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Translational

Regulation of glucose dynamics by noninvasive peripheral electrical stimulation in normal and insulin-resistant rats

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ABSTRACT

Background. The epidemic nature of type 2 diabetes mellitus (T2DM), along with the downsides of current treatments, has raised the need for therapeutic alternatives.

Methods. We studied normo-glycemic and high-fat diet (HFD), induced insulin-resistant Wistar Han rats for 2 to 3 weeks. Rats received peripheral electrical stimulation (PES) treatment (2 Hz/16 Hz bursts, 10 mA) in their hind limbs for 3 min, 3 times per week. Glucose tolerance was evaluated by using a glucose tolerance test at the beginning and again at the end of the study. The effect of an acute PES treatment on metabolic rates of glucose appearance and turnover was measured by using the hyperinsulinemic–euglycemic clamp (HEGC) test.

Results. Repeated PES treatment significantly inhibited the progression of glucose intolerance in normal and insulin-resistant rats and prevented HFD-induced gains in body weight and fat mass. Acute treatment induced a prolonged effect on glucose turnover, as evaluated by the HEGC test. Increased hepatic glucose output was observed during the basal state ($P < 0.005$). Under hyperinsulinemic conditions, PES improved tissue sensitivity to insulin (41.1%, $P < 0.01$), improved suppression of hepatic glucose production (58.9 ± 4.4% vs. 87.1 ± 4.4%, $P < 0.02$) and significantly elevated the rate of glycogenesis ($P < 0.01$), compared with controls.

Conclusions. The present study indicates that a noninvasive PES treatment of very short duration is sufficiently potent to stimulate glucose utilization and improve hepatic insulin

Abbreviations: T2DM, type 2 diabetes mellitus; PES, peripheral electrical stimulation; ipGTT, intraperitoneal glucose tolerance test; HEGC, hyperinsulinemic euglycemic clamp; EA, electro-acupuncture; FFA, free fatty acids; HFD, high fat diet; AUC, areas under the curve; HGP, hepatic glucose production; GIR, glucose infusion rate.

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sensitivity in rats. Repeated PES treatment may have a beneficial effect on HFD-induced adiposity and control of body weight.

1. Introduction

The global burden of type 2 diabetes mellitus (T2DM) has been a challenge for researchers for many years due to the growing need to manage and monitor solutions. Given the chronic nature of diabetes, treatments focus on controlling the symptoms through medications, regular physical activity, and dietary interventions [1].

It is well established that muscle contraction in response to voluntary (physical exercise) or nonvoluntary conditions induces skeletal muscle glucose uptake in normal individuals and T2DM patients through non-insulin dependent pathways [2]. Exercise may also improve insulin sensitivity and regulate glycogen synthesis in skeletal muscle after depletion of glycogen induced by contractile activity [3,4]. However, most patients show poor adherence to therapeutic exercise-training regimens because of a busy lifestyle or a medical disability [5]. Several groups of antidiabetic drugs, which have differing biological mechanisms of action for lowering blood glucose levels, are on the market [6]. In most cases, these medications tend to lose much of their efficacy after a few years of treatment even when used in combination with other medications. Furthermore, many of these medications have side effects such as weight gain and hypoglycemia, as well as an increased risk for congestive heart failure and gastrointestinal adverse events [7].

Electrotherapy, which is related to muscle contraction or nerve stimulation, is widely used in clinical practice for the treatment of a variety of medical disorders including cardiac diseases, spinal cord and peripheral nerve disorders, and pain [8–10]. Two main treatment approaches that use electrical stimulation have been investigated for improving glycemic control. One approach is the use of implanted stimulators to apply direct stimulation to pancreatic or gastrointestinal tract cells to regulate their neural electrical activity [11–13].

The other approach, which has been evaluated in several studies, is electro-acupuncture (EA). In this approach, low frequency (1–25 Hz) stimulation is applied through needle electrodes inserted in traditional skin acupuncture points. Several studies demonstrated changes in glucose dynamics after 30–90 min of EA treatment in anesthetized normal rodents and rodent models of type 1 and type 2 diabetes. In fasting conditions, an insulin-induced hypoglycemic effect was observed after EA treatment in the abdomen [14–16] and points in the hind limbs [17]. Furthermore, in response to glucose intake, improvement in glucose tolerance and glucose uptake was demonstrated with a glucose tolerance test (GTT) and a glucose clamp test [16,18,19]. Due to the relatively long duration of the treatment, these observations were, at least in part, related to muscle contraction, which directly influences skeletal muscle and adipose tissue signaling pathways. The effect of EA on insulin levels [16,20], expression of the glucose transporter GLUT4 [21,22], and serum free fatty acids (FFA) [19] is similar to the effect achieved after voluntary exercise as expressed by an increased glucose disposal rate [23–25]. However, despite its beneficial effects, this type of treatment is invasive and uncomfortable and requires professional assistance for administration.

Interestingly, only a few studies have investigated the involvement of neural pathways in regulation of glucose with EA stimulation [19,26]. It has been shown that EA applied for a relatively short duration (10 min) in the hind limbs triggers a neural reflex response that increases hepatic glucose output in anesthetized rats [27]. In addition, EA treatment for 10 min improved responses of plasma glucose to an intravenous administration of insulin in diabetic rats [28]. These effects were abolished after alpha and beta blockers were administered and after the sciatic and femoral nerves were severed. Although these studies provide strong evidence of the effect of EA on glucose regulation through neural pathways, quantitative data regarding the changes in peripheral and hepatic insulin sensitivity, as well as the glycogen synthesis rate, remain to be determined.

The aims of this study were twofold: (a) to evaluate the effect of noninvasive peripheral electrical stimulation (PES) in the hind limbs on glucose regulation in conscious and unrestrained rats, and (b) to investigate quantitatively acute and long-term effects of a very short duration of PES treatment (3 min) on blood glucose uptake and production evaluated with a GTT and an accurate calculation model of a hyperinsulinemic euglycemic clamp (HEGC) test.

2. Materials and Methods

The experiments were performed in three phases. First, normal rats were studied so that a possible acute and prolonged effect of noninvasive stimulation could be evaluated. Next, rats in which insulin resistance had been induced by diet were studied. In the third stage, an acute effect of PES treatment on blood glucose uptake and production was explored quantitatively in normal rats.

2.1. *Animals*

Three-month-old male Wistar Han rats (Harlan, Jerusalem) were housed in individual cages and subjected to a standard light (6:00 A.M. to 6:00 P.M.)/dark (6:00 P.M. to 6:00 A.M.) cycle. Normal rats were fed ad libitum with regular rat chow, which consisted of 50% carbohydrate, 18.8% protein, and 6% fat with a physiological fuel value of 3.3 kcal/g (2018SC Teklad Global Harlan, Jerusalem). Whole body insulin resistance was induced by feeding the rats a high-fat diet (HFD), which resembles a typical Western diet, for 3 weeks, as previously reported [29,30]. The diet consisted of 49.9% carbohydrate (34% sucrose), 17.8% protein, and 20% fat (0.15% cholesterol and 19% milk fat), with a physiological fuel value of 4.55 kcal/g, with 40% of the energy coming from fat (AIN-76 A Western diet, Harlan, Jerusalem). HFD-fed animals displayed a moderate increase in fasting glycemia and impaired glucose tolerance