Original Paper

Biology of the Neonate

Biol Neonate 1999;75:337–342

Treadmill Training Enhances Glucose Tolerance More in Pregnant than in Virgin Rats

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Key Words

Rat · Pregnancy · Exercise · Lactate · Glucose tolerance

Abstract

To determine whether aerobic training throughout gestation modifies glucose tolerance, female Wistar rats were mated or kept nonpregnant and run or not on a 10° slope treadmill for 5 days/week at 20 m/min, starting with a 20-min run, and with a progressive daily increase of 5 min, reaching a 75-min run on the 20th day of protocol or gestation. The exercise protocol did not modify food intake, maternal and fetal weights, litter size or blood lactic acid levels. The rise in blood glucose after an oral glucose load (2 g/kg body weight) did not differ between trained and untrained nonpregnant rats but was lower in trained than in untrained pregnant rats. In the untrained rats the rise in plasma insulin levels after the glucose load was much greater in pregnant than in nonpregnant rats; in trained rats this difference between groups was attenuated by the greater effect of exercise decreasing the plasma insulin response to the glucose load in pregnant than in nonpregnant rats. Thus, an aerobic exercise protocol that does not modify the outcome of pregnancy does significantly reduce the altered oral glucose tolerance in pregnant rats and only has a minor effect in nonpregnant rats.

Introduction

Late pregnancy has been characterized by an insulin resistance both in women [2, 11, 26] and experimental animals [9, 17, 18, 22] that is due to the decreased insulin sensitivity of peripheral tissues, principally skeletal muscle [17, 25]. This insulin resistance is normally manifested by elevated plasma insulin concentrations and normoglycemia in response to meals or a glucose challenge [18, 22, 28], and reduced sensitivity to the hypoglycemic ef-

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0006–3126/99/0755–0337$17.50/0

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fects of exogenous insulin [18, 22, 24, 26]. Physical training in the nonpregnant state has been associated with an increase in insulin sensitivity [13, 15, 23, 30], an increase in insulin-induced glucose uptake by skeletal muscle [20] and enhanced glucose tolerance [1, 8, 27]. Acute exercise in trained pregnant animals also increased glucose uptake in maternal skeletal muscle without compromising glucose uptake by fetus or placenta [21]. The objectives of the present study are both to ascertain whether a regular aerobic training affects fetal outcome in pregnant rats and to determine the effects of training on maternal glucose tolerance.

Materials and Methods

Animals

Female Wistar rats from our own colony (Center for Animal Experimentation 86/609/UE; ref. No. 28005-22A) were used throughout. They were housed in individual cages at 22 ± 2 °C, relative humidity 55 ± 10% and lights on from 12:00 to 24:00 h. Rats had free access to tap water and chow pellets (Panlab SL, Barcelona, Spain) with a composition of 17% protein, 3% fat, 4.3% fiber, 5% minerals, 58.7% carbohydrates, and 12% humidity (2,900 kcal/kg diet). Rats were mated when weighing 150–160 g, and the day spermatozoa appeared in vaginal smears was considered day 0 of gestation. Age-matched virgin rats were used as controls. Parallel studies were performed on trained and untrained animals on day 20 of pregnancy or protocol.

Experimental Design

Training was performed by running on a 10° sloped rodent tape (Treadmill LI 8706; Letica, Spain). Rats were pretrained by running for 5–7 min/day on the treadmill, for 3 days before beginning the protocol. In pregnant rats, the exercise protocol was initiated on day 0 of gestation and virgin rats were studied in parallel. Both virgin and pregnant rats were exercised during their dark cycle 5 days/week starting at the speed of 20 m/min for 20 min, with a progressive daily increase of 5 min until a total of 75 min on day 20 of protocol or gestation.

To evaluate lactate concentration, blood samples were collected from the tip of the tail on day 20 of pregnancy or the exercise protocol just before and at different times during the run. The samples were immediately deproteinized with ice-cold perchloric acid (0.165 mol/l) and after centrifugation, supernatants were analyzed by a lactate dehydrogenase method using a commercial kit (Boehringer Mannheim, Germany).

A glucose tolerance test [18, 22] was performed on rats that were not subjected to the lactate analysis. On day 20 of the experiment, 3 h after the last run in case of the exercised rats, each rat received, by stomach tube, a glucose load of 2 g glucose/kg of body weight dissolved in distilled water. Blood samples were collected from the tip of the tail just before glucose load and at 7.5, 15, 22.5, 30, 60 and 90 min after the glucose load. Blood glucose was measured with a Reflolux II analyzer (BM-Test-Glycemie 20-800R; Boehringer Mannhiem) and plasma aliquots were kept at −80 °C until insulin determination [10] using a rat-specific radioimmunoassay kit from Incstar Corp. USA.

Expression of the Results

Results are expressed as means ± SE. Statistical comparisons were carried out using the Student's t-test for two groups and by the Student-Newman-Keuls test for more than two groups with the IBM 'Graph Pad Instat'. A significant difference between the groups was taken as $p < 0.05$.

Results

As shown in table 1, body weight and average daily food intake in both untrained and trained pregnant animals were significantly higher on days 12 and 20 than in both virgin groups. The exercise protocol did not modify either conceptus-free body weight (net weight), total conceptus weight or mean fetal and placental weights or the number of fetuses per litter (table 1).

Blood lactic acid levels were measured on day 20 of the experiment just before and at different times during the run. As shown in table 2, there was no difference between pregnant and virgin rats in blood lactate levels at any of the running times versus time 0.

Table 1. Effect of aerobic exercise on body weight, food intake and pregnancy outcome in 20-day pregnant and virgin rats

	Group			
	virgin		pregnant	
	untrained	trained	untrained	trained
Increment body weight, percentage versus day 0				
Day 12	110.7±1.8	110.9±2.0	130.4±6.4[a]	126.6±2.5[d]
Day 20	117.7±2.2	117.5±3.2	166.4±10.4[b]	164.3±3.9[d]
Average daily food intake, g				
Day 0	17.0±1.4	16.0±0.8	17.0±1.4	16.4±0.8
Day 12	16.4±0.9	16.7±1.0	21.6±0.6[b]	21.3±0.9[c]
Day 20	15.8±0.8	15.4±1.0	21.0±0.7[b]	21.0±0.9[c]
Conceptus weight, g	–	–	50.3±3.0	56.0±2.9
Net maternal body weight, g, as % of day 0	–	–	137.8±8.1	132.2±3.9
Litter size	–	–	11.00±1.0	12.00±1.0
Fetus weight, g	–	–	3.03±0.1	3.25±0.1
Placenta weight, g	–	–	0.59±0.02	0.59±0.01

Statistical comparisons between pregnant and virgin rats are shown by superscript letters: [a] $p < 0.01$ and [b] $p < 0.001$ for control rats, and [c] $p < 0.01$ and [d] $p < 0.001$ for exercised rats. n = 8–9 rats/group.

Changes in blood glucose and plasma insulin after the oral glucose load are shown in figures 1 and 2. Blood glucose levels before the glucose load in late pregnant rats were similar in trained and untrained rats and significantly lower than in virgin animals. Although the oral glucose load produced a rise and a subsequent decrease in blood glucose values in the four groups, the response to exercise was clearly different between virgin and pregnant rats (fig. 1). Whereas the exercise protocol did not modify glucose tolerance in virgin rats, in pregnant rats the rise in blood glucose was lower and later in the trained than in the untrained animals (fig. 1). As shown in figure 2, basal plasma insulin values in the pregnant rats were the same in trained and untrained rats and were significantly higher than in the respective virgin groups. After the oral glucose load there was a higher and faster rise

Table 2. Effect of treadmill exercise on blood lactate levels (mmol/l) in 20-day pregnant and virgin rats

Exercised virgin (t: 0)	1.23±0.38
Time of training, min	
t: 15	1.61±0.30
t: 35	1.54±0.23
t: 55	1.11±0.19
t: 65	1.12±0.08
t: 75	1.21±0.24
Exercised pregnant (t: 0)	0.90±0.19
Time of training, min	
t: 15	1.69±0.31
t: 35	1.54±0.16
t: 55	1.30±0.09
t: 65	1.64±0.34
t: 75	1.41±0.17

Statistical comparisons between the groups were not significant ($p > 0.05$). n = 8 rats/group.

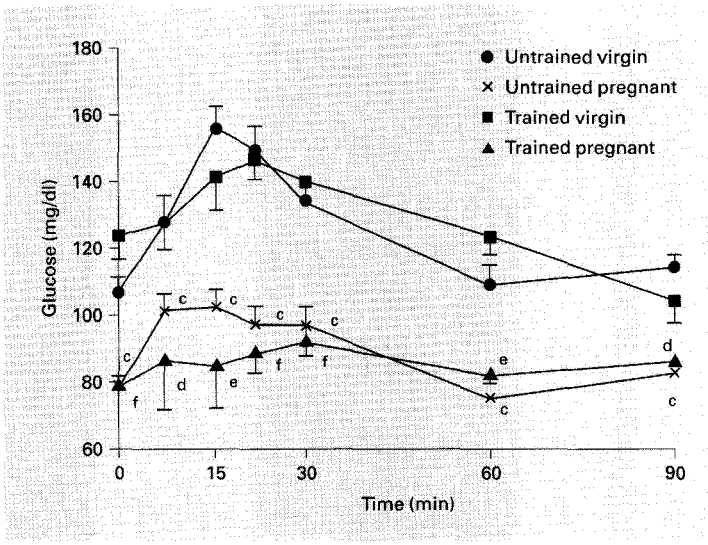

Fig. 1. Effect of exercise in trained 20-day pregnant and virgin rats on blood glucose levels after an oral glucose load (2 g/kg). Statistical comparison between the groups is shown by letters: untrained pregnant versus untrained virgin rats, c ($p < 0.001$); trained pregnant versus trained virgin rats, d ($p < 0.05$), e ($p < 0.01$), f ($p < 0.001$). n = 5–29 rats/group.

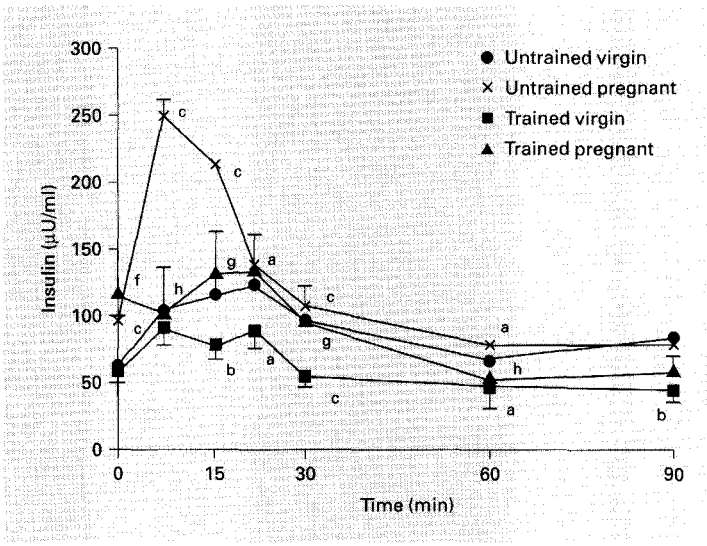

Fig. 2. Effect of exercise in trained 20-day pregnant and virgin rats on plasma insulin levels after an oral glucose load (2 g/kg). Statistical comparison between the groups is shown by letters: untrained pregnant and trained virgin versus untrained virgin rats, a ($p < 0.05$), b ($p < 0.01$), c ($p < 0.001$); trained pregnant versus trained virgin rats, f ($p < 0.001$); trained pregnant versus untrained pregnant rats, g ($p < 0.05$), h ($p < 0.01$). n = 5–29 rats/group.

in plasma insulin levels in untrained pregnant than in untrained virgins, and this difference was significant at 7.5, 15, 22.5, 30 and 60 min (fig. 2). The exercise protocol decreased the rise in plasma insulin levels after the glucose load in both virgin and pregnant rats, although the effect in the latter was greater. Thus, in virgin rats plasma insulin levels at 15, 22.5, 30, 60 and 90 min after the glucose load were significantly lower in trained than in the untrained rats. In trained pregnant rats, plasma insulin was significantly lower than in the untrained pregnant rats at 7.5 and 15 min, but not at 30, 60 and 90 min, reflecting the rapid decline in plasma insulin levels found in the control pregnant rats (fig. 2).

Discussion

The present results show that moderate aerobic exercise protocol in pregnant rats does not modify body weight, food intake or pregnancy outcome, but enhances oral glucose tolerance, even though it decreases the rise in plasma insulin after an oral glucose load, suggesting enhanced insulin sensitivity. In agreement with previous findings [18], nontrained pregnant rats showed a normal proportional increase in blood glucose levels and a higher increase in plasma insulin after an oral glucose load than did virgin rats, confirming the insulin-resistant condition of the late pregnant rat. The present results therefore suggest that an aerobic exercise protocol corrects the insulin resistance in the late pregnant rat. This effect differs from what is found in virgin rats subjected to the same exercise protocol, since their oral glucose tolerance is not modified by exercise and the rise in plasma insulin after the glucose load is only slightly smaller in the trained than in the nontrained virgin rats.

Using different protocols, previous studies have also shown greater responsiveness to exercise in pregnant than in nonpregnant rats [21, 29], although the present study is the first one to show that regular exercise can normalize the altered glucose tolerance normally present during late pregnancy. An important aspect of the protocol used here is that the lack of change in the blood lactic acid levels during the runs shows that the moderate, regular and progressive exercise is aerobic.

Although maternal exercise has been questioned during pregnancy because it may negatively effect fetal growth, metabolism and even viability [7, 29], the present study shows that moderate aerobic exercise in the rat does not affect pregnancy outcome whereas it greatly improves maternal oral glucose tolerance. It is well known that maternal insulin resistance and the needs for an enhanced pancreatic β-cell function normally present during late pregnancy [3, 6, 16] predispose the mother toward developing diabetes; thus strategies that decreased this risk without affecting fetal growth or metabolism, as is the case with moderate exercise protocols, would be highly desirable. This strategy is already being successfully applied in diabetic pregnant women [14], although it is still a controversial matter [4, 5, 12, 19], since the possible effects on the fetus depend on the type and intensity of the exercise protocol followed. Studies in the rat have shown that maternal exercise may affect fetal metabolism, growth and even viability, depending on the type of exercise protocol followed. Acute exercise has been shown to compromise fetal glucose uptake because it increases the glucose uptake by maternal skeletal muscles [21, 29], and high intensity regular exercise reduces the weight and the number of offspring [7], reflecting some deleterious effect on the fetus. Nevertheless, our present results show that moderate regular aerobic exercise has a positive effect in correcting insulin resistance in the trained late pregnant rat without affecting the outcome of pregnancy.

Acknowledgment

This investigation was supported by a grant from the Dirección General de Investigación Científica y Técnica (PB92/0175), Ministerio de Educación y Cultura of Spain.

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