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Short Report

Exercise Training Suppresses Expression of Tumor Necrosis Factor-α mRNA by Adipocytes in Noninsulin- dependent Diabetes Mellitus Mice

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Abstract

To investigate the effect of exercise training on glucose uptake and TNF-α mRNA expression by adipocytes in noninsulin-dependent diabetes mellitus (NIDDM), male C57BL/Ks-J-*db/db* mice were assigned to a sedentary group (SE) and a swimming-trained group (SW). Glucose uptake by adipose tissue was significantly higher in the SW mice than in the SE mice. In addition, TNF-α mRNA expression in adipose tissue was significantly lower in the SW mice than in the SE mice. These results suggest that exercise-induced glucose uptake in adipocytes might depend on suppression of TNF-α mRNA expression in NIDDM mice.

Introduction

Aerobic exercise training is generally considered to be beneficial for noninsulin-dependent diabetes mellitus (NIDDM) patients [1]. The effect of exercise training on blood glucose regulation has been mainly attributed to an increase in glucose uptake and metabolism in the skeletal muscles [2]. Some previous studies have shown that exercise training increases glucose uptake in the adipose tissue of non-diabetic rats [3], but its influence on glucose uptake in adipose tissue of NIDDM patients is not well understood. Recently, the development of obesity and NIDDM has been consistent with the involvement of as a central mediator [4]. TNF-α has been found to inhibit glucose uptake and play a role in hyperglycemia in obesity diabetes [5]. Previous investigators have speculated that overexpression of TNF-α in adipose tissue would inhibit the transport of glucose in an autocrine or paracrine fashion. In this study, the effect of exercise training on glucose uptake and TNF-α expression by adipocytes was investigated in NIDDM mice.

Materials and Methods

Male C57BL/Ks-J-*db/db* mice (4 wks) were assigned to a sedentary group (SE, n = 8) and a swimming-trained group (SW, n = 8). SW swam in a water tank at 37 ℃ for 10 wks (60 min $\cdot$ day^{-1}, 5 days $\cdot$ wk^{-1}). After 10 wks of swimming exercise, fasting plasma glucose and insulin concentrations were measured by the glucose oxidase method and EIA, respectively. Measurement of glucose uptake by adipose tissue was done using the method of Hotamisligil et al. [5] with some modifications. Under anaesthesia with

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an intraperitoneal injection of pentobarbital sodium, epididymal adipocytes were isolated from the mice by an aseptic technique. The tissue was stirred in incubation in RPM1640 with 10% fetal calf serum under 95% O_2 and 5% CO_2 at 37 ℃ with the addition of 2 g · L^{-1} glucose. TNF-α mRNA from the adipose tissue was treated by the RT-PCR method (GeneAmp, RNA PCR kit, Roche Molecular Systems Inc.). The first strand of cDNA was synthesized by reverse transcriptase, and then the cDNA samples were adjusted to PCR buffer conditions and were run for PCR simultaneously. The primers for TNF-α were 5'-GGCAGGTCTACTTTGGAGTCATTGC and 5'-ACATTCGAGGCTCCAGTGAATTCGG [6]. The PCR cycle program was carried out with 30 sec denaturation at 95 ℃, 30 sec annealing at 61 ℃, and 30 sec extension at 72 ℃. The PCR product was 307 base pairs. Ten to forty PCR cycles were run to determine the dependency of the number of PCR products on cycle numbers. Linearity between the amount of amplified cDNA and applied RNA was checked in the preplateau exponential phase. The values for specific mRNA were normalized on the basis of β-actin mRNA. The primers for β-actin were 5'-TGGAATCCTGTGGCATCCATGAAAC and 5'-TAAAACGCAGCTCAGTAACAGCCG [6].

Results and Discussion

The characteristics of the sedentary and swimming-trained NIDDM mice are shown in Table 1. Although plasma insulin concentrations in the two groups did not differ, plasma glucose concentrations were significantly lower in the SW mice than in the SE mice ($p < 0.05$, Table 1). Glucose uptake by adipose tissue was significantly higher in the SW mice than in the SE mice ($p < 0.05$, Fig. 1). In addition, TNF-α mRNA expression in adipose tissue after one hour incubation was significantly lower in the SW mice than in the SE mice ($p < 0.05$, Fig. 1).

Table 1 Effect of 10 wks of swim training on body and adipose tissue weights and plasma glucose and insulin levels.

	SE	SW
BW (g)	31 ± 2	33 ± 2
Epididymal AT/BW (%)	2.6 ± 0.3	2.2 ± 0.2*
Fasting plasma glucose (mg/dl)	563 ± 68	426 ± 98*
Fasting plasma insulin (ng/ml)	2.98 ± 0.76	2.74 ± 0.31

Values are the means±standard error. SE, sedentary; SW, swimming-trained; BW, body weight; AT, adipose tissue. *P < 0.05 vs. SE. The differences were analyzed by Mann Whitney U test.

The decrease in plasma glucose concentrations after an overnight fast in the swiming-trained mice as compared with the sedentary *db/db* mice, was in agreement with earlier reports [1]. To date, improvement in hyperglycemia by exercise training has been mainly attributed to insulin sensitivity and the glucose transport system in the skeletal muscles [2]. The present study showed an increase in glucose uptake by the adipose tissue in swimming-trained NIDDM mice. Previous studies demonstrated that exercise training increases glucose uptake in the adipose tissue of non-diabetic rats *in vitro* [7] and *in vivo* [3]. To increase glucose uptake, suppression of TNF-α mRNA expression in adipose tissue may be necessary, because an increase in the production of TNF-α in adipose tissue may play an important role in the insulin resistance mechanism associated with central obesity [8]. Exercise training induced suppression of TNF-α mRNA expression in the adipocytes of NIDDM mice when increased glucose uptake was observed. Decreases in fasting serum glucose and TNF-α levels have been reported in NIDDM patients after the treatment of

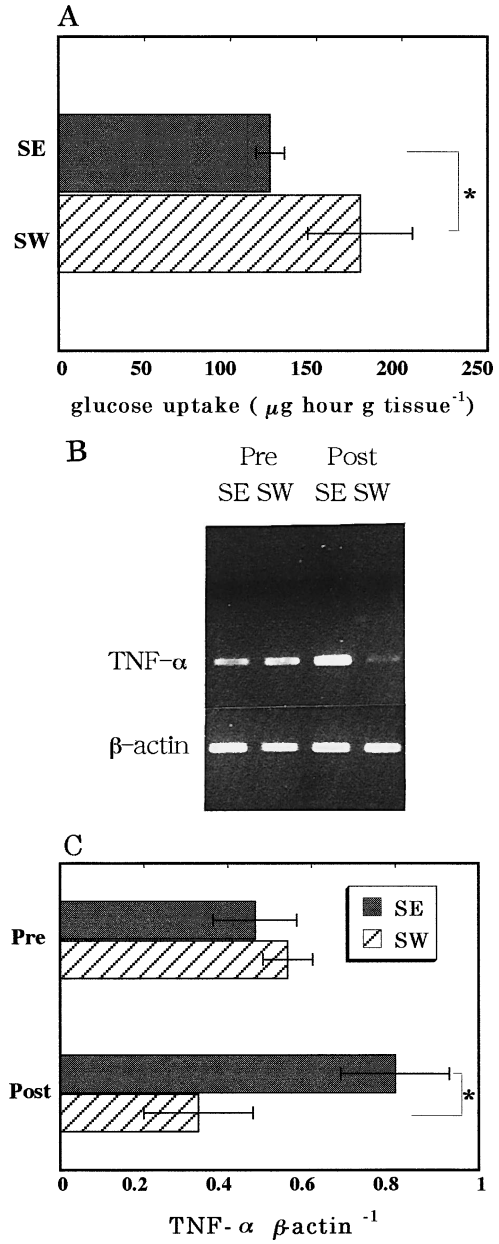

Fig. 1 Changes in glucose uptake and TNF-α mRNA expression in adipose tissue of NIDDM mice following exercise training. A ; glucose uptake was calculated by the following equation:

$$\text{Glucose uptake} = \frac{\frac{G0 - G60}{60} + \frac{G0 - G120}{120}}{2}$$

G0, G60, G120 indicate the glucose concentrations in medium collected at 0, 60 and 120 min during incubation. B ; the expression pattern of TNF-α mRNA was revealed by agarose gel electropboresis after performance of the RT-PCR. The adipose tissues were collected before (Pre) and after (Post) incubation in 10%FCS RPMI1640 with glucose. C ; Analysis of TNF-α mRNA expression. *p < 0.05.

diet and exercise [9]. However, the reason for the suppression of TNF-α mRNA expression in adipocytes following exercise training in NIDDM mice remains unclear. In conclusion, our results suggest that exercise training induces an increase in glucose uptake and suppresses TNF-α mRNA expression by the adipocytes in NIDDM mice.

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