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1 Section V: Psychology of Sport, Exercise, and Health (Andreas Ivarsson)

2 Article

Effects of three-month Qigong exercise on heart rate variability and respiration in anxious college students

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11 Abstract: Objective: This longitudinal study aimed to investigate the effects of Qigong on the anxiety state, heart rate variability (HRV), and breathing of anxious college students. Methods: A total of 37 individuals (18-25 years old) were 12 13 randomly allocated to the control (n = 19) and intervention (n = 18) groups. Qigong interventions were conducted five times 14 weekly for 12 weeks, with each session lasting 60 min. Hamilton Anxiety Scale, Fatigue Scale 14, Pittsburgh Sleep Quality 15 Index, and 36-item Short Form Survey (SF-36), HRV, and respiration data were collected before and after the 3-month 16 intervention Results: Individuals who participated in the three-month Qigong exercise intervention showed a significant 17 reduction in anxiety, particularly mental anxiety (P < 0.05). Subjects in the intervention group presented a decrease in skin temperature (P < 0.05) and an increase in blood volume pulsation (P < 0.05). Meanwhile, HRV exhibited a significant 18 19 increase in the standard deviation of interbeat interval before and after comparisons (P < 0.05) and between the two groups 20 (P = 0.039) and a reduction in the normalized low-frequency power after the intervention. Moreover, the intervention group 21 experienced increased abdominal breathing depth and abdominal breathing per minute (P < 0.05). Conclusion: These findings 22 indicate that Qigong is an effective mind-body exercise strategy for relieving anxiety. HRV and breathing were improved 23 accordingly among college students after the completion of the 3-month Qigong program.

24 Keywords: Qigong; HRV; respiration; college students; anxiety

25

26 1. Introduction

Excessive academic, social, and life pressure among college students and increased sedentary time have 27 serious detrimental effects on their physical and mental health, which can easily lead to negative emotions¹. 28 Contemporary college students face more negative emotions than ever before, with the incidence of anxiety, 29 sleep disorders, and depression reaching 32.73%, 15.43%, and 62.91%, respectively². On a psychological 30 level, anxiety shows association with a poor mental health³; on a medical level, it is associated with an 31 increased all-cause mortality⁴. These data indicate the necessity of developing a method that can effectively 32 relieve anxiety in college students. Although some drug treatments have been developed, they may be 33 accompanied with side effects, and occasionally, their effectiveness is uncertain⁵. By contrast, exercise is an 34 effective, inexpensive, and simple means of reducing anxiety⁶. 35

Fitness Qigong, as a traditional sport in China, has ameliorative effects on a variety of chronic diseases, such as diabetes⁷, hypertension⁸, chronic heart disease⁹, and cancer¹⁰, particularly in reducing anxiety and depression levels¹¹. However, studies on fitness Qigong primarily focused on its effects on the physical fitness among the elderly ¹², and a limited number are related to anxious college students. Compared with highly confrontational, high-intensity, and difficult exercise programs, fitness Qigong is more suitable for college students with poor physical fitness, no exercise habits, and are prone to anxiety symptoms because of its "three tunes in one" for regulating the body, breath, and mind. Fitness Qigong includes many types, which all have different fitness effects¹³. Yijinjing enhances physical fitness and prevents diseases through postural adjustments and breathing exercises, whereas Liuzijue improves the functions of internal organs and alleviates negative emotions through vocalization¹⁴. Therefore, this study combined Yijinjing and Liuzijue to develop an exercise intervention program for anxious college students.

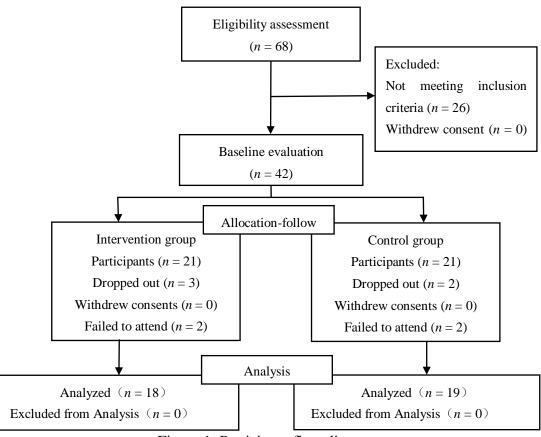
Anxiety and negative emotions can be assessed through certain physiological measures, particularly heart 47 rate variability (HRV)¹⁵. HRV is a measure of cardiac vagal tone. It can be quantified through the spectral 48 analysis of interbeat interval (RR)¹⁶. A low HRV is related to a number of psychopathological states, including 49 anxiety¹⁷. However, the effect of Qigong on the HRV of anxious college students remains poorly understood. 50 Breathing exercises (as long as the inhalation/exhalation ratio is equal) as short as 5 min promote a balance 51 between sympathetic and parasympathetic activities ¹⁸, which increases HRV and improves 52 psychophysiological coherence¹⁹. Therefore, fitness Qigong may reduce anxiety by modulating HRV, 53 breathing depth, and breathing frequency. 54

In this study, we initially determined the alleviating effect of combined Qigong practices on the anxiety of college students. We also analyzed the possible mechanisms of fitness Qigong in improving anxiety by comparing the changes in HRV and respiratory indexes before and after fitness Qigong practice and identified multidimensional, subjective, and objective measurement and evaluation to illustrate its improvement effect on the anxiety status of college students.

60 2. Methods

61 2.1 Participants

62 A total of 68 undergraduate students (30 males and 38 females; mean age = 21.20 years, range: 18-25years; mean body mass index (BMI) = 22.17, range: 18–30) participated in this experiment (Table 1). Males 63 64 and females were grouped together because differences in the results showed no statistical significance for any of the variables measured. The students were asked to complete a basic questionnaire prior to exercise 65 intervention. Those who satisfied the following criteria were qualified to participate in the study: (1) aged 66 between 18 and 25 years; (2) sedentary lifestyle (<150 min of moderate-to-high-intensity physical activity per 67 week, and >8 h daily sedentary time); (3) severe anxiety (scores in the Hamilton Anxiety Rating Scale [HAM-68 A] >14); (4) willingness to participate in the test and sign an informed consent form. Participants were 69 70 excluded if they met the following exclusion criteria: (1) diagnosed with a disease that affects the performance of movements; (2) history of heart disease, severe arrhythmia, or pacemaker use; (3) habitual smoking or 71 heavy consumption of alcohol or other stimulant beverages; (4) have taken drugs that may affect emotional 72 reaction, such as antihypertensives, anticholinergics, anxiolytics, or antidepressants, at least 1 month before 73 the start of the study. Of the included individuals, 26 did not meet the screening criteria and hence were 74 eliminated from the study (Fig. 1). The remaining 42 participated in the experiment. The subjects were 75 76 randomly divided into two groups: the control group, who did not conduct any regular exercise, and the intervention group, who participated in regular Qigong exercise. Two and three individuals dropped out after 77 being assigned to the control (n = 19) and intervention (n = 18) groups. 78



79 80

Figure 1. Participant flow diagram

81 2.2 Experimental procedures

Participants completed the HAM-A for anxiety, Fatigue Scale 14 (FS-14), Pittsburgh Sleep Quality Index 82 (PSQI), and 36-Item Short Form Survey (SF-36). Prior to the test, they were asked to inhibit from drinking 83 caffeinated or alcoholic beverages for at least 24 h. All the subjects were tested individually, and data were 84 collected in a constant-temperature (24 °C–26 °C) and bright lounge between 14:00 and 17:00 pm. Then, the 85 participants were instructed to sit on a chair and rest for 5 min to determine the indicators of multiple 86 physiological parameters (MPPs), HRV, and respiration (pretest). Finally, signals were collected using a 87 88 biofeedback instrument and transmitted to a laptop for postanalysis. The intervention group adopted the combined Yijinjing and Liuzijue Qigong styles as the training method under the guidance of experienced 89 coaches. The Qigong program lasted 12 weeks, with a frequency of 5 days a week and 60 min a day and 90 including a 10 min preworkout warm-up, 40 min exercise workout, and 10 min cool down. At the end of the 91 92 3-month intervention, the subjects were reassessed in accordance with the preintervention steps (posttest). All exercise and measurement procedures were conducted under professional guidance. The study was approved 93 by the Ethics Committee of Tongji University (2021tjdx024). 94

95 2.3 Measurements

96 2.3.1 Scale evaluation

Prior to Qigong interventions, the participants were assessed using the HAM-A, FS-14, PSQI, and SF-97 36. HAM-A measures the severity of anxiety symptoms in patients with neuroses and others who have 98 different conditions, primarily adults with anxiety symptoms²⁰. FS-14 consists of 14 entries, each of which 99 focuses on a fatigue-related issue. These 14 questions reflect the severity of fatigue from various perspectives, 100 and they are divided into two categories, including physical fatigue and mental fatigue, through principal 101 component analysis²¹. PSQI refers to a quantity scale that is used to evaluate sleep quality in general²². SF-36 102 is a frequently used, well-researched measurement of health. It comprises 36 questions, including eight 103 domains of health, and commonly used to measure a population's life quality ²³. 104

105 2.3.2 MPP measurement

MPPs, including skin conductance level, skin temperature (TEMP), blood volume pulsation, blood volume amplitude, and pulse, were evaluated using biofeedback instruments (2000x-pert, Biopac Systems Inc.). The skin conduction sensor had two electrodes. The electrode was tightly tied to the end of a finger to maintain a good contact but not extremely tight to allow blood circulation. For MPP measurement, Bioneural Infiniti v5.1 operating platform, which can formulate single-data reports or trend reports of MPP data, was used for data acquisition, analysis, and management.

112 2.3.3 HRV measurement

As an important indicator of cardiac sympathetic–parasympathetic tone, the HRV primarily reflects changes in the RR²⁴. HRV can be measured in the time and frequency domains. In time-domain analysis, we used SDNN and PNN50 metrics, which can reflect the total variation and magnitude of HRV, respectively, as measures.

HRV was measured using biofeedback instruments(2000x-pert, Biopac Systems Inc.). R-wave detection was used to identify R-wave peaks in recorded electrocardiographic signals and measure continuous RR during tracking. HRV-related data were collected before and after the Qigong intervention and labeled for separation.

121 2.3.4 Respiration measurement

The respiratory index, including the amplitude and frequency of breathing in the chest and abdomen, was 122 assessed using a biofeedback instrument (2000x-pert, Biopac Corporation). Circumferential changes were 123 measured using ultrasound signals to maintain the accuracy of readings. Breathing measurements included 124 four indicators: (1) maximum and minimum of the abdominal breathing curve (RESPA-A); (2) the number of 125 breaths per minute (calculated from the abdominal respiratory curve [RESPF-A]); (3) maximum and minimum 126 127 of the thoracic respiratory curve (RESPA-C); (4) the number of breaths per minute, which was calculated on the basis of the chest respiratory curve (RESPF-C). The participants were required to breathe naturally before 128 and after the exercise and coordinate their movements and breathing during Oigong exercises. 129

130 2.4 Statistics and analysis

131 Statistical analyses were performed using the SPSS statistical package (IBM, SPSS Statistics 20). The extracted data included participant characteristics, questionnaire scores, HRV, respiration, and MPPs. The 132 means and standard deviation (SD) of key outcome indicators were extracted at baseline and at time point 133 closest to the end of exercise. Levine's and Shapiro-Wilk tests were used to check the homogeneity of variance 134 135 and normality of data, respectively. All data were normally distributed. Data that did not meet the test parameters were converted when necessary via regression. Independent sample t-test was used to compare the 136 baseline characteristics of the two groups, and the same was performed for the results of HAM-A, FS-14, 137 PSQI, and SF-36 between the two groups. Independent and paired sample t-tests were conducted to compare 138 the differences between pretesting and posttesting, respectively, of MPP, HRV, and respiration. The 139 confidence level was set at 95% (P < 0.05) in all statistical analyses. 140

141 3. Results

142 3.1 Demographic characteristics and screening criteria for participants in the two groups

The study initially screened 68 sedentary college students, 26 of whom were prescreened out for failing to meet the criteria and 5 of whom dropped out. Finally, 37 students, all of which were anxious young adults, participated in this research. Similarities in their basic information and subhealth levels were considered during grouping to ensure that no significant differences exist in various baseline parameters between the two groups (males and females and control versus intervention groups). Table 1 shows the basic characteristics of the participants between the two groups, which showed no significant differences in their age, sex, body weight, body height, BMI, and education level.

150

Table 1.	Characteristic	s of participants
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Variable	Control (<i>n</i> = 19)	Intervention $(n = 18)$	Total
N (%)	19 (51.4%)	18 (48.6%)	37 (100%)

Age	20.2 ± 3.5	21.7 ± 5.1	20.9 ± 4.3
Sex (M/F)	11/8	7 /11	18/19
Height (cm)	165.2 ± 10.2	164.9 ± 8.4	165.1 ± 9.3
Weight (kg)	62.46 ± 5.25	59.18 ± 4.53	60.86 ± 4.90
BMI (kg/m ²)	19.76 ± 2.06	21.23 ± 5.82	20.48 ± 3.89
Education level (bachelor degree and postgraduate, %)	89.34	82.11	85.82

151 Data are presented as mean \pm SD. BMI, body mass index.

152 3.2 Effect of Qigong intervention on anxiety status among college students

After the three-month Qigong exercise intervention, significant improvements (decrease in 153 questionnaires scores) in the total anxiety (P = 0.039) and mental anxiety (P = 0.004) were observed. Physical 154 anxiety did not change, which was possibly because the amount of Qigong exercise did not reach the level of 155 changing physical anxiety for a three-month beginner. Compared with the control group, the intervention 156 group showed a significant improvement in physical pain (P = 0.013) In addition, the intervention group 157 attained a lower sleep quality score (P = 0.027), which indicated an improvement. "Life quality" and "Bodily 158 pain" showed high SDs, which can be explained by the anxiety conditions of participants, who could have 159 different perceptions (Table 2). 160

Table 2. Effect of Qigong on anxiety, fatigue, and sleep quality among anxious college students

	Control (<i>n</i> = 19)	Intervention $(n = 18)$	Change (95%)	P value
Total anxiety (Ham-a)	15.67±5.94	11.63±5.50*	4.04 (0.217 to 7.853)	0.039
Physical anxiety (Ham-a)	6.50±3.09	5.89±3.11	0.61 (-1.465 to 2.675)	0.714
Mental anxiety (Ham-a)	9.17±4.29	5.74±5.05*	3.21 (1.094 to 5.332)	0.004
Total fatigue (FS-14)	8.39±2.83	8.16±2.81	0.13 (-1.654 to 2.116)	0.805
Sleep quality (PSQI)	7.61±2.59	$6.06 \pm 1.58*$	1.66 (0.208 to 3.120)	0.027
Life quality (SF-36)	511.51±98.63	523.91±104.3	-5.21 (-50.737 to 40.313)	0.817
Bodily pain (SF-36)	63.29±30.55	84.13±11.28*	-20.83 (-36.866 to -4.795)	0.013

162 Data are presented as mean \pm SD, * P < 0.05 represents statistically significant difference.

163 3.3 Effect of Qigong intervention on MPPs of anxious college students

Psychological improvements related to anxiety levels also cause changes in basic physiological indicators. Thus, in this study, the effects of Qigong on these indicators were investigated through measurement of several parameters. After the Qigong intervention, the TEMP index in the intervention group was significantly lower than the preintervention level (P < 0.05). Meanwhile, the BVP in the intervention group showed a significant increase after the intervention (P < 0.05) and responded more prominently to Qigong intervention than that in the control group (P = 0.029). Other indices exhibited no significant change between the groups before and after the intervention (Table 3).

171 Table 3. Effects of Qigong on the MPPs of pre- and posttests in two groups (control and intervention) of

172 anxious college students

161

Index	Control (<i>n</i> = 19)			Intervention $(n = 18)$			
	Pre	Post	Change (95%CI)	Pre	Post	Change (95%CI)	P value
SCL	1.24±0.70	1.24±0.96	0.00 (-0.649 to 0.653)	1.43±0.83	4.29±6.31	-2.86 (-6.500 to 0.774)	0.114
TEMP	32.04±3.76	30.09±4.40	1.95 (-2.196 to 6.105)	34.43±2.78	31.52±3.81*	2.91 (1.641 to 4.171)	0.620

BVP	49.45±0.45	49.74±1.20	-0.29 (-1.421 to 0.831)	48.03±2.38	51.40±3.61*	-3.36 (-5.935 to -2.881)	0.029
PVA	67.88±22.97	44.14±21.47	23.74 (-5.598 to 53.091)	54.64±23.02	48.53±19.65	6.12 (-12.195 to 24.427)	0.235
PULS	68.66 ± 5.25	67.40±4.19	1.26 (-2.145 to 4.673)	73.93±7.90	74.87±5.13	-0.94 (-6.834 to 4.945)	0.478

Data are presented as mean \pm SD. * represents statistically significant difference in pre- and post-test. *P* value indicates differences in variables between the control and intervention group. SCL, skin conductance level; TEMP, skin temperature; BVP, blood volume

175 pulsation; PVA, blood volume amplitude, relative maximum recorded range of BVP peak values; PULS, number of heart beats per

176 minute.

177 3.4 Effects of Qigong on HRV of anxious college students

178 In the time-domain analysis, SDNN in the intervention group presented a significant increase in after the intervention (P < 0.05). Furthermore, the changes in SDNN (P = 0.039) between the two groups showed a 179 significant difference after the intervention. Frequency-domain analysis enables the detailed observation of 180 sympathetic and vagal modulation, which compensates for the poor sensitivity and specificity of time-domain 181 methods²⁵. Therefore, we further analyzed the frequency domain and selected three indicators: the low-182 frequency power spectrum band (LF: 0.04–0.15 Hz), high-frequency power spectrum band (HF: 0.15–0.4 Hz), 183 and LF/HF ratio. The results showed that the normalized LF in the intervention group was significantly lower 184 after Qigong intervention than before intervention (P < 0.05). No statistically significant difference in 185 normalized HF and LF/HF was observed within and between groups, and the results are reported in Table 4. 186

187 Table 4. Effects of Qigong on the HRV of the control and intervention groups in the pre- and posttests.

Index	Control (n = 19)			Intervention $(n = 18)$			
	Pre	Post	Change (95%CI)	Pre	Post	Change (95%CI)	P value
SDNN	87.79±71.81	66.91±25.36	20.88 (-43.343 to 85.102)	78.59+39.04	123.61+59.58*	-45.02	0.335
SDIG	01119211101	00.71_20.00	20100 (101010 10 10 001102)	/0.0/_0/01	120:01_07.00	(-79.737 to -10.304)	
PNN50	29.90±19.44	30.93±10.03	-1.03 (-17.848 to 15.786)	33.18±23.48	31.93±19.08	1.25 (-12.061 to 14.553)	0.523
nLF	$40.80{\pm}10.17$	44.94 ± 9.76	-3.13 (-14.159 to 7.900)	43.78 ± 10.17	37.77±6.67*	5.01 (1.314 to 8.715)	0.351
nHF	59.18±10.17	56.52±12.36	2.67 (-8.343 to 13.682)	58.68 ± 11.27	60.57 ± 7.52	-1.89 (-7.825 to 4.041)	0.307
LF/HF	0.73±0.29	0.87 ± 0.54	-0.14 (0.195 to 0.326)	0.77 ± 0.38	0.80±0.34	-0.03 (-0.273 to 0.213)	0.195

188Data are presented as mean \pm SD. * represents statistically significant difference in pre- and post-test. P value indicates189differences in variables between the control and intervention group. SDNN, the standard deviation of R-R intervals; PNN50,

190 the percentage of difference over 50 ms between adjacent normal-normal R-R intervals. nLF, normalized low-frequency

191 power; nHF, normalized high-frequency power; LF/HF, low-frequency/high-frequency ratio.

192 3.5 Effect of Qigong exercise on respiration regulation in anxious college students

The two main modes of physiological breathing include thoracic and abdominal breathing, which are 193 also known as shallow and deep breathing. We analyzed the differences in the amplitude and frequency 194 between thoracic and abdominal respiration to gain insights into the effect of Qigong exercises on respiratory 195 regulation. Compared with those in the pretest, RESP-A (P < 0.05) decreased, and RESPF-C (P < 0.05) 196 increased after the test in the control group. Compared with those in the pretest, RESP-A (P < 0.05) decreased, 197 and RESPA-A (P < 0.05), RESPF-A (P < 0.05), and RESPF-C (P < 0.05) increased in the intervention group 198 after Qigong exercise, which indicate the important role of Qigong in regulating the amplitude and frequency 199 of respiration, particularly in abdominal breathing of anxious college students (Table 5). 200

201 Table 5. Effects of Qigong on the respiration of the control and intervention groups during pre- and posttest

202 conducted among anxious college students.

	Control (<i>n</i> = 19)			Intervention $(n = 18)$			
Index	Pre	Post	Change(95%CI)	Pre	Post	Change(95%CI)	P value

RESP-A	9.77±0.32	9.18±0.44*	0.58 (0.114 to 1.055)	9.31±0.48	8.88±0.37*	0.43 (0.0816 to 0.778)	0.335
RESPA-A	0.60 ± 0.25	0.70 ± 0.28	-0.10 (-0.246 to 0.046)	0.72 ± 0.28	$0.89 \pm 0.19*$	-0.18 (-0.351 to 0.007)	0.523
RESPF-A	14.10±3.81	15.62±3.89	-1.53 (-3.676 to 0.625)	13.13±3.35	16.18±4.37*	-3.05 (-5.564 to -0.540)	0.351
RESP-C	9.11±1.06	9.22±0.38	-0.11 (-0.948 to 0.726)	8.93±0.78	9.49±0.63	-0.56 (-1.133 to 0.008)	0.307
RESPA-C	0.65 ± 0.38	0.76 ± 0.40	-0.11 (-0.311 to 0.088)	0.80 ± 0.42	0.63±0.23	0.17 (-0.179 to 0.523)	0.195
RESPF-C	13.88±3.84	17.48±2.72*	-3.60 (-0.311 to 0.088)	13.30±3.15	16.53±3.95*	-3.23 (-5.856 to -0.606)	0.825

Data are presented as mean \pm SD. * represents statistically significant difference between pre- and post-test in the control and intervention group. *P* value indicates difference in variables between the control and intervention group. RESP-A, abdominal breathing; RESPA-A, depth of abdominal breathing; RESPF-A, frequency of abdominal breathing; RESP-C, chest breathing; RESPA-C, depth of chest breathing; RESPF-C, frequency of chest breathing.

208 4. Discussion

209 This study innovatively selected the combination of Yijinjing and Liuzijue from various types of Qigong exercise to illustrate the effect of breathing training in Qigong on anxiety relief. We observed that Yijinjing 210 exercises strengthen the body, prevent disease, and improve cognitive function and negative emotion, as also 211 observed previously from other studies that employed different Qigong techniques²⁶. Liuzijue emphasizes the 212 use of abdominal breathing during practice, which allows for deep and long breaths²⁷. Therefore, the addition 213 214 of Liuzijue exercises to Yijinjing can enhance the breathing depth through vocal training while retaining the easy-to-grasp and rhythmically diverse characteristics of Yijinjing, which can relieve anxiety in college 215 students. Our results revealed that the combined practice of Yijinjing and Liuzijue can effectively improve the 216 217 anxiety state and sleep quality among college students. Overall, these findings suggest that a 3-month Qigong intervention can improve anxiety status, sleep quality, and physical pain among college students with anxiety 218 219 disorders.

The occurrence of psychological disorders is closely related to physiological factors, such as TEMP and 220 skin resistance indicators associated with stress levels^{28,29}. In this study, short-term Oigong exercises can 221 considerably reduce psychological stress, as observed in previous works²⁸ that examined stress-related 222 physiological indicators and revealed reductions in heart rate and finger temperature. We also observed a 223 considerable reduction in subjective stress responses. We also showed that Qigong practice achieved and 224 maintained remarkable decreases in the heart rate, respiratory rate, and systolic blood pressure and maintained 225 long-term stress reduction. These results are supported by those of previous research, albeit the literature on 226 this point is limited²⁹. In addition, our study indicated a substantial improvement in physical pain perception 227 of anxious college students after 3-month Qigong training, which is consistent with that of a previous study³⁰; 228 Qigong exercise has also a positive effect on some people with neck pain³¹. Our study adds knowledge to the 229 understanding of Qigong as a therapeutic tool although its effects on pain relief must be further explored. In 230 general, these results can be explained as the improvement in autonomic nervous system functioning induced 231 by the relaxing effect of Qigong. 232

Our findings are in agreement with those of studies performed in the last decades, which provided 233 considerable evidence showing that Qigong improves autonomic nerves, which increases vagal regulation³²⁻ 234 ³⁴. Most studies have shown that after Qigong intervention, HRV indicators, such as time-domain indicators 235 (SDNN and RMSSD), increased, and frequency-domain indicators (LF and LF/HF) decreased³⁵, which are 236 consistent with our findings. Therefore, we hypothesized that Qigong may relieve anxiety through HRV 237 regulation, and it possibly acts on the autonomic innervation system of the heart and lungs. First, the slow 238 239 movement patterns of Yijinjing and Liuzijue can alter the autonomic system and modulate its balance toward parasympathetic dominance, which can improve HRV. Second, Qigong can improve HRV by changing the 240 breathing pattern and depth. These results are supported by those of previous studies¹⁹. Finally, the unique 241 psychological conditioning of Qigong, such as meditation, leads to a decrease in sympathetic excitability and 242 an increase in vagal excitability, which results in an improved HRV³⁵. The abovementioned hypothesis about 243 Qigong explains its beneficial use as a means of improving HRV, which will contribute to anxiety relief. 244

The adverse effects of stress and negative emotions can be counteracted through different forms of 245 breathing techniques, such as meditation and relaxation³⁶. Liuzijue Qigong typically involves training with 246 breathing techniques²⁷. Breathing techniques with equal inhalation/exhalation ratios effectively promote the 247 balance between sympathetic and parasympathetic activities and increases HRV³⁷. Abdominal breathing is an 248 effective intervention that can increase inspiratory volume and achieve an equal inhalation/exhalation ratio³⁷. 249 All these effects have been confirmed by our study, which showed the beneficial effect of two different forms 250 251 of Qigong. These results can be ascribed to the activation of the autonomic nervous system. A possible effect on the brain might have also caused the improvement of physiological parameters, as demonstrated by a study 252 on the effect of Qigong on increasing alpha-waves in electroencephalogram³⁸. The results of the present 253 research showed that the depth and frequency of abdominal breathing among anxious college students were 254 effectively improved through conscious breathing training during Qigong exercise. Therefore, breathing 255 256 techniques of Qigong can be used as first-line and supplemental treatments for anxiety, depression, and other emotional disorders. In the follow-up study, more attention should be paid to the mechanisms and differences 257 in the effects of various breathing techniques of Qigong exercise. In addition, the physiological importance of 258 breathing training in Qigong for anxious college students should be considered. Although this study provided 259 promising results, it had some limitations, e.g., its inapplicability to college students with other related 260 disorders, such as psychological disorders, frailty, or metabolic syndrome when the applicability and 261 distinction between different kinds of Oigong interventions are considered. In the future, the number of 262 subjects and duration of training intervention must be increased, and different types of Qigong must be 263 associated with various chronic diseases in the form of exercise prescriptions. 264

265 5. Conclusions and perspectives

Qigong has been studied as a means of improving psychological and associated physiological factors. 266 Compared with the more recent relaxation methods, this form of exercise is grounded in a millenary traditional 267 culture, which piques the interest of people. Qigong is accepted more by eastern (Chinese) populations, which 268 could have added a placebo effect on the results. This study revealed that Qigong exercise may have a vital 269 mood-enhancing effect on anxious college students with a 3-month training, and these changes are affected 270 by the improvement in HRV and breathing regulation. In terms of clinical relevance, this study unveiled the 271 potential effect of two Qigong styles on improving the quality of life of sedentary and anxious university 272 students and ameliorating their basic functional physiological parameters. Follow-up studies should also 273 explore the mechanisms behind such phenomena and compare the differences between traditional Qigong and 274 other exercise modalities in terms of anxiety relief. 275

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282 CONFLICT OF INTEREST

283 The authors declare no conflicts of interest.

284 DATA AVAILABILITY STATEMENT

285 The data are available from the corresponding author upon reasonable request.

286 AUTHOR CONTRIBUTIONS

Author JYS and JTZ have given substantial contributions to the conception and the design of the manuscript; author HC, JW, BL, TFL, YNX and HZ performed the experiment; author TC performed the acquisition of the data; author AC and JMD contributed to analysis and interpretation of the data. All authors
 read and approved the final version of the manuscript.

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