-anxiety drugs, anxiety combined with compulsion, phobia, hypochondria, neurasthenia, mania, depression or schizophrenia.

We obtained informed consent from all subjects, and conducted the study in accordance with the ethical requirements of the *Helsinki Declaration*.

Methods

Experimental procedures

All experiments were performed from 19:00 to 24:00 in the same quiet comfortable environment. Before the experiment, all students wore Aearo-Express ear plugs (lot No. R5A018; Aearo Company, Indianapolis, IN, USA) and a black eye shield (lot No. MRX003-C011, Annan Tanye Technology Co., Ltd., Lishui, Zhejiang Province, China). After the Actiheart equipment was set up, the students lay on their back for 10 minutes, and the laboratory technician began to record the time. 10 minutes later, the laboratory technician removed the ear plug on one side, sterilized the auricle, and then stuck a cowherb seed (Guangzhou Suixin Medical Instruments Co., Ltd., Guangzhou, Guangdong Province, China) onto a 0.5 × 0.5 cm² piece of medical tape. The tape was stuck on the auricular point using a forceps. The ear plug was then replaced. Auricular pressing was conducted and heart rate variability was monitored.

Auricular point selection and stimulation method

The main stimulation points included the heart, liver, kidney, and *Shenmen*. Adjunct points included the endocrine and adrenal gland. Acupoint locations were chosen in accordance with the *Nomenclature and*

Location of Auricular Points^[27].

The stimulation method was performed as follows: auricular points were compressed by the same laboratory technician using the forefinger and thumb, positioned vertically, applying firm and well-distributed pressure, so that some tenderness of the ear was felt. Stimulation was applied with a frequency of twice per second, and each acupoint was stimulated in order. Each main point was stimulated for 4 minutes, and each adjunct point for 2 minutes. Compression was terminated 20 minutes later, and the time was recorded. The students still lay on their back for 20 minutes. The laboratory technicians removed the equipment and recorded the time.

Monitoring heart rate variability

After the operational procedure was installed, the Actiheart card reader connected to an USB interface of a computer (Actiheart, Cambridge Neurotechnology Ltd., Cambridge, UK). "Short-range record" was selected to record general conditions of each subject, including name, date of birth, gender, height and weight. The subjects lay on their back and rested for 10 minutes. An Ambu electrocardiogram electrode slice (Ambu blue sensor VL-00-S, Ambu A/S, Ballerup, Denmark) was attached to the xiphoid process, and the Actiheart electrocardio-monitor was placed on this electrode slice. The wire from the monitor was spread out to the left and kept horizontally straight, while another electrode slice was attached at the end of this wire. Monitoring began 10 minutes before stimulation, and continued until the end of 20-minute stimulation period. The record was conducted by the same person for each subject^[28-29].

Data processing

The monitor was placed on the card reader, and the data were read and stored. After entering the short-range record procedure, sampling time was set at 5 minutes to increase the comparability with pre-stimulation data. Three time periods (pre-stimulation, first 10 minutes of stimulation and second 10 minutes of stimulation) were selected for analysis, and data were entered into Excel. Time domain indices of heart rate variability were recorded, consisting of standard deviation of all normal to normal intervals, root mean square of standard deviation of normal to normal intervals and heart rate variability triangular index. The whole experiment was divided into consecutive 5-minute time periods. The average normal to normal interval was calculated for every 5-minute period, followed by the standard deviation of the average of normal to normal interval. Frequency domain indices were recorded, containing very-low-frequency power, low-

frequency power and high-frequency power, and ratio of low-frequency to high-frequency power^[23, 30]. Logarithmic transformation was carried out on very-low-frequency power, low-frequency power and high-frequency power.

Statistical analysis

Using SPSS 13.0 software (SPSS, Chicago, IL, USA), analysis of variance and non-parametric testing were used to compare the difference in heart rate variability indices before stimulation, the first 10 minutes of stimulation and the second 10 minutes of stimulation in both groups. Results were expressed as mean \pm SD. A value of $P < 0.05$ was considered statistically significant.

Acknowledgments: We thank all the participants and Professor Gustav Wik from Department of Clinical Medicine, University of Bergen for technical support.

Funding: This project was funded by the Undergraduate Innovation Experimental Design Program of Guangdong Province, No. 1212110041; and the Extracurricular Scientific Research Project of Southern Medical University, No. 2009kw034.

Author contributions: Yong Huang participated in study concept and design, manuscript authorization and obtaining funding. Pei Zhang and Jiao Wang were in charge of recruiting volunteers. Huanlin Huang and Yingyi Ou performed data analysis. Junqi Chen stored and integrated the data and wrote the manuscript. Wocao Wu was responsible for manuscript writing and experiment enforcement. Erchuan Zhen performed the experiments. All authors approved the final version of the paper.

Conflicts of interest: None declared.

Ethical approval: This study was registered on the Chinese clinical trial register, registration No. ChiCTR-OCH-12002332; no ethics inspection, because of noninvasive intervention.

Author statements: The manuscript is original, has not been submitted to and is not under consideration by another publication, has not been previously published in any language or any form, including electronic, and contains no disclosure of confidential information or authorship/patent application/funding source disputations.

REFERENCES

- [1] Shen YC. Pathergasiology. Beijing: People's Medical Publishing House. 2009.
- [2] Shu S, Li TM, Fang FF, et al. Relieving pre-exam anxiety syndrome with wrist-ankle acupuncture: a randomized controlled trial. *Zhong Xi Yi Jie He Xue Bao*. 2011;9(6): 605-610.
- [3] Xu J, Xie YN, Zhao JB, et al. Effects of self-concept on test anxiety level among sophomores in a medical college. *Di Yi Jun Yi Da Xue Xue Bao*. 2005;25(6):759-760.

- [4] Lu SK. Subject of Acupuncture and Moxibustion Technique. Beijing: China Press of Traditional Chinese Medicine. 2007.
- [5] Wang QC. Subject of Acupuncture and Moxibustion Therapy. Beijing: China Press of Traditional Chinese Medicine. 2007.
- [6] Huang LC. Subject of Auricular Pressing Therapy. Beijing: Scientific and Technical Documentation Press. 2005.
- [7] Wang XH, Yuan YD, Wang BF. Clinical observation on effect of auricular acupoint pressing in treating sleep apnea syndrome. *Zhongguo Zhong Xi Yi Jie He Za Zhi*. 2003;23(10):747-749.
- [8] Dong WY, Wu C, Jin ZT, et al. Clinical observation on auricular-plaster therapy combined with chiropractics for treatment of emaciation in children. *Zhongguo Zhen Jiu*. 2007;27(3):185-187.
- [9] Song SJ. Observation on therapeutic effect of ear point blood-letting combined with cupping on Back-shu points for treatment of acne vulgaris. *Zhongguo Zhen Jiu*. 2007;27(8):626-628.
- [10] Wu RD, Zhang HD, Lin LF. Observation on ear point taping and pressing therapy for treatment of primary dysmenorrhea. *Zhongguo Zhen Jiu*. 2007;27(11):815-817.
- [11] Chen Q, Huang HM, Xu YJ, et al. Controlled study of auricular point taping and pressing therapy for treatment of vascular dementia. *Zhongguo Zhen Jiu*. 2009;29(2):95-97.
- [12] Jin YB, Sun ZL, Jin HF. Randomized controlled study on ear-electroacupuncture treatment of endometriosis-induced dysmenorrhea in patients. *Zhen Ci Yan Jiu*. 2009;34(3):188-192.
- [13] Wang SX, Wang ZH. Debating some issues on the application of auricular point. *Zhongguo Zhen Jiu*. 2009;29(8):637-638.
- [14] Tong PJ, Wang HD, Ma ZC. Application and effect of auricular acupoint pressing for analgesia in perioperative period of total knee joint replacement. *Zhongguo Zhong Xi Yi Jie He Za Zhi*. 2010;30(9):931-934.
- [15] Gong RL. Observation on therapeutic effect of child amblyopia treated with auricular point sticking therapy. *Zhongguo Zhen Jiu*. 2011;31(12):1081-1083.
- [16] Sharma RK, Balhara YP, Sagar R, et al. Heart rate variability study of childhood anxiety disorders. *J Cardiovasc Dis Res*. 2011;2(2):115-122.
- [17] Zhong SZ. Systematic Anatomy. Beijing: Higher Education Press. 2003.
- [18] Wang Y, Li GS. Effect of vagal nerve stimulation on heart rate and heart rate variability in the toads. *Zhongguo Ying Yong Sheng Li Xue Za Zhi*. 2010;26(2):229-232.
- [19] Kong LX, He XH. Relationship between heart rate variability and cold-heat/deficiency-excess syndromes in TCM. *Zhongyi Zazhi*. 2010;51(4):348-351.
- [20] Bauer A, Malik M, Schmidt G, et al. Heart rate turbulence: standards of measurement, physiological interpretation, and clinical use: International Society for Holter and Noninvasive Electrophysiology Consensus. *J Am Coll Cardiol*. 2008;52(17):1353-1365.
- [21] Lv PW. Prevention of exam syndrome using auricular pressing. *Jiankang Bolan*. 2007;13(1):27.
- [22] Thayer JF, Friedman BH, Borkovec TD. Autonomic characteristics of generalized anxiety disorder and worry. *Biol Psychiatry*. 1996;39(4):255-266.
- [23] Guo JH, Zhang P. Dynamic Electrocardiography. Beijing: People's Medical Publishing House. 2004.
- [24] Gao YH, Chen SP, Wang JY, et al. Effects of electroacupuncture of different acupoints groups on blood pressure and heart rate variability in rats. *Zhen Ci Yan Jiu*. 2009;34(1):21-26.
- [25] Wang CK, Liu Y. Correlation among general self-efficacy, trait anxiety, state anxiety and test anxiety. *Zhongguo Xinli Weisheng Zazhi*. 2000;8(4):229-230.
- [26] Wang CK. Reliability and validity of test anxiety scale-Chinese version. *Zhongguo Xinli Weisheng Zazhi*. 2001;15(2):96-97.
- [27] General Administration of Quality Supervision, Inspection and Quarantine of the People's Republic of China, Standardization Administration of the People's Republic of China. GB/T 13734-2008 Nomenclature and Location of Auricular Points. Beijing: Standards Press of China. 2008.
- [28] Brage S, Brage N, Franks PW, et al. Reliability and validity of the combined heart rate and movement sensor Actiheart. *Eur J Clin Nutr*. 2005;59(4):561-570.
- [29] Fasmer OB, Liao H, Huang Y, et al. A naturalistic study of the effect of acupuncture on heart-rate variability. *J Acupunct Meridian Stud*. 2012;5(1):15-20.
- [30] Kleiger RE, Stein PK, Bigger JT Jr. Heart rate variability: measurement and clinical utility. *Ann Noninvasive Electrocardiol*. 2005;10(1):88-101.

(Edited by Ju DH, Zhang L/Qiu Y/Wang L)