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Research Article
Influence of Laser Puncture on Endothelial Dysfunction on Hypertensive Patients

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Abstract

Background and Objective: The purpose of the study was to investigate the influence of laser applied on acupuncture points in endothelial dysfunction on hypertensive obese patients. **Materials and Methods:** Sixty patients from both genders aged between 40-50 years old were assigned into two groups equal in number. Group A: Received a program of aerobic exercises and laser on acupuncture points 16 J per point and group B: Received a program of aerobic exercises and sham laser (placebo) on acupuncture points. The program was 2 times per week for 24 weeks. Urinary Nitric Oxide (NO) and blood superoxide dismutase (SOD) were measured at the beginning and at the end of the study. **Results:** The results of study group revealed that there was a significant increase in nitric oxide (81.82%) and superoxide dismutase (78.72%) levels, which means reduction in oxidative stress and improvement in endothelial function. **Conclusion:** It could be concluded that for hypertensive patients using laser puncture two times per week improved their nitric oxide and superoxide dismutase levels which is important antioxidants in the body. Laser applied on acupuncture points improve general health.

Key words: Hypertension, endothelial dysfunction, laser, acupuncture, nitric oxide, superoxide dismutase

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Hypertension is the most common cardiovascular disease (CVD), affecting about one billion individuals worldwide[1].

There are regulation of vascular tone in response to the vascular endothelium which is a very active organ through the effects of synthesized mediators, predominantly Nitric Oxide (NO), endothelial nitric oxide synthase (eNOS) and superoxide. The NO is abundantly evident in normally functioning vasculature where it acts as a vasodilator[2].

Endothelial dysfunction is present in essential hypertension in both resistance and conduit arteries and constitutes an early independent predictor of cardiovascular events[3].

Low Level Laser Therapy (LLLT) is a 100% safe technique that functions at the atomic level by carrying electrons on the soliton waves to needy areas without producing any side effects. Loss of electrons leads at the atom level leads to cellular dysfunction, tightening of smooth muscles, inflammation, less production of nitric oxide which allows smooth muscles in the artery walls to remain tight resulting in hypertension[4].

The specific aim of the current study was to determine the influence of laser puncture in endothelial dysfunction on hypertensive patients.

There's a lack of quantitative knowledge in the published literatures and studies related to the effect of laser puncture on endothelial dysfunction in hypertensive patients. So, the result of this study may be of clinical value to physicians, physical therapists and patients.

MATERIALS AND METHODS

Sixty patients from both genders (36 men and 24 women) with pre and stage 1 hypertensive (the range 130-149 mmHg Systolic Blood Pressure (SBP) and Diastolic Blood Pressure (DBP) in the range 85-99 mmHg) they were of class (I and II) of obesity according to BMI equation:

$$\text{BMI} = \frac{\text{Weight (kg)}}{\text{Height (m}^2\text{)}} = 30.0{:}39.9 \text{ kg m}^{-2}$$

participated in this study from internal medicine department and obesity unit of El Sahel Teaching Hospital, Egypt and their ages were ranged from 40-50 years.

Inclusion criteria of the study group:

- The selected patients were diagnosed clinically with pre and stage 1 hypertensive (the range 130-149 mmHg systolic BP and diastolic BP in the range 85-99 mmHg)
- Their Body Mass Index (BMI) within the range of obesity class (I and II) of obesity according to BMI equation:

$$\text{BMI} = 30.0{:}39.9 \text{ kg m}^{-2}$$

- The age of selected subjects was ranged from 40-50 years

Exclusion criteria of the study:

- Had previous history of cigarette smoking or alcohol drinking
- Pulmonary and vascular diseases
- Endocrine disorders
- All neurological disorders or balance problem e.g., hemiplegic and epilepsy
- Individuals with severe coronary disease (continuous angina)
- Conduction abnormalities on electrocardiogram (ECG)
- Uncontrolled diabetes mellitus

Design: Pre test post test design was used in this study. Sixty patients with hypertension were assigned randomly into two equal groups A and B.

Group A: Thirty patients with pre and stage 1 hypertensive received a program of aerobic exercises training on treadmill and laser on acupuncture points.

Group B: Thirty patients with pre and stage 1 hypertensive received a program of aerobic exercises training on treadmill and sham laser on acupuncture points.

Instrumentation for assessment:

- Mercurial sphygmomanometer and stethoscope that was used for measuring BP for all patients participated in the study to select them
- Weight and height scale that was used in order to calculate BMI
- **Spectrophotometer:** The UV 2300 Spectrophotometer with resolution of 1.5 nm that was used for measuring SOD

Instrumentation for treatment:

- **Laser device:** Gymna I.R laser device made in Belgium was used in the study with wave length 904 nm, maximal power was 27 mw and frequency ranged from 1 Hz up to 1000 Hz. The device was calibrated by the manufacturer routinely and it was calibrated before starting the study

- **Electrical motorized treadmill:** Used to conduct the aerobic exercise

Procedure: All patients signed informed consent about the purpose and nature of the study. A complete history was being taken and physical examination was being conducted for each patient before participation in the study.

Evaluation procedure: The evaluation procedure had been done for all patients in the two groups before starting the program:

- Blood pressure measurement
- BMI
- Measurement of superoxide dismutase activity
- Measurement of urinary nitrates/nitrites SOD and NO had been measured after 24 weeks of treatment

Treatment procedure: Treatment had been carried out in physical therapy outpatient clinic of El-Sahel teaching hospital. The program of the study was given for two groups two sessions per week for 24 weeks.

Group A: Patients received laser with 16 J cm^{-2} for 4 min for each point over the following acupuncture points:

- **Distal point: Large intestine 4 (LI 4):** To locate, squeeze the patient thumb against the base of the index finger. The point is located on the highest point of the bulge of the muscle, level with the end of the crease
- **Proximal point: Large intestine 11 (LI 11):** To locate, bend patient forearm with my hand towards his neck, the point is located at the end of the crease at the elbow. This is halfway up the side of the arm

Aerobic exercise training program: It were conducted twice a week for 24 weeks (total of 48 treatments), training used the electrical motorized treadmill. The aerobic exercise training program was subdivided into three phases: The warm-up phase (5-10 min), the main-exercise phase: (15-20 min) intensity of the exercise was 60-70% of maximum heart rate (MHR) and the cool-down phase: 5-10 min.

Control group B: Patients received sham laser (Placebo laser) acupuncture points followed by aerobic exercise training program the same as which applied on the study group A.

Data analysis: Statistical analysis of the data was performed using SPSS software for medical statistics version 20. The significance level was set at $p<0.05$. To test the study hypothesis the following statistical methods were used:

- Descriptive statistics (mean and standard deviation) for NO and SOD analysis measurement for two groups
- Percentage of difference of post treatment as compared with pre one
- Inferential statistical by using paired t test for the same group and unpaired t test between the two groups

RESULTS

There was no statistical significant difference of mean of age, BMI, SBP and DBP between study group and control group pre-treatment (p values $p>0.05$) which mean of study group age was (45.10±3.99 year) and control group was (44.60±3.62 year) p value = 0.614, BMI in study group was (34.04±3.12 kg m^{-2}) and control group was (34.62±3.53 kg m^{-2}) p value = 0.591, SBP in study group was (137.83±6.11 mm Hg) and control group was (138.80±5.29 mm Hg) p value = 0.515, DBP in study group was (92.00±5.11 mm Hg) and control group was (91.90±4.29 mm Hg) p value = 0.935.

Table 1 of study group represents the NO (μmol L^{-1}) ranged from 0.84-2.80 μmol L^{-1} with a mean value (1.32±0.45 μmol L^{-1}) for pre-treatment, while it ranged from 1.20-4.70 μmol L^{-1} with a mean value (2.41±0.89 μmol L^{-1}) for post-treatment. The statistical analysis of the mean differences of NO (μmol L^{-1}) by paired t-test between pre and post-treatment revealed that a significant difference ($p = 0.0001$ and $p<0.05$) in NO (μmol L^{-1}) with a percentage of improvement equal to (81.82%).

Table 2 of study group represents the SOD (U L^{-1}) ranged from 0.11-2.50 U L^{-1} with a mean value (0.47±0.47 U L^{-1}) for pre-treatment, while it ranged from 0.08-1.70 U L^{-1} with a mean value (0.84±0.70 U L^{-1}) for post-treatment. The statistical analysis of the mean differences of SOD (U L^{-1}) by paired t-test between pre and post-treatment revealed that a significant difference ($p = 0.0001$ and $p<0.05$) in SOD (U L^{-1}) with a percentage of improvement equal to (78.72%).

Table 3 of control group represents the NO (μmol L^{-1}) ranged from 0.87-2.73 μmol L^{-1} with a mean value (1.37±0.41 μmol L^{-1}) for pre-treatment, while it ranged from 1.01-4.00 μmol L^{-1} with a mean value (2.20±0.56 μmol L^{-1}) for post-treatment. The statistical analysis of the mean differences of NO (μmol L^{-1}) by paired t-test between pre and post-treatment revealed that a significant difference ($p = 0.0001$ and $p<0.05$) in NO (μmol L^{-1}) with a percentage of improvement equal to (60.58%).

Table 1: Mean values between pre and post nitric oxide (μmol L^{-1}) in study group

	Nitric oxide (μmol L^{-1})		
Items	Pre-treatment		Post-treatment
Mean±Standard Deviation	1.32±0.45		2.41±0.89
Minimum to maximum	0.84-2.80		1.20-4.70
t-value		9.035	
Level of significance (p-value)		0.0001	
Significance (p<0.05)		S	
Improvement (%)		81.82 ⇧	

Table 2: Mean values between pre and post superoxide dismutase (U L^{-1}) in study group

	Superoxide dismutase (U L^{-1})		
Items	Pre-treatment		Post-treatment
Mean±Standard Deviation	0.47±0.47		0.84±0.70
Minimum to maximum	0.11-2.50		0.08-1.70
t-value		6.635	
Level of significance (p-value)		0.0001	
Significance (p<0.05)		S	
Improvement (%)		78.72 ⇧	

Table 3: Mean values between pre and post nitric oxide (μmol L^{-1}) in control group

	Nitric oxide (μmol L^{-1})		
Items	Pre-treatment		Post-treatment
Mean±Standard Deviation	1.37±0.41		2.20±0.56
Minimum to maximum	0.87-2.73		1.01-4.00
t-value		8.317	
Level of significance (p-value)		0.0001	
Significance (p<0.05)		S	
Improvement (%)		60.58 ⇧	

Table 4 of control group represents the SOD (U L^{-1}) ranged from 0.09-1.30 U L^{-1} with a mean value (0.42±0.36 U L^{-1}) for pre-treatment, while it ranged from 0.16-1.65 U L^{-1} with a mean value (0.63±0.46 U L^{-1}) for post-treatment. The statistical analysis of the mean differences of SOD (U L^{-1}) by paired t-test between pre and post-treatment revealed that a significant difference (p = 0.0001 and p<0.05) in SOD (U L^{-1}) with a percentage of improvement equal to (50.00%).

As observed in Table 5, mean pre-treatment of NO (μmol L^{-1}) in study group was (1.32±0.45 μmol L^{-1}) and pre-treatment of NO (μmol L^{-1}) in control group was (1.37±0.41 μmol L^{-1}). The statistical analysis of the mean differences of pre-treatment NO (μmol L^{-1}) by independent t-test revealed that there was no statistical significant difference (p = 0.441 and p>0.05) in mean value of pre-treatment NO (μmol L^{-1}) between both groups. Mean post-treatment of NO (μmol L^{-1}) in study group was (2.41±0.89 μmol L^{-1}) and post-treatment of NO (μmol L^{-1}) in control group was (2.20±0.56 μmol L^{-1}). The statistical analysis of the mean differences of post-treatment NO (μmol L^{-1}) by independent t-test revealed that there was

Table 4: Mean values between pre and post superoxide dismutase (U L^{-1}) in control group

	Superoxide dismutase (U L^{-1})		
Items	Pre-treatment		Post-treatment
Mean±Standard Deviation	0.42±0.36		0.63±0.46
Minimum to maximum	0.09-1.30		0.16-1.65
t-value		7.347	
Level of significance (p-value)		0.0001	
Significance (p<0.05)		S	
Improvement (%)		50.00 ⇧	

Table 5: Comparative analysis of nitric oxide (μmol L^{-1}) between study group and control group

	Nitric oxide (μmol L^{-1})	
Items	Pre-treatment	Post-treatment
Study group	1.32±0.45	2.41±0.89
Control group	1.37±0.41	2.20±0.56
t-value	0.441	1.052
Level of significance (p-value)	0.660	0.045
Significance (p<0.05)	NS	S

Table 6: Comparative analysis of superoxide dismutase (U L^{-1}) between study group and control group

	Superoxide dismutase (U L^{-1})	
Items	Pre-treatment	Post-treatment
Study group	0.47±0.47	0.84±0.70
Control group	0.42±0.36	0.63±0.46
t-value	0.476	1.160
Level of significance (p-value)	0.636	0.043
Significance (p<0.05)	NS	S

statistical significant difference (p = 0.045 and p<0.05) in mean value of post-treatment NO (μmol L^{-1}) between both groups.

Table 6 shows the mean pre-treatment of SOD (U L^{-1}) in study group was (0.46±0.47 U L^{-1}) and pre-treatment of SOD (U L^{-1}) in control group was (0.42±0.36 U L^{-1}). The statistical analysis of the mean differences of pre-treatment SOD (U L^{-1}) by independent t-test revealed that there was no statistical significant difference (p = 0.636 and p>0.05) in mean value of pre-treatment SOD (U L^{-1}) between both groups.

Mean post-treatment of SOD (U L^{-1}) in study group was (0.84±0.70 U L^{-1}) and post-treatment of SOD (U L^{-1}) in control group was (0.63±0.46 U L^{-1}). The statistical analysis of the mean differences of post-treatment SOD (U L^{-1}) by independent t-test revealed that there was statistical significant difference (p = 0.043 and p<0.05) in mean value of post-treatment SOD (U L^{-1}) between both groups.

DISCUSSION

The results of the current study showed that there was a significant difference between study and control group post treatment in NO and SOD values.

Hamed and Al Maghraby[5] studied the effectiveness of laser acupoint therapy and exercise program on oxidative stress and antioxidant response in mild essential hypertensive patients and they concluded that a combination therapy of both laser therapy and exercise program for hypertensive patients will produce more reduction of blood pressure as well as reduction of peripheral vascular resistance in response to improving endothelial dysfunction.

In agreement with the results of the current study the results of Park *et al.*[6] who investigate the acute effect of acupuncture on endothelial dysfunction in patients with hypertension and he concluded that the acute treatment of acupuncture in hypertensive patients improves endothelial dysfunction. The beneficial effect of acupuncture could be of clinical importance to prevent the progression of cardiovascular diseases in hypertensive patients.

The results of the current study agree with the results of Lai *et al.*[7] who concluded that an increase in antioxidant enzymes in the spontaneously hypertensive rats subject to acupuncture and up-regulated SOD, which may provide partial explanation for the antihypertensive effect of acupuncture.

Also Dan[8] suggested that the hypotensive effect of acupuncture is much more potent than that of placebos and is comparable with that of certain conventional hypotensive agents. In addition, acupuncture is often effective for relieving subjective symptoms and it has no side-effects. He reported that the influence of acupuncture on hypertension might be related to its regulatory effect on the level of serum nitrogen monoxide.

Zhang *et al.*[9] reported that infrared laser irradiation contributes to correction of the elevated level of hydro peroxidase and increased level of superoxide dismutase.

Also Hwang *et al.*[10] investigated whether Electro Acupuncture (EA) could reduce early stage hypertension by examining Nitric Oxide (NO) levels in plasma and Nitric Oxide Synthase (NOS) levels in the mesenteric resistance artery; they concluded that EA could attenuate the blood pressure elevation of Spontaneously Hypertensive Rats (SHR), along with enhancing NO/NOS activity in the mesenteric artery in SHR.

Also, Xiong *et al.*[11] demonstrated a relaxation of vascular smooth muscle cell mediated via release of endothelium derived nitric oxide from arteries irradiated by visible light laser.

The results of the present study come in line with[12] who studied the effect laser stimulation in anesthetized rats and found that laser stimulation at the left or right pericardium (PC-6), stomach (ST-36) and liver (LR-3) as well as at the right LI-11 can modulate the cardiovascular functions in anesthetized rats.

Also Sutalangka *et al.*[13] stated that acupuncture enhanced SOD and catalase (CAT) activities and suppressed acetylcholinesterase activity in the hippocampus.

The results of the present study come in line with Schuller and Neugebauer[14] who evaluated the evidence for laser acupuncture in hypertension. A positive effect had been assumed in systolic hypertension and laser acupuncture was advantageous in terms of side effects compared to classical acupuncture techniques. They reported changes of the neuronal and inducible nitric oxide synthase (nNOS and iNOS) and their ribonucleicacid messenger (RNAs) in the rostral ventrolateral medulla (RVLM) of stress-induced hypertensive increased significantly.

Also Mittermayr *et al.*[15] stated that the laser irradiation improves local tissue perfusion in a controlled manner stimulating NO release.

Also Pandey *et al.*[4] stated that the laser system is a very effective and safe system of treating stage I and II hypertension.

In agreement with the results of the current study the results of Ma *et al.*[16] who assessed the therapeutic outcomes of combining acupuncture and aerobic exercises to treat hypertension and found that all patients showed improvement in quality of life. Systolic blood pressure was controlled better by acupuncture while DBP improved following aerobic exercises, while both methods induced the best outcomes. But he stated that acupuncture effect on BP is due to the release of various chemicals of the central nervous system.

CONCLUSION

In conclusion using laser acupuncture on LI4 and LI11 points in pre and stage 1 hypertensive patients 2 times\week for 24 weeks results in significant increase in NO and SOD level. So, laser acupuncture can be recommended for those patients to improve endothelial dysfunction.

Adding laser applied on acupuncture points to aerobic exercise is more effective in improving endothelial dysfunction of hypertensive patients.

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