

• Supplementary File •

Electroacupuncture reduces posterior cingulate cortex activation and functional connectivity during food cue stimulation in overweight/obese subjects

Yang He¹, Karen M. von Deneen^{1*}, Guanya Li¹, Mingzhu Xu¹, Wenjing Zhang²,
Xiaoping Li², Yuanyuan Ren^{2*} & Yi Zhang^{1*}

¹Center for Brain Imaging, School of Life Science and Technology, Xidian University, Xi'an, Shaanxi 710071, China;

²Department of Acupuncture and Massage, Xi'an Traditional Chinese Medicine Hospital, Shaanxi University of Chinese Medicine, Xi'an, Shaanxi 710021, China

Appendix A Experimental design

In the electroacupuncture (EA) group, paramagnetic (0.25 mm × 50 mm) needles were inserted to a depth of 2.0-3.0 cm into nine main acupoints: Zhongwan (CV12), Tianshu (ST25), Xiawan (CV10), Daheng (SP15), Wailing (ST26), Liangmen (ST21), Huaroumen (ST24), Daju (ST27) and Fujie (SP14); additional distribution acupoints included Shangjuxu (ST37), Xiajuxu (ST39), Quchi (LI11), Neiting (ST44), Zhigou (TE6), Hegu (LI4) and Fenglong (ST40) (Figure 1). The acupuncturist manually rotated the needles till the tingling sensation called *De-qi* was achieved, and this stimulation lasted 1 minute. Needles placed in the bilateral Tianshu (ST25), Liangmen (ST21), Shangjuxu (ST37), and Neiting (ST44) acupoints were connected to an electrical stimulator and stimulated with a 20 Hz continuous wave for 30 minutes. The needle was manually rotated for 1 minute after the needle remained in place for 15 minutes except for the EA acupoints. In the Sham group, needles were inserted superficially to a depth of less than 5 mm and 2 cm adjacent to the acupoints selected in the EA group, but no manipulation of the needle and current stimulation were performed during the needle retention time.

Appendix B MRI scanning

Magnetic resonance imaging (MRI) scan was implemented inside of a 3T Signa Excite HD scanner. High-resolution 3D image was obtained with a magnetization-prepared rapid acquisition gradient-echo sequence with a voxel size of 1mm x 1mm x 1mm and with an AFSGE sequence (TR = 7.8 ms, TE = 3.0 ms, matrix = 256 x 256, FOV = 256 x 256 mm², slice = 1 mm, 166 slices). T2*-weighted EPI sequence was used to acquire resting-state functional images (TR = 2000 ms, TE = 30 ms, matrix = 64 x 64, FOV = 256 mm x 256 mm, angle = 90, resolution = 4mm x 4mm, slice = 4 mm, 32 axial slices).

Appendix C Imaging data processing

Imaging data were processed using SPM 12. Functional images were slice-timed, and then correction for head motion was done using a frame-wise method. EPI images underwent co-registration to a T1 anatomical image and normalization to the MNI template and re-sampled to a 3 x 3 x 3 mm³ voxel size. Images were smoothed with an isotropic Gaussian kernel (FWHM = 6 mm).

A general-linear-model (GLM), including high-calorie (HC)/low calorie (LC) food stimuli and neutral cue (NC) regressors of the conditions, was developed. Every regressor was produced by convolving the canonical-hemodynamic-response-function with a box-car function associated with the condition timing. Six realignment parameters were regressed out. Contrast images for HC vs. NC, LC vs. NC and HC vs. LC were calculated and sent to level 2 ANOVAs using the group (EA, Sham), time (Baseline, one course of treatment) and condition (HC vs. NC, LC vs. NC, HC vs. LC) factors. Findings underwent multiple comparisons via family-wise-error (FWE) corrections ($P_{FWE} < 0.05$, cluster size of $k = 50$ voxels; cluster-defining threshold of $P < 0.001$).

* Corresponding author (email: vondenk@ufl.edu, yuanyuan_ren@126.com, yizhang@xidian.edu.cn)

† Yang He and Karen M. von Deneen have the same contribution to this work.

Table 1 Demographic and clinical information of overweight/obese subjects in both electroacupuncture and Sham acupuncture groups.

	PreEA(n = 27)	PostEA(n = 27)	PreSham(n = 19)	PostSham(n = 19)	PreEA vs. PreSham		
	(Mean ± SE)	(Mean ± SE)	(Mean ± SE)	(Mean ± SE)	T	P	
Age(yrs)	33.50 ± 1.15	33.54 ± 1.67	34.63 ± 2.07	34.84 ± 2.02	-0.508	0.614	
Gender	7M/20F	7M/20F	4M/15F	4M/15F	0.146	0.703	
Weight (Kg)	86.19 ± 2.13	79.34 ± 2.20	85.08 ± 2.99	82.21 ± 2.74	0.311	0.757	
BMI (Kg/m ²)	30.85 ± 0.64	28.35 ± 0.64	31.07 ± 0.91	29.95 ± 0.80	-0.209	0.835	
WC (cm)	104.92 ± 1.79	97.48 ± 1.51	106.33 ± 2.28	102.84 ± 1.95	-0.502	0.618	
Depression (SDS)	48.26 ± 2.21	44.74 ± 2.29	48.05 ± 2.23	43.94 ± 1.99	0.064	0.949	
Anxiety (SAS)	45.15 ± 2.50	38.04 ± 1.78	44.63 ± 2.08	43.57 ± 2.51	0.149	0.882	
Craving	High-Calorie	54.26 ± 5.74	35.93 ± 4.47	45.52 ± 6.26	45.78 ± 6.59	1.04	0.304
	Low-Calorie	55.74 ± 5.09	43.89 ± 4.88	47.36 ± 7.48	49.47 ± 6.23	0.96	0.343
	Restraint eating	2.91 ± 0.11	3.73 ± 0.12	2.91 ± 0.12	3.34 ± 0.09	-0.018	0.985
DEBQ	Emotional eating	2.43 ± 0.18	2.26 ± 0.17	2.58 ± 0.15	2.25 ± 0.14	-0.634	0.529
	External eating	3.23 ± 0.12	2.90 ± 0.11	3.13 ± 0.10	2.97 ± 0.08	0.614	0.542
	Cognitive control	9.22 ± 0.77	13.52 ± 0.83	9.63 ± 0.75	11.94 ± 0.85	-0.367	0.716
TFEQ	Disinhibition	7.44 ± 0.56	5.59 ± 0.42	7.73 ± 0.71	7.31 ± 0.73	-0.328	0.745
	Hunger	6.19 ± 0.61	3.63 ± 0.52	5.74 ± 0.81	5.05 ± 0.75	0.451	0.654

Table 2 Interaction effects (Group × Time) and Post-hoc tests for behavioral measurements.

	ANOVA				Post-hoc tests						
	Interaction effect		PreEA vs. PreSham		PostEA vs. PostSham		PreEA vs. PostEA		PreSham vs. PostSham		
	F	P	T	P	T	P	T	P	T	P	
Weight (Kg)	19.615	<0.001	0.311	0.757	-0.821	0.416	10.768	<0.001	4.997	<0.001	
BMI (Kg/m ²)	17.629	<0.001	-0.209	0.835	-1.576	0.122	10.909	<0.001	5.177	<0.001	
WC (cm)	6.049	0.018	-0.502	0.618	-2.198	0.033	6.647	<0.001	3.415	0.003	
Depression (SDS)	0.03	0.864	0.064	0.949	0.248	0.806	1.545	0.134	1.676	0.111	
Anxiety (SAS)	3.406	0.072	0.149	0.882	-1.854	0.07	3.029	0.005	0.517	0.612	
Craving	High-Calorie	4.172	0.047	1.04	0.304	-1.285	0.206	3.264	0.003	<0.001	1
	Low-Calorie	2.319	0.135	0.96	0.343	-0.714	0.479	2.373	0.025	-0.253	0.803
	Restraint eating	3.17	0.082	-0.018	0.985	2.321	0.025	-4.836	<0.001	-4.206	0.001
DEBQ	Emotional eating	1.21	0.277	-0.634	0.529	-0.011	0.991	1.835	0.078	3.066	0.007
	External eating	0.918	0.343	0.614	0.542	-0.498	0.621	2.3	0.03	1.85	0.081
	Cognitive control	1.929	0.172	-0.367	0.716	1.294	0.203	-3.996	<0.001	-3.139	0.006
TFEQ	Disinhibition	2.972	0.092	-0.328	0.745	-2.18	0.035	3.312	0.003	0.715	0.484
	Hunger	3.604	0.064	0.451	0.654	-1.619	0.113	3.286	0.003	1.753	0.097
	EA (n = 27)				Sham (n = 19)				EA vs. Sham		
	(Mean ± SE)				(Mean ± SE)				T	P	
BMI(Kg/m ²)	2.50 ± 0.23				1.12 ± 0.22				4.199	<0.001	
EWL%	40.14% ± 5.59%				14.55% ± 3.08%				4.013	<0.001	

Table 3 The average Acupuncture Sensation Scale (*De-qi*) after the first acupuncture and the last acupuncture

<i>De-qi</i>	EA (n = 27) (Mean ± SE)	Sham (n = 19) (Mean ± SE)	EA vs. Sham	
			T	<i>P</i>
Soreness	3.32 ± 0.45	1.57 ± 0.53	2.52	0.016
Numbness	3.11 ± 0.44	1.03 ± 0.38	3.606	<0.001
Fullness	3.86 ± 0.40	1.29 ± 0.35	4.641	<0.001
Coolness	1.52 ± 0.44	0.34 ± 0.27	2.306	0.026
Warmth	1.83 ± 0.44	0.16 ± 0.12	3.655	0.001
Sharp Pain	3.40 ± 0.45	2.32 ± 0.41	1.714	0.094
Dull Pain	2.88 ± 0.46	0.73 ± 0.24	4.115	<0.001
Heaviness	2.56 ± 0.40	0.44 ± 0.25	4.453	0.001
Tingling	2.96 ± 0.46	1.94 ± 0.39	1.607	0.115
Itching	1.52 ± 0.35	0.32 ± 0.16	3.138	0.003
Aching	2.27 ± 0.38	0.29 ± 0.17	4.802	<0.001
Pressure	1.40 ± 0.32	0.11 ± 0.11	3.869	<0.001
Hunger	1.25 ± 0.26	0.24 ± 0.14	3.355	0.002

Table 4 The peak activation coordinates and corresponding Brodmann areas of the activated region

Region	BA	Cluster size	Peak coordinates		
			X	Y	Z
dACC	24, 32	132	0	18	33
PCC	23	83	3	-9	36
Postcentral	3, 4	130	24	-30	60

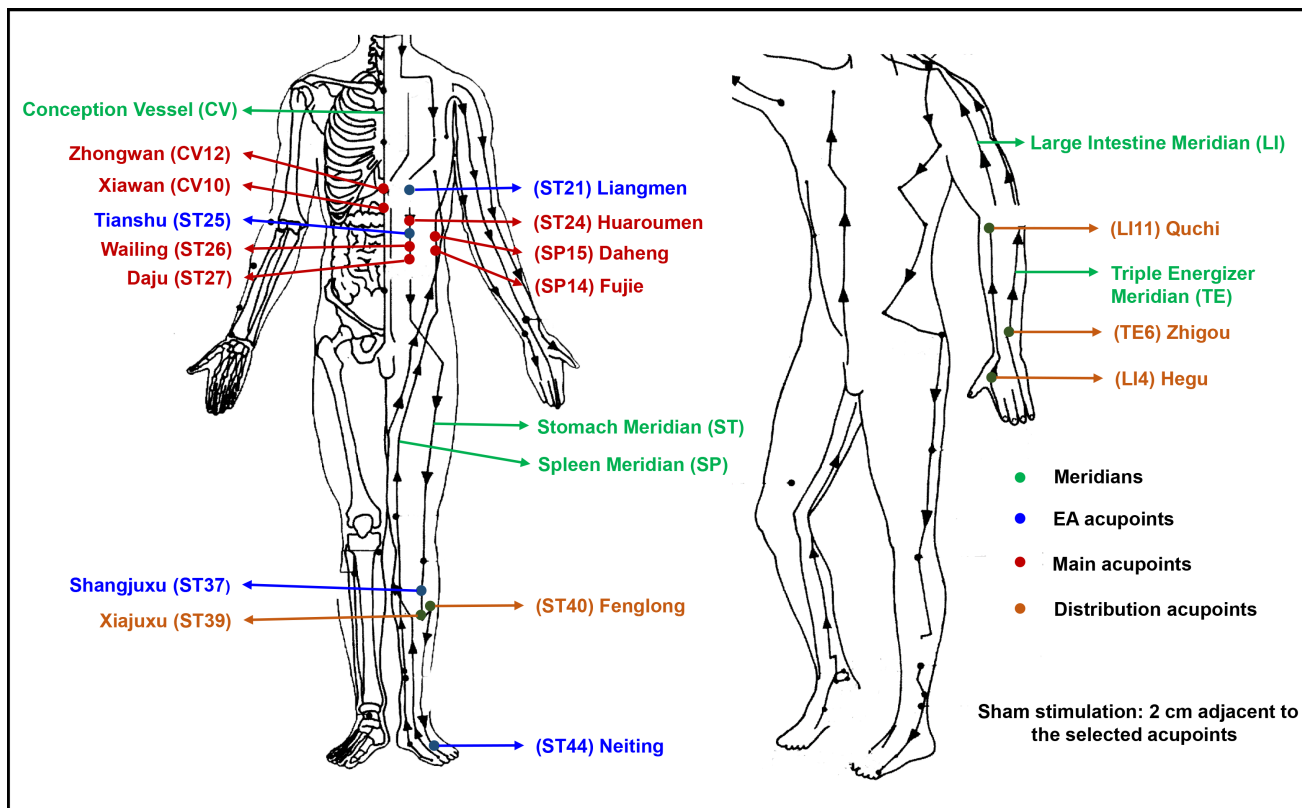


Figure 1 Selected acupoints