

The effect of 6 weeks swimming training on plasma antioxidants activity in diabetic rats

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ABSTRACT

The purpose of this study was to evaluate the effect of 6 weeks endurance swimming training on plasma antioxidants in diabetic rats. For this purpose, 30 male wistar rats were divided into three groups, one group of Diabetic control rats (DC, n=10), one group of Healthy control (HC, n=10), one group of diabetic and Swimming training (S, n=10). Results were analyzed using the one-way ANOVA followed by a Tukey test. Significance level was 0.05. The diabetic control group exhibited a significant decrease in body weight ($P<0.05$). S and HC group have shown significant increase in cardiac antioxidant enzymes than DC group. The present results indicate that endurance swimming training is useful for treating metabolic diseases related to obesity and diabetes.

Key words: Endurance swimming training, Fenugreek seeds extract Heart antioxidants

INTRODUCTION

Diabetes mellitus is a group of metabolic diseases characterized by hyperglycemia resulting from defective insulin secretion or resistance insulin action, or both [1]. Oxidative stress may constitute a focal point for multiple therapeutic interventions, and for therapeutic synergy. Hyperglycemia may perturb cellular antioxidant defense systems and damage cells. Free radicals are formed disproportionately in diabetes by glucose oxidation, non-enzymatic glycation of proteins, and the subsequent oxidative degradation of glycated proteins. Oxidative stress plays an important role in the etiology of diabetes and diabetic complication [2]. Ihera et al. [1999] examined oxidative stress marked in diabetic rats and found increased reactive oxygen species (ROS) in pancreatic islets [30]. Cells continuously produce free radicals and ROS as part of metabolic processes. These free radicals are neutralized by an elaborate antioxidant defense system consisting of enzymes such as catalase, superoxide dismutase, and glutathione peroxidases.

Diabetes is associated with significant oxidative stress, and oxidative damage to tissues may be a contributory factor in several diabetic complications [4]. Normal levels of the antioxidant defense mechanism are not sufficient for the eradication of free radical induced injury. Therefore, the administration of antioxidants from a natural origin has a promising role to play.

The therapeutic use of physical exercise for diabetes treatment has been promoted since 600 B.C. before the discovery of insulin in 1922. Much controversy exists concerning the effects of endurance training on the oxidative status and antioxidant defense systems of the myocardium as decrease, increase, or even remain unchanged (for a comprehensive review see reference of 5). Some controversy might arise from the different methodologies used for

determinations, and differences in the models employed (running vs. swimming, rats vs. mice, male vs. female). Although insulin treatment and other chemical therapies can control the disease to various extents, the complications are very common; whose pathologic base is microangiopathy. Among various forms of treatments for diabetic mellitus, exercise and diet are of vital importance.

The present study was aimed to evaluate the effect of endurance swimming training and fenugreek seed extract combination on plasma glucose and heart tissue oxidative stress in diabetic rats and this study is the first study that evaluates the effect of physical exercise and fenugreek seeds extract combination on cardiac antioxidants in diabetic rats.

MATERIAL AND METHODS

Animals

Thirty male wistar albino rat, weighting 200-250 g, and averaging 12 weeks old were used in this study. They house in metal cage under standard laboratory condition (12:12-h light-dark cycle and were fed regular pellets and distilled water ad libitum. The room had a temperature of 20–25°C, humidity of 50–60%, and average illuminance of 150–200 lux in the daytime. The rats were randomly divided into three groups: 1. diabetic and swimming training (S) (n=10), 2. Health control (HC) (n=10), 3. Diabetic control (DC) (n=10) this group received normal saline (5 ml/kg B.W). Saline was treated orally by gastric gavage.

Induction of diabetes

After fasting for 12 hours, the animals received an intraperitoneal injection (60 mg/kg body weight) of streptozotocin (STZ, Sigma, St. Louis, USA), diluted in 1.0 ml of sodium citrate buffer (0.1 M, pH 4.5). Seven days after application of STZ and fasting for 12 hours, blood glucose was measured. Blood samples were collected by tail nipping and assessed for glucose by an electronic glucometer. Animals with levels of fasting blood glucose above 300 mg\dl were considered diabetic. Fasting blood glucose and body weight were monitored at first and end in the experimental period.

Endurance training program

Swimming exercise training protocol was conducted in 2 phases, adaptation and training. The adaptation phase consisted of the first 7 days of training. On the first day, the animals exercised in a round plastic tank (140x60x45 cm and water temperature about 34-36°C) for 10 minutes. The exercise period was extended by 5 minutes every day until the rats could swim for 40 minutes. The training phase consisted of swimming 40 min/day, 5 days/week for a total of 6 weeks.. Swimming exercise was selected because it did not cause foot injuries, and is physically less traumatic for the animal.

Plasma preparation

At the end of the training programs, 24 h after the last exercise-training session and 12 h fasting, the rats were weighed, sacrificed by decapitation and then Blood samples were obtained from heart of rats, whole blood were collected in an EDTA tube. The blood was centrifuged for 10 minutes at 3000 rpm in 4 °C. Plasma was separated carefully in a number of ependrooff tubes and then stored at -80°C until analyze time.

SOD Activity

Superoxide dismutase (SOD) activity was determined using a RANSOD kit (Randox labs. Crumlin, UK) according to Delmas-Beauvieux et al [12].SOD activity was measured at 505 nm on a spectrophotometer on supernatant.

CAT Activity

Catalase activity (CAT) was measured as previously described by Aebi [13]. The decomposition of H₂O₂ was followed directly by the decrease in absorbance at 240 nm and 20 °C.

GPX Activity

Glutathione peroxidase (GPX) activity was determined using a RANSEL kit (Randox labs.), according to the method of Paglia and Valentine [14].

Statistical Analysis

Results are presented as means ± standard deviation and were analyzed using One- Way ANOVA with Tukeys Post Hoc comparisons. A probability of 0.05 or less was considered as statistically significant.

RESULTS

Body weight

There was no significant difference in the groups at the beginning of the study. In this study shown body weight reduced in all of the groups ($P < 0.05$) exception of HC group (Table 1).

Table 1: The effect of 6 weeks swimming training on body weight

weeks groups	W1 M±SD	W2 M±SD	W3 M±SD	W4 M±SD	W5 M±SD	W6 M±SD
S	204.1±13.95	201.3±13.23	192.6±9.95	180.3±7.83	177.3±7.38	165.2±8.07*
HC	210.4±9.3	212.8±8.7	215.2±8.8	215.3±6.9	217.4±7.3	222.2±9.1
DC	200.7±16.94	192±16.24	181.8±18.43	170.1±17.94	161±15.89	136.7±14.17*
P-value	0.61	0.59	0.13	0.04	0.007	0.002

S: Swimming training, HC: Health control, DC: Diabetic control. * Significant decrease

Results shown that there was significant difference plasma SOD, CAT and GPX in DC than other groups ($P < 0.05$) (Table 2).

Table 2: The effect of swimming training on plasma SOD, CAT and GPX

		Df	F	Sig
SOD (U/mg protein)	Between Groups	4	19.657	0.005
CAT (U/mg protein)	Between Groups	4	18.156	0.009
GPX(U/mg protein)	Between Groups	4	5.578	0.02

Antioxidants activity

SOD activity

The results have showed that there were significant differences between the groups in plasma SOD; S group than DC group ($P=0.03$), HC group than DC group ($P=0.005$), S group than HC group ($P=0.02$) (Figure 1).

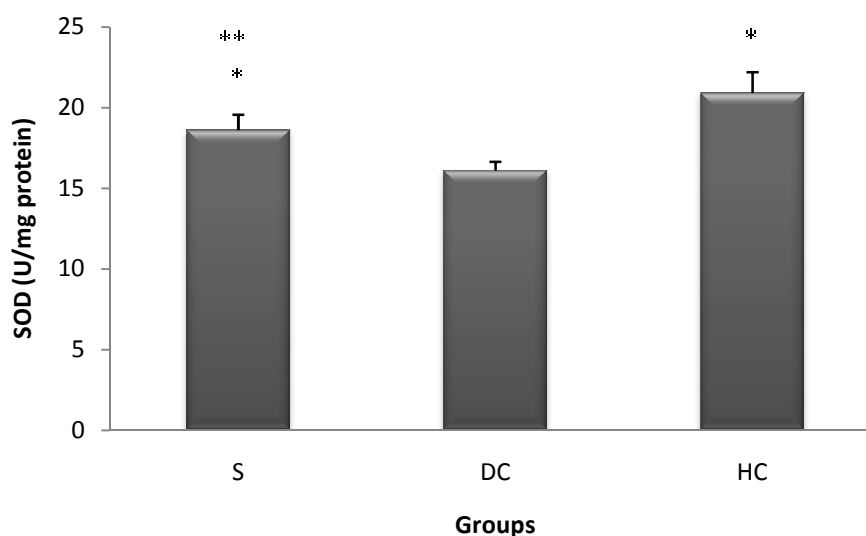


Figure 1: The effect of swimming training on plasma SOD.

S: Swimming training, HC: Health control, DC: Diabetic control. * Values are statistically significant at $p < 0.05$ in S and HC than DC. ** Values are statistically significant at $p < 0.05$ in S than HC.

CAT activity

The results have showed that there were significant differences between the groups in plasma CAT; S group than DC group ($P=0.04$), HC group than DC group ($P=0.001$), S group than HC group ($P=0.009$). Thus, significant increase observed at all of the groups' than DC group (Figure2).

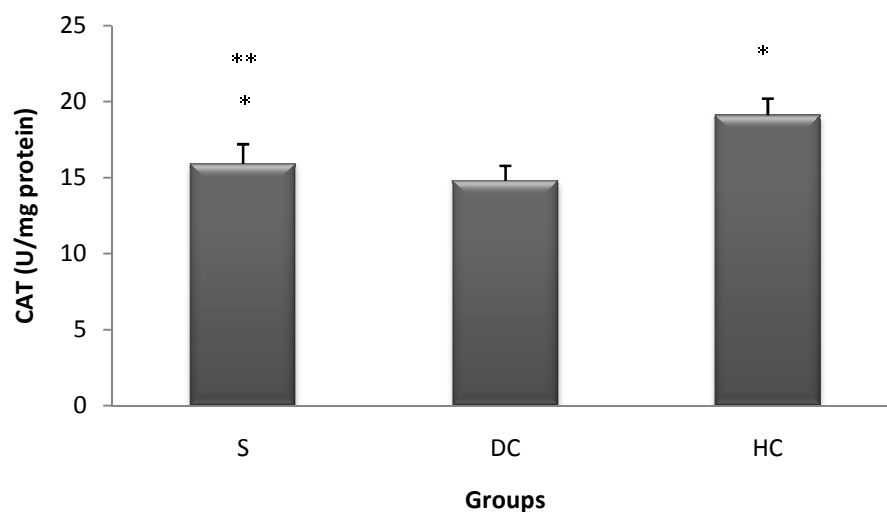


Figure 2: The effect of swimming training on plasma CAT.

*S: Swimming training, HC: Health control, DC: Diabetic control. * Values are statistically significant at $p < 0.05$ in S and HC than DC. ** Values are statistically significant at $p < 0.05$ in S than HC.*

GPX activity

The results have showed that there were significant differences between the groups in cardiac GPX; S group than DC group ($P=0.001$), HC group than DC group ($P=0.000$), but there wasn't significant difference between other groups ($P>0.05$) (Figure 3).

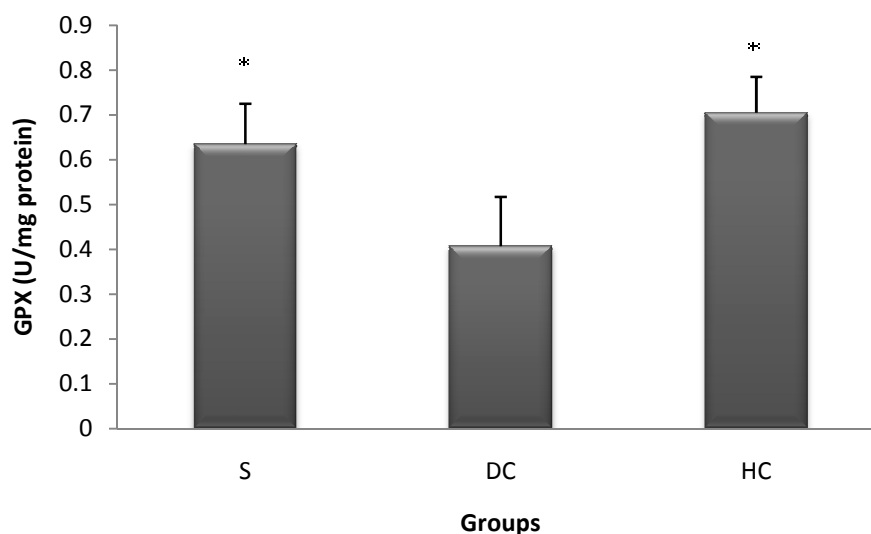


Figure 3: The effect of swimming training on plasma GPX

*S: Swimming training, HC: Health control, DC: Diabetic control. * Values are statistically significant at $p < 0.05$ in S and HC than DC.*

DISCUSSION

The current study determined the effect of swimming training on plasma antioxidants in diabetic rats. We showed swimming training lead to significant increase in plasma SOD, CAT and GPX in diabetic rats than diabetic control ($P<0.05$).

Diabetics and experimental animal models exhibit high oxidative stress due to persistent and chronic hyperglycemia, thereby deplete the activity of anti-oxidative defense system and thus promote free radicals generation [15]. Oxidative stress recently been shown to be responsible, at least in part, for the β -cell dysfunction caused by glucose toxicity. Under hyperglycemia, production of various reducing sugars such as glucose-6-phosphate and fructose

increases through glycolysis. During this process, reactive oxygen species (ROS) are produced and cause tissue damage [16, 17].

Swimming was chosen as a suitable model since it is a natural behavior of animals. The method causes less mechanical stress and injury, and leads to a better redistribution of blood flow among tissues without significant variations in cardiac output and heart rate which in turn may minimize the magnitude of injury caused due to the generation of ROS [18]. Regular exercise is recommended to diminish the fat reserves and to control the glucose levels in diabetic individuals [19]. Hyperglycemia increases the production of reactive oxygen species through protein glycation [20] and patients with type 1 diabetes mellitus have higher plasma concentrations of free radicals than healthy individuals [21]. In this way, increased glucose level is an important factor implicated with the development of the diabetes-associated complications.

Endurance training increases the energy supply to tissues in activity, resulting in oxidative stress conditions [22, 23]. Other studies have reported a similar behavior in different tissues [24]. Thus, an efficient antioxidant system is necessary to attack the free radicals produced during exercise. Thus, we believe that diabetic hyperglycemia is an important factor modulating oxidative stress. Some studies have evaluated the adaptive response of these systems to a long-term training protocol and shown increase in CAT, GPX and SOD activities that is consistent with our results [25] but they used from treadmill for endurance training but we used from swimming training that reduced injury in subjects.

Previous investigations have shown decreases [25, 26] and no changes [27] that are inconsistent with our results or even increases [28, 29] in antioxidant enzyme after training that are consistent with our results. Differences in these factors might explain the controversial results about antioxidants activity in response to training. Results have shown that changes in antioxidant activity in various tissues follow different pattern, that the pattern of these changes is not yet known. The overall, Inconsistent results in this studies could be due to some factors such as Age, sex and animals race, different techniques in the assessment, type of exercise, duration and intensity of exercise and the tissue that is used.

CONCLUSION

Overall, the present study provides evidence that the swimming training is capable of influencing plasma antioxidant enzymes in diabetic rats. We showed that this training increase plasma antioxidant enzymes that it can lead to decreases cell apoptosis.

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