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ORIGINAL ARTICLE

Effects of Fenugreek Seed Extract and Swimming Endurance Training on Plasma Glucose and Cardiac Antioxidant Enzymes Activity in Streptozotocin-induced Diabetic Rats

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Abstract
Objective: Diabetes mellitus is a group of metabolic diseases characterized by chronic hyperglycemia condition resulting from defective insulin secretion or resistance insulin action, or both. The purpose of this study was to evaluate the effect of 6 weeks swimming training and *Trigonella foenum-graecum* seed (fenugreek) extract, alone and in combination, on plasma glucose and cardiac antioxidant enzyme activity of streptozotocin-induced diabetic rats.
Methods: Fifty male Wistar rats were divided into five groups: diabetic control (DC, $n = 8$); healthy control (HC, $n = 11$); swimming training (S, $n = 11$); swimming training + fenugreek seed extract (1.74 g/kg body weight; SF1, $n = 11$); and swimming training + fenugreek seed extract (0.87 g/kg body weight; SF2, $n = 9$). Streptozotocin was used for the induction of diabetes. Results were analyzed using one-way analysis of variance followed by Tukey test.
Results: In comparison with the DC group, all groups exhibited a significant decrease in body weight ($p < 0.05$), except for the HC group. SF1 and HC groups showed significant decreases in plasma glucose levels compared with the DC group ($p < 0.05$). S, SF1, SF2, and HC groups showed significant elevations in cardiac antioxidant enzymes activity in comparison with the DC group.
Conclusion: The results indicated that the combination of endurance swimming training and fenugreek seed extract can significantly reduce the plasma glucose levels and increase cardiac antioxidant enzymes activity in diabetic rats. Our findings suggest that this combination could be useful for the treatment of hyperglycemia and cardiac oxidative stress induced by diabetes mellitus.

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1. Introduction

Diabetes mellitus is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion or insulin action, or both [1]. Hyperglycemia may perturb cellular antioxidant defense systems and damage cells. Free radicals are formed disproportionately in diabetes by glucose oxidation, nonenzymatic glycation of proteins, and the subsequent oxidative degradation of glycated proteins. Oxidative stress plays an important role in the etiology of diabetes and diabetic complications [2]. Oxidative stress may constitute a focal point for multiple therapeutic interventions, and for therapeutic synergy. There is considerable evidence that oxidative stress from superoxide and other reactive oxygen species (ROS) contributes to the development of cardiovascular diseases, diabetes, and renal insufficiency [3]. Ihara et al [4] examined oxidative stress marked in diabetic rats and found increased ROS in pancreatic islets. 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 peroxidase [4].

Diabetes is associated with significant oxidative stress, and oxidative damage to tissues may be a contributory factor in several diabetic complications [5]. Diabetic patients have an increased incidence of vascular disease and it has been shown that oxidative stress elevated during diabetes [6]. Abnormally high levels of free radicals and the simultaneous decline of antioxidant defense mechanisms can lead to damage of cellular organelles and enzymes, increased lipid peroxidation, and development of insulin resistance [7].

Much controversy exists concerning the effects of endurance training on the oxidative status and antioxidant defense systems of the myocardium, which may decrease, increase, or even remain unchanged [8,9]. 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). Among various forms of treatments for diabetes mellitus, exercise and diet are of vital importance. The hypoglycemic effects of fenugreek seed have been studied in many animal model systems [10,11], as well as in humans in diabetic patients [12], but the results were controversial. In addition, fenugreek seeds have been shown to possess an encouraging antioxidant property [13] and can be a valuable candidate in the treatment and prevention of diabetes complications.

In several previous studies, the effects of training and fenugreek seed extract on the metabolic traits of diabetes were examined alone, and the results were controversial. To our knowledge, this is the first study to evaluate the effect of physical exercise and fenugreek seed extract in combination on the cardiac antioxidants in diabetic rats.

2. Materials and methods

2.1. Animals

Fifty male Wistar albino rats, weighing 200−250 g, and averaging 12 weeks old were used in this study. They were housed in metal cages under standard laboratory condition (12:12 hours 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 luminance of 150−200 lux in the daytime. The rats were randomly divided into five groups: (1) swimming training-fenugreek extract [1.74 g/kg body weight (BW); SF1, $n = 11$]; (2) swimming training (S, $n = 11$); (3) swimming training-fenugreek extract (0.87 g/kg BW; SF2, $n = 9$); (4) healthy control (HC, $n = 11$); and (5) diabetic control (DC, $n = 8$) this group received normal saline (5 mL/kg BW). Fenugreek and saline were treated orally by gastric gavage separately. The procedures used were in accordance with the guiding principle of the responsible committee for the care and use of animals.

2.2. Induction of diabetes

After fasting for 12 hours, the animals received an intraperitoneal injection (60 mg/kg BW) of streptozotocin (STZ; Sigma−Aldrich, St Louis, MO, USA), diluted in 1.0 mL of sodium citrate buffer (0.1M, 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 BW were monitored at the beginning and end of the experimental period.

2.3. Plant material

Fenugreek seeds were purchased from the local herbal market, cleaned, dried, and finely powdered in a grinding machine. The powdered fenugreek (1.5 kg) was boiled in 15 L distilled water for 30 minutes. The decoction was cooled for 30 minutes at room temperature, then filtered twice through a coarse sieve. Finally, the filtrate was concentrated by flash evaporation at 358°C to a thick paste (total 300 g).

2.4. Endurance training program

The swimming training protocol was conducted in two phases, adaptation and training. The adaptation phase consisted of the first 7 days of training. On the 1st day, the animals exercised in a round plastic tank (140 cm × 60 cm × 45 cm, water temperature 34−36°C) for 10 minutes. The exercise period was