Attenuation of Fructose-Induced Hypertension in Rats by Exercise Training

GERALD M. REAVEN, HELEN HO, AND BRIAN B. HOFFMAN

SUMMARY This study was initiated to see if the insulin resistance, hyperinsulinemia, and hypertension that follow feeding normotensive Sprague-Dawley rats a fructose-rich diet could be prevented by letting rats run spontaneously in exercise wheel cages. Blood pressure in sedentary rats increased from (mean ± SEM) 125 ± 2 to 148 ± 3 mm Hg in response to 2 weeks of a high fructose diet, and this increment was significantly ($p < 0.001$) attenuated in exercising rats (from 121 ± 1 to 131 ± 2 mm Hg). In addition, mean (±SEM) plasma insulin concentration was lower in fructose-fed rats allowed to run spontaneously (44 ± 2 vs 62 ± 5 μU/ml; $p < 0.01$). Finally, resistance to insulin-stimulated glucose uptake was assessed by determining the steady state plasma glucose response to a continuous glucose and exogenous insulin infusion during a period in which endogenous insulin secretion was suppressed. The results of these studies indicated that the mean (±SEM) steady state plasma glucose concentration was significantly lower in the exercise-trained rats (127 ± 5 vs 168 ± 6 mg/dl; $p < 0.001$), despite the fact that the steady state plasma insulin levels were also lower in rats allowed to run spontaneously (75 ± 4 vs 90 ± 5 μU/ml; $p < 0.05$). Thus, the ability of exercise-trained rats to stimulate glucose disposal was enhanced as compared with that of sedentary rats fed the same fructose-rich diet. These data demonstrate that the insulin resistance, hyperinsulinemia, and hypertension produced in normotensive rats by feeding them a high fructose diet can be attenuated if rats are allowed to run spontaneously. These results provide further support for the hypothesis that insulin resistance and hyperinsulinemia play a role in the pathogenesis of fructose-induced hypertension. (Hypertension 12: 129–132, 1988)

KEY WORDS • fructose-induced hypertension • insulin resistance • hyperinsulinemia • exercise training

IN a previous report we found that hypertension can be produced by feeding a high fructose diet to normal rats.[1] Fructose-fed rats also develop resistance to insulin-stimulated glucose uptake and hyperinsulinemia,[1-3] phenomena that have recently been identified as occurring in patients with hypertension.[4-7] Based on these findings, we raised the possibility that the changes in insulin action and concentration noted in fructose-fed rats with hypertension may play a role in the pathogenesis of the elevated blood pressure seen in this situation. In this context, we have previously demonstrated that insulin-stimulated glucose uptake is enhanced in exercise-trained rats[8] and that the hyperinsulinemia associated with feeding rats a high fructose diet is attenuated if rats are allowed to run spontaneously.[9] Thus, in the present report, we tested the hypothesis that insulin resistance and hyperinsulinemia are involved in the development of hypertension in fructose-fed rats by comparing the effects of fructose-rich diets on both insulin metabolism and blood pressure in sedentary rats with those seen in rats allowed to exercise spontaneously in running wheels.

Materials and Methods

General Protocol

Male Sprague-Dawley rats (Harlan, Indianapolis, IN, USA) were used for all experiments. The rats were approximately 6 weeks old and weighed approximately 150 g when the study was started. Prior to any manipulation of diet or level of physical activity, rats were fed standard rat chow (Wayne Lab Blox, Allied Mills, Chicago, IL, USA) containing 60% vegetable starch, 11% fat, and 29% protein, and maintained on a 12-hour light/dark (0600/1800) cycle.

Rats were divided into two groups at the start of the study. One group of 18 rats was housed three to a standard cage, while the 18 rats in the other group

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were placed individually in exercise wheel cages (Wahman, Timonium, MD, USA) and allowed to run at their own pace. The cages, as supplied by the manufacturer, consist of a rotating wheel cage (with the number of revolutions/day recorded by a cyclometer attached to the wheel axis) plus an adjoining feed cage. To maximize exposure to the wheel, we replaced the feed cage attachment with a small feeding trough large enough to hold a 4- to 5-day food supply. Thus, the exercising animals remained entirely in the wheel portion of the cage to run at will and feed on laboratory chow ad libitum from the attached feeding trough. Rats of this age consume approximately 32 g of food per day when housed in running wheels, as compared with 25 g/day for sedentary rats housed three to a cage.[8]

At the start of the second week rats were acclimated to the procedure of blood pressure measurement at 1300 daily for 1 week. All rats were then given a diet containing 66% fructose, 12% fat, and 22% protein (Teklad Labs, Madison, WI, USA), and they continued to eat this diet until the end of the study. The mineral content of the two diets used was reasonably comparable: The chow diet contained sodium, 2.6 g/kg, and potassium, 10.8 g/kg, as compared with sodium, 4.9 g/kg, and potassium, 4.9 g/kg, for the fructose diet. The fat and protein were essentially identical in the two diets. During this period blood pressure was measured 5 times/week. Food was removed at 0800 on the 14th day, rats were weighed, and blood was taken by tail-bleeding at 1300. The diets were continued for 2 more days, and insulin suppression tests were performed as will be described.

Blood Pressure Measurement

Rats were removed from the animal room and taken to the laboratory at 0900, allowed free access to diet and water, and kept in a quiet area before the blood pressure was measured at 1300. The tail-cuff method, without external preheating, was used to measure the systolic blood pressure.[10] Ambient temperature was kept at 30 °C. The equipment used included magnetic animal holders, connected with manual scanner (Model 65-12, IITC, Woodland Hills, CA, USA), pulse amplifier (Model 59, IITC), and dual-channel recorder (Model 1202, Linear Instruments, Reno, NV, USA). The systolic blood pressure was measured in the conscious state and has been shown using this technique to be similar to that obtained by direct arterial cannulation.[11] The mean of five consecutive readings was used as the measurement of the systolic blood pressure of each rat for that day, and the average blood pressure on the last 2 days of each study period was calculated and used for statistical comparisons.

Measurement of In Vivo Insulin Action

In vivo insulin action was quantified by a modification of a method previously used in humans[12] and rats.[13] Food was withdrawn at 0800 the morning of the study, and the procedure was started at 1200. Rats were anesthetized by an intraperitoneal injection of sodium thiamylal (7.5 mg/100 g body weight), and the right internal jugular was exposed and cannulated for administration of the infusate. Rats received a continuous infusion of glucose (8 mg/kg/min) and insulin (2.5 mU/kg/min) for 180 minutes. With this technique, comparable steady state plasma insulin levels are reached in all animals during the last hour of the study. By measuring the steady state plasma glucose concentration during the third hour, it is possible to get a direct assessment of the ability of a fixed concentration of insulin to stimulate glucose uptake in the various groups. Steady state plasma glucose and insulin values were calculated from the mean of tail blood samples taken at 10-minute intervals during the last 60 minutes of the infusion.

Biochemical Measurements

Tail blood samples were taken at the beginning of each experiment and 14 days later. They were centrifuged, aliquoted, frozen, and later assayed for glucose[14] and insulin.[15]

Results

Rats housed in the exercise wheel cages ran a mean (±SEM) of 3.4 ± 0.5 miles/day. They weighed 177 ± 2 g before the fructose diet was started, which was slightly less than the weight of the sedentary controls (183 ± 2 g; $p < 0.06$). Weight gain over the 2-week fructose diet period was greater in the sedentary group (100 ± 3 vs 71 ± 5 g; $p < 0.001$), with a final weight of 283 ± 4 g in the control as compared with 248 ± 6 g in the rats allowed to run spontaneously.

The effect of exercise training on fructose-induced hypertension is illustrated in Figure 1. Blood pressure rose from 125 ± 2 to 148 ± 3 mm Hg in sedentary rats in response to the high fructose diets, as compared with a smaller increase from 121 ± 1 to 131 ± 2 mm Hg in exercising rats. Both the

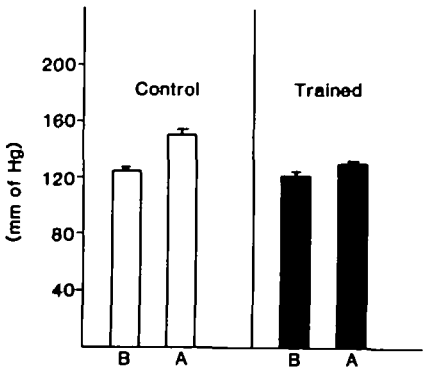

FIGURE 1. *Mean (±SEM) blood pressure before (B) and after (A) 2 weeks of a fructose-enriched diet in sedentary control and exercise-trained Sprague-Dawley rats. There were 18 rats in each group.*

incremental change (10 ± 2 vs 23 ± 3 mm Hg) and the final blood pressure were lower ($p < 0.001$) in the exercise-trained rats. Blood pressure was also somewhat lower before the fructose diet was started in the rats housed in the exercise wheel cages.

Plasma insulin concentrations in the two groups are illustrated in Figure 2. Plasma insulin concentrations were lower in the exercising rats both before (20 ± 1 vs 26 ± 1 μU/ml; $p < 0.01$) and after (44 ± 2 vs 62 ± 5 μU/ml; $p < 0.01$) 2 weeks of the fructose-enriched diet. In addition, the incremental rise in plasma insulin in response to the high fructose diet was lower (23 ± 2 vs 36 ± 5 μU/ml; $p < 0.05$) in the spontaneously running rats.

Steady state plasma glucose and insulin concentrations of the two groups are shown in Figure 3. Mean (±SEM) steady state plasma glucose concentrations were lower (127 ± 5 vs 168 ± 6 mg/dl; $p < 0.001$) in the exercise-trained rats. This difference was found even though steady state plasma insulin levels turned out to be lower (75 ± 4 vs 90 ± 5 μU/ml; $p < 0.05$) in the rats allowed to run spontaneously. Thus, resistance to insulin-stimulated glucose uptake was greater in fructose-fed rats that were kept sedentary.

Discussion

The results of the current experiments confirm the previous report from our group that hyperinsulinemia and hypertension can be induced in normal rats by feeding a fructose-enriched diet. Fructose is not unique in this regard, and increases in intake of either sucrose or glucose have also been shown to raise blood pressure in experimental animals.[16–20] However, the hypertensive effect in the latter two sugars has been shown to occur when the carbohydrate was given to rats with spontaneous hypertension or to normal rats in combination with a high salt diet. On the other hand, insulin resistance and hyperinsulinemia have been shown to develop when normal rats eat a high sucrose diet,[21] and it is certainly possible that hypertension also develops in this situation.

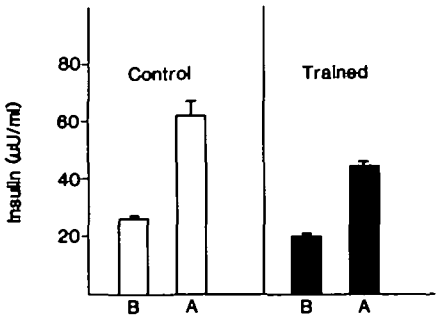

FIGURE 2. *Mean (±SEM) plasma insulin concentration before (B) and after (A) 2 weeks of a fructose-enriched diet in sedentary control and exercise-trained Sprague-Dawley rats. There were 18 rats in each group.*

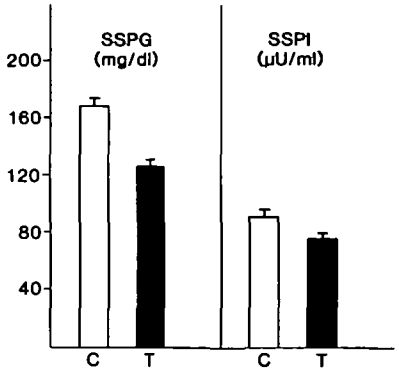

FIGURE 3. *Mean (±SEM) steady state plasma glucose (SSPG) and insulin (SSPI) concentrations in control sedentary (C) and exercise-trained (T) rats after 2 weeks of a fructose-enriched diet. There were 18 rats in each group.*

The rationale for the present study was based on our previous finding[8] that insulin-stimulated glucose uptake is enhanced when rats are permitted to run spontaneously and that this effect is associated with a reduction in circulating insulin concentration. This phenomenon has been demonstrated in normal rats[8] as well as in rats fed diets enriched with glucose, fructose, or sucrose.[9] The current study was initiated to take advantage of this effect of exercise on insulin metabolism and was designed to test the hypothesis that the development of hypertension in fructose-fed rats may be secondary to the insulin resistance and hyperinsulinemia associated with the high carbohydrate diet. We reasoned that if the magnitude of insulin resistance and hyperinsulinemia usually associated with feeding rats a fructose-enriched diet[1–3] were attenuated in spontaneously running rats, then this effect should inhibit the increase in blood pressure previously shown to occur in fructose-fed rats.[1] It is clear from the data presented that the prediction was borne out—insulin-stimulated glucose uptake was significantly greater and plasma insulin levels were lower in the exercise-trained rats fed a high fructose diet than in sedentary rats consuming a similar diet. However, we cannot tell if the enhanced insulin sensitivity and lower insulin levels noted in the running rats were due to the effect of exercise training or simply the reflection of the metabolic impact of spontaneous running. In either event, the results indicate that insulin action was better and insulin levels were lower in fructose-fed rats allowed to run spontaneously. However, the finding that these changes in insulin action and concentration in exercise-trained rats were associated with an attenuation of the fructose-induced increase in blood pressure does not prove that these events are causally related. On the other hand, these results provide support for the hypothesis that insulin resistance and hyperinsulinemia are involved in the patho-

genesis of the hypertension resulting from feeding rats a high fructose diet.

The observation that fructose-induced hypertension was greatly inhibited in spontaneously running rats does not prove that this was because insulin-stimulated glucose uptake was greater and plasma insulin levels were lower in the exercise-trained rats. For example, exercise training has been shown to lower blood pressure in patients with hypertension,[22] and there is no a priori reason to believe that this effect is mediated by a change in insulin action and circulating insulin level. However, in support of a role for plasma insulin level is the observation that the fall in blood pressure that occurred as the result of an exercise training program in patients with hypertension was highly correlated with the degree of hyperinsulinemia present before training was initiated.[22]

Another possible explanation for the ability of exercise training to modulate the effects of a high fructose diet on blood pressure in normal rats is the fact that the spontaneously running rats weigh less than the sedentary controls, despite the fact that they consume approximately 30% more calories per day.[8] However, the difference in weight between the two groups was only 12%, and the blood pressure did not change over the 2-week experimental period in the control rats, even though the weights differed by 36%. Furthermore, previous studies in humans have shown that the blood pressure–lowering effects of exercise training are independent of changes in weight.[22] Indeed, it has been suggested on the basis of these studies in humans that the hypertension associated with obesity may be related to the presence of obesity-related variables such as hyperinsulinemia.[22] Given these considerations, the hypothesis that fructose-induced hypertension is attenuated in rats allowed to run spontaneously because exercise training reduces the insulin resistance and hyperinsulinemia usually associated with a high fructose diet appears to be a tenable one. As such, the results of the current experiments provide further evidence that insulin resistance and hyperinsulinemia may play an important role in the pathogenesis of hypertension.

Additional evidence that insulin may play a role in blood pressure regulation is the observation that the induction of insulin-deficient diabetes with streptozotocin in spontaneously hypertensive rats has been associated with a dose-dependent reduction in blood pressure,[23] whereas this was not the case in Wistar-Kyoto rats made equally insulin-deficient. Whether or not hyperinsulinemia ultimately turns out to play a causal role in the development of hypertension will depend on the results of future studies, but the results of experiments to date support the view that this hypothesis is worthy of continued consideration.

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