Leptin responses to glucose infusions in obesity-prone rats

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Levy, James R., John Lesko, Richard J. Krieg, Jr., Robert A. Adler, and Wayne Stevens. Leptin responses to glucose infusions in obesity-prone rats. Am J Physiol Endocrinol Metab 279: E1088–E1096, 2000.—The secretion of leptin is dually regulated. In fasting animals, plasma leptin concentrations reflect body fat stores, whereas the incremental leptin response to fasting or refeeding most likely reflects insulin-mediated energy flux and metabolism within adipocytes. Impaired secretion of leptin in either pathway could result in obesity. We therefore measured plasma leptin concentrations in fasted animals and plasma leptin concentrations after an intravenous glucose infusion in a rat model of obesity. Young Sprague-Dawley (S-D) and Fischer 344 (F344) rats had similar percent body fat and fasting glucose and fasting leptin concentrations. However, F344 animals had higher insulin concentrations and leptin responses to intravenous glucose than did the S-D animals. The animals were then fed a control or high-fat diet for 6 wk. High-fat fed animals gained more weight and body fat than the control fed animals. Control and high-fat fed F344 animals gained ~40% (P < 0.0001) more weight and >100% (P < 0.01) more body fat than the S-D animals. Fasting leptin concentrations and leptin concentrations after intravenous glucose infusions and feeding were more than double (P < 0.05) in F344 animals compared with S-D animals. Whether an animal is fed a control or high-fat diet had little effect on the leptin response to intravenous glucose. In conclusion, young, lean F344 animals, before the onset of obesity, demonstrated a greater acute leptin response to intravenous glucose than similarly lean S-D animals. After a 6-wk diet, F344 animals had a greater percent increase in body weight and insulin resistance and exhibited higher fasting leptin concentrations and a greater absolute leptin response to intravenous glucose compared with the S-D animals. The chronic diet (control or high fat) had little impact on the acute leptin response to intravenous glucose. F344 animals exhibit leptin resistance in young, lean animals and after aging and fat accumulation.

secretion; Fischer 344; body fat

The chronic and acute regulation of leptin secretion may modulate food-seeking behavior and eventual caloric intake. Therefore, dysregulation of either pathway may alter body weight homeostasis and cause thinness or obesity. Both increased or decreased fasting serum leptin concentrations have predicted future weight gain. In an animal model of leptin resistance induced by a leptin receptor mutation, fasting leptin concentrations are increased at birth, and they precede the onset of increased fat storage (25). In animals or humans with no known leptin receptor mutations,
lower baseline serum leptin concentrations have predicted a propensity for weight gains that exceed those in weight-matched control animals or humans with elevated serum leptin concentrations (34, 44, 56, 60). To our knowledge, the correlation between the acute leptin response and the propensity for future weight gain has not yet been studied in animals. Hypothetically, either blunted or exaggerated acute leptin secretory responses may predict relative weight and body fat gain. A blunted acute leptin secretory response may result in inadequate satiety signals, overeating, and obesity. An exaggerated leptin secretory response may signal ineffective delivery of leptin to the hypothalamus or relative leptin resistance. Therefore, we chose to study the acute leptin secretory pathway in two strains of rat with different propensities for weight and body fat gain. Sprague-Dawley (S-D) rats are known to become obese when fed a high-fat diet (4, 8, 41, 51). Compared with the obesity-prone S-D rats, we studied Fischer 344 (F344) rats as a strain purported to be relatively resistant to obesity (4, 13). Fischer rats have been studied in the past as a model of aging-induced insulin resistance precisely because it was thought that Fischer rats appeared to age without appreciable amounts of weight gain or of body fat accumulation (14, 35). We compared the fasting leptin concentrations and the leptin response to intravenous glucose in S-D and F344 rats before and after 6 wk of a control or high-fat diet.

METHODS

Animals and diets. All animals were humanely treated, and the experimental protocols were reviewed and accepted by the Institutional Animal Care and Use Committee at Virginia Commonwealth University. Two-month-old male S-D and F344 rats were purchased from Harlan Sprague Dawley (Indianapolis, IN) and housed in individual cages at 22°C. Lighting was controlled on a natural-dark-light cycle (lights out at 1800-lights on at 0600). S-D rats are outbred, and it has been reported that there is considerable variation in the strain in susceptibility to obesity (6, 29). In this study, we did not attempt to identify obesity-prone and obesity-resistant S-D animals. F344 rats are highly inbred. The percent body fat was measured (see Body fat), and the acute leptin response to intravenous glucose, animal feeding. To measure the fasting plasma leptin concentrations and the acute leptin response to intravenous glucose, animals were anesthetized with methoxyfluorane, and a catheter was inserted into the external jugular vein as previously described (27). The infusion cannulas were tunneled subcutaneously to the back of the neck. The catheter was infused with heparinized saline to prevent clot formation at the intravenous tip of the catheter, and the external end of the catheter was plugged. The animals were allowed to recover for 3 days. On the 3rd day, the animals were fasted overnight. The next morning, 0.5 ml of blood (for fasting leptin levels) was withdrawn through the catheter, and then 17 ml/kg of a glucose solution (40%) were infused through the catheter over a 2-min period. At 2, 3, 4, and 6 h after the glucose infusion, 0.5 ml of whole blood was withdrawn through the catheter. These time points were chosen to reveal the peak leptin concentrations after an intravenous glucose infusion, as shown in previous studies (31, 57). The catheter was then plugged. The animals were allowed to recover and to eat and drink ad libitum. One week later, the animals were fasted overnight. The following morning, 8 g of chow were given to each animal. Invariably, the total quantity of chow was ingested within 1 h. Three hours after the food was provided, the animals were killed, blood was collected, and the epididymal fat pads were weighed.

Initially, 5 animals from each strain were fed the control diet and 10 animals from each strain were fed the high-fat diet for 6 wk. All animals had DEXA measurements at the end of the 6 wk. During the surgery for insertion of the intravenous catheter, 4 of the 30 animals (1 F344 animal fed the control diet; 2 S-D and 1 F344 animals fed the high-fat diet) expired from complications of the anesthesia and surgery. The intravenous catheters were patent in only two S-D animals fed a control diet, three S-D animals fed a high-fat diet, three F344 animals fed a control diet, and six F344 animals fed a high-fat diet. Therefore, six more S-D animals were obtained from the supplier with identical weights as the original and fed either a control or high-fat diet for 6 wk. The final weights of the animals were similar to the weights of the animals from the initial experiment. After surgery, catheters were patent in two S-D animals fed the control diet and three S-D animals fed the high-fat diet. Results of the leptin response to intravenous glucose were pooled from both experiments (total of 4 S-D animals fed a control diet, 6 S-D animals fed a high-fat diet, 3 F344 animals fed a control diet, and 6 F344 animals fed a high-fat diet). DEXA was not performed on the second group of animals.

Plasma glucose and hormone measurements. Glucose, insulin, and leptin levels were measured in the plasma. Blood was collected in heparinized microtubes and centrifuged at 4°C, 10,000 g for 5 min, and the plasma was separated and immediately stored at −70°C. The plasma was thawed at room temperature before the measurements listed below were performed. Glucose was measured by an automated colorimetric glucose oxidase system (Vitros 700 System, Johnson and Johnson). Plasma levels of leptin and insulin were measured by rat radioimmunoassay kits (Linco-Re-
significant difference (74 ± 6 ng/ml vs. 69 ± 3 ng/ml). These findings demonstrated a relative insulin resistance in F344 animals than in the S-D animals (0.5 ± 0.06 vs. 0.2 ± 0.01 ng/ml, P < 0.0001). Nevertheless, the glucose concentrations in the fasting animals were not significantly different (74 ± 5 vs. 78 ± 3 ng/dl). These findings demonstrated a relative insulin resistance in the F344 strain.

The leptin concentrations were very low in fasting animals (Fig. 1A). After the glucose infusion, most of the leptin values in the S-D animals remained below the limits of detection of the leptin assay (0.5 ng/ml). *P = 0.008 for difference in strain by 2-way ANOVA.*

### Table 1. Comparison of weights of S-D and F344 rats fed a control and high-fat diet

<table>
<thead>
<tr>
<th>Diet</th>
<th>Initial Weight, g</th>
<th>Final Weight, g</th>
<th>% Weight Gain</th>
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<tbody>
<tr>
<td>Control diet</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>S-D</td>
<td>284 ± 6</td>
<td>433 ± 14</td>
<td>47 ± 3</td>
</tr>
<tr>
<td>F344</td>
<td>185 ± 3</td>
<td>310 ± 7</td>
<td>67 ± 2†</td>
</tr>
<tr>
<td>High-fat diet</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S-D</td>
<td>284 ± 6</td>
<td>431 ± 10</td>
<td>52 ± 1*</td>
</tr>
<tr>
<td>F344</td>
<td>182 ± 3</td>
<td>322 ± 4</td>
<td>77 ± 3*†</td>
</tr>
</tbody>
</table>

Values are means ± SE; n = 5 rats for control diet; n = 10 rats for high-fat diet. S-D, Sprague-Dawley rats; F344, Fischer 344 rats. *P = 0.008 for difference in diet; †P < 0.0001 for difference in strain by 2-way ANOVA.

usually above the limits of detection of the leptin assay, and the concentration peaked about 3 h after the intravenous glucose injection. Three hours after intravenous glucose injection or feeding, leptin concentrations in F344 rats were significantly higher than in the S-D rats (Fig. 1B). Within a strain of animal, the leptin concentration after an intravenous glucose bolus and after feeding was similar (Fig. 1B).

Responses to 6-wk diets. A second group of 2-mo-old S-D and F344 rats were fed either a control or a high-fat diet for 6 wk. At the end of this period, S-D rats had increased their weight by ~50%, regardless of whether the rats were fed the control or high-fat diet.
(Table 1). In the same period of time, F344 rats gained proportionately more weight than did the S-D rats. Furthermore, F344 rats fed a high-fat diet gained more weight than did the rats fed a control diet. The total amount of food consumed by the animals that received the control diet did not differ statistically from the amount consumed by the animals that were fed the high-fat diet (Table 2). However, because caloric content of the high-fat diet was greater than that of the control diet, the animals from both strains that were fed a high-fat diet consumed more calories during the 6-wk period than did the animals that were fed the control diet. When expressed as kilocalories consumed per gram of final body weight, the F344 rats consumed significantly more calories than the S-D rats.

After the animals received the control or high-fat diets for 6 wk, body composition was analyzed by DXA. The percentages of body fat in S-D and F344 rats fed control and high-fat diets over 6 wk are shown in Table 3. The initial percentage of body fat was slightly lower in the S-D strain than in the F344 strain and in animals chosen randomly for the high-fat diet. After 6 wk, the body fat percentage increased in all groups. In the S-D rats, the fractional increase in percent body fat was significantly higher in animals fed the high-fat diet than in the animals on the control diet, although the final weights were not statistically different. F344 rats gained fat at more than double the rate than did the S-D rats. As with the S-D strain, the F344 animals that were fed a high-fat diet accumulated significantly more body fat than did the F344 animals that were fed a control diet. At the end of 6 wk, the F344 rats that were fed the high-fat diet weighed more and had a higher percentage of body fat than did the rats that were fed the control diet. The high body fat percentage in the F344 animals, as measured by DXA, was reflected also in the weights of the epididymal fat pads. Although the F344 animals weighed less than the S-D animals, the epididymal fat in the F344 strain was nearly double that in the S-D strain (control diet, 9.5 ± 0.8 vs. 5.6 ± 0.3 g, P < 0.001; high-fat diet, 13.3 ± 0.7 vs. 7.8 ± 0.6 g, P < 0.001).

Over the 6-wk observation period, the fractional gain in fat-free mass (which was ~1.4 times the initial fat-free mass) was virtually constant in all groups (Table 4). By contrast, fat mass increased significantly more than did the fat-free mass. The F344 rats accumulated more fat than did the S-D rats, and the high-fat diet produced more body fat than did the control diet. The F344 rats were much more efficient in storing energy per control calorie ingested (0.74 ± 0.1 vs. 1.0 ± 0.01 kJ/kcal, P < 0.001) and per high-fat calorie ingested (0.72 ± 0.1 vs. 1.00 ± 0.02 kJ/kcal, P < 0.001) than were the S-D rats.

We next measured fasting glucose, insulin, and leptin levels and their responses to an intravenous glucose infusion in animals after 6 wk of either the control or high-fat diet. The observation times were chosen to maximize the detection of the leptin peak. Glucose and insulin concentrations were probably greatest within 1 h after the intravenous infusion of glucose; therefore, the glucose and insulin concentrations measured in our analysis were likely not to be peak responses. Fasting and postintravenous infusion glucose concentrations did not differ significantly in S-D and F344 animals (Fig. 2A). In addition, glucose concentrations did not differ in animals fed a control or high-fat diet (Fig. 2A). Insulin concentrations in the S-D animals usually remained below the detection of the insulin assay (Fig. 2B). However, fasting and postinfusion insulin concentrations in the F344 animals increased approximately fivefold over the 6-wk period (Fig. 2B). This finding suggested that the insulin sensitivity had deteriorated in both control and high-fat fed animals.

Fasting leptin concentrations were four- to fivefold higher in F344 rats than in the S-D rats (Fig. 3). The effect of the high-fat diet to raise fasting leptin concentrations was found to be of borderline significance (P = 0.065). To examine whether the strain of rat or diet had an effect on the acute leptin response to an intravenous glucose infusion, a two-factor repeated measures analysis of covariance model was run. It was found that the fasting leptin concentration was strongly associated with the magnitude of the acute leptin response, and therefore, the fasting leptin was included as a covariate in the model. The analysis demonstrated that, after the baseline fasting leptin level was adjusted for, the leptin response to intravenous glucose was larger in the F344 rats for all time points, and the pattern of the response between strains was roughly parallel over

Table 2. Comparison of total amount of food and energy consumed in S-D and F344 animals fed a control and high-fat diet

<table>
<thead>
<tr>
<th></th>
<th>Total Food Consumed, g</th>
<th>Total Kcal Consumed</th>
<th>Kcal/Final Body Weight</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Control diet</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S-D</td>
<td>1.033 ± 0.33</td>
<td>4.031 ± 129</td>
<td>9.3 ± 0.1</td>
</tr>
<tr>
<td>F344</td>
<td>0.806 ± 0.24†</td>
<td>3.143 ± 92†</td>
<td>10.1 ± 0.1†</td>
</tr>
<tr>
<td><strong>High-fat diet</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S-D</td>
<td>1.020 ± 0.23</td>
<td>4.896 ± 115†</td>
<td>11.3 ± 0.1*</td>
</tr>
<tr>
<td>F344</td>
<td>0.823 ± 0.12†</td>
<td>3.952 ± 60†</td>
<td>12.2 ± 0.1*†</td>
</tr>
</tbody>
</table>

Values are means ± SE; n = 5 rats for control diet; n = 10 rats for high-fat diet. *P < 0.0001 for diet; †P < 0.0001 for strain by 2-way ANOVA.
time (Fig. 3). Diet did not affect the leptin response, although it appeared that the peak leptin concentrations in the animals fed a high-fat diet occurred after the peak leptin concentrations in animals fed the control diet.

We also investigated whether the acute leptin response varied according to the method of delivery of calories. Plasma leptin concentrations 3 h after intravenous infusion of glucose or after per os feeding (Fig. 4) were significantly greater than the fasting plasma leptin concentrations in both strains of rat that were fed either diet. The peak serum leptin levels after intravenous glucose infusion or after per os feeding did not differ statistically in any of the experimental groups.

DISCUSSION

In this study, we have examined the steady-state leptin levels and the acute leptin responses to intravenous glucose infusion and per os feeding in two strains of rat with different propensities for weight and body fat gain. In animals that were 2 mo of age, the adipose tissue fraction of body weight was similar in the S-D and F344 strains (Table 3). Predictably, the fasting leptin concentrations, which reflect the stores of adipose tissue, were similarly low in these young, lean animals. However, young F344 animals had higher fasting insulin concentrations and a greater absolute leptin response to intravenous glucose than did young S-D animals (Fig. 1A). After 6 wk of either a control or high-fat diet, fasting leptin concentrations increased in proportion to the gain in percent body fat. Therefore, the animals fed a high-fat diet had a greater percentage of body fat and greater fasting leptin concentrations than did animals fed the control diet. The F344 animals gained a markedly greater percentage of body fat and had higher fasting leptin concentrations than did the S-D animals. The most novel finding of this work was that the acute leptin response to intravenous glucose was greater in the relatively obese F344 animals than in the S-D animals (Fig. 3). The leptin response was significantly higher in the F344 rats over all time points, even after adjusting for the fasting leptin level. In contrast, the leptin response to intravenous glucose was no different in the relatively obese animals in both strains that were fed a high-fat diet than in animals that were fed control diets.

Table 4. Body composition in S-D and F344 rats fed a control and high-fat diet

<table>
<thead>
<tr>
<th></th>
<th>Fat Mass</th>
<th>Fat-Free Mass</th>
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<tbody>
<tr>
<td></td>
<td>Initial</td>
<td>Final</td>
</tr>
<tr>
<td>Control diet</td>
<td></td>
<td></td>
</tr>
<tr>
<td>S-D</td>
<td>39.9 ± 2</td>
<td>74.8 ± 5</td>
</tr>
<tr>
<td>F344</td>
<td>27.1 ± 1‡</td>
<td>82.5 ± 2‡</td>
</tr>
<tr>
<td>High-fat diet</td>
<td></td>
<td></td>
</tr>
<tr>
<td>S-D</td>
<td>31.0 ± 4*</td>
<td>84.6 ± 4†</td>
</tr>
<tr>
<td>F344</td>
<td>25.3 ± 1*‡</td>
<td>103.2 ± 2†‡</td>
</tr>
</tbody>
</table>

Values are means ± SE. Total fat was calculated by multiplying % body fat (Table 2) by weight (Table 1), and fat-free mass was calculated by subtracting total fat from weight. *P = 0.06 for diet; †P < 0.001 for diet; ‡P < 0.001 for strain by 2-way ANOVA.

Fig. 2. Effect of iv glucose infusion on serum glucose and insulin concentrations in rats fed either a control (Ctrl) or high-fat (HF) diet. S-D (●, ○) and F344 (□, △) were fed either control (●, □) or high-fat (○, △) diets for 6 wk. Glucose was then infused intravenously, and serum concentrations of glucose (A) and insulin (B) were measured at above times as described in METHODS. Each data point is mean ± SE (n = 4, S-D control; n = 6, S-D high-fat; n = 3, F344 control; n = 6, F344 high-fat).
Leptin has been shown to reduce food intake and prevent the decrease in energy expenditure associated with weight loss, actions that should reduce body weight and percent body fat (19, 43, 45, 58). A few studies (1, 19) have shown that administration of leptin by injection or with constant subcutaneous infusion results in a dose-dependent decrease in body weight at incremental increases of plasma leptin levels within the physiological range. Only in more obese animals with higher fasting leptin levels does exogenous administration of leptin have less effect on weight loss (19). The higher fasting leptin concentrations and the more exaggerated absolute leptin response to intravenous glucose observed in the obese F344 strain are paradoxical to the biological actions of leptin. There have been a number of hypotheses to explain this paradox. Arch et al. (2) proposed that the leptin concentration-response curve may not be linear; as the leptin concentration rises, the biological response (i.e., satiety) may flatten. This concept fits nicely with the genetic-evolutionary theory espoused by Flier (15), who proposed that starvation and weight loss might be the main stimulus for leptin action. When body fat stores become depleted during starvation, low leptin levels stimulate food-seeking behavior and caloric intake and modulate other neuroendocrine processes (i.e., stimulate the hypothalamic-pituitary-adrenal axis and suppress the thyroid and reproductive axes; Ref. 15). When body fat stores increase, the higher leptin levels may or may not inhibit appetite and diminish total body weight. An alternative hypothesis for higher leptin levels in obese animals is that entry of leptin into cerebrospinal fluid may be limiting in obesity, which could result when the plasma leptin levels exceed the capacity of the transport system (9, 53). We believe that the first two hypotheses proposed above do not provide adequate reasons for the apparent lack of responsiveness on satiety of the increased levels of leptin observed in the obesity-prone F344 animals. Specifically, even though the young, lean F344 animals demonstrate a relatively exaggerated absolute leptin response to intravenous glucose, the absolute leptin levels are quite low and should be on the early, linear portion of the concentration-response curve. Likewise, it is doubtful that these low leptin levels observed early in the life of an F344 animal saturate the blood-brain leptin transport system. A third explanation for the leptin paradox is resistance to leptin action. We believe that F344 animals are most likely leptin resistant. As in animals with leptin receptor gene mutations (10, 25), the elevated serum leptin concentrations in young, lean animals point to a leptin-resistant state. The cause for the leptin resistance in F344 was not investigated. To date, the leptin receptor gene and the post...
receptor signaling in F344 animals have not been characterized.

Fasting leptin concentrations vary directly with the percentage of body fat. In this study, we have found that the more obese F344 animals had higher fasting leptin levels compared with the thinner S-D animals (Fig. 3). In addition, animals fed a high-fat diet tended to be fatter and have higher fasting leptin levels compared with animals fed a control diet. The incremental rise in plasma leptin levels above fasting levels in response to an intravenous infusion of glucose is caused by leptin secretory mechanisms that are independent from percent body fat. Despite differences in body fat, animals fed control or high-fat diets had similar leptin responses to intravenous glucose. However, F344 animals secrete more leptin in response to intravenous glucose. The most likely candidates for mediating the acute, incremental leptin response are insulin and/or glucose. In several rodent studies, insulin stimulates leptin gene expression and secretion. Insulin administered to a starved or lean rodent increases adipocyte leptin gene expression and serum leptin concentration (38, 50, 61). Leptin gene expression in streptozotocin-treated rats rapidly increases with insulin supplementation (33). Experiments in cultured adipocytes support the stimulatory role of insulin on leptin gene expression and secretion (28). Barr et al. (3) observed that insulin stimulates leptin secretion from isolated adipocytes within 10 min. In vitro studies (30, 40) have suggested that the acute leptin response is mediated by insulin-induced delivery and metabolism of energy-producing substrates within adipocytes. The exact mechanism of a greater leptin response to intravenous glucose in F344 animals than in S-D animals was not investigated. It is possible that the delivery and metabolism of energy-producing substrates in adipocytes in F344 animals were greater than in S-D animals despite the greater insulin resistance in the F344 strain; the F344 strain required higher insulin concentrations to perform this function. Alternatively, the greater leptin responses to intravenous glucose may simply reflect a strain variation in the trait of leptin production per unit of adipose tissue. Other yet unexplained mechanisms may be playing roles in the acute leptin response to intravenous glucose.

There are a number of recent reports showing dual regulation of leptin secretion in humans, and insulin and/or glucose may play a role in the body-fat independent regulation. A diurnal variation of plasma leptin has been observed in humans (55), and the diurnal rhythm of plasma leptin has been shown to be entrained to meal timing (52) and to be dependent on energy availability (24). Saad et al. (49) have shown that physiological insulinemia can acutely regulate plasma leptin and that the pattern of insulin secretion could explain the diurnal changes in leptin. The importance of insulin-induced glucose utilization in adiposity-independent leptin secretion was demonstrated by the greater amplitude of the diurnal variation of leptin secretion in human volunteers who were fed a high-carbohydrate, low-fat diet compared with an isocaloric high-fat, low-carbohydrate diet (22). Furthermore, a carbohydrate meal induced higher postprandial leptin levels than an isoeneric fat meal (46). There is some evidence to suggest that the acute leptin regulatory pathway may have some role in the pathophysiology of human obesity. Saad et al. have demonstrated that obesity is associated not only with higher leptin levels but also with blunted diurnal excursions and dampened pulsatility. Another study has shown that the gain in body fat is inversely related to the nocturnal rise in serum leptin level in young females (34). Therefore, because leptin secretion is dually and independently regulated, it is imperative to examine both pathways as etiologies for pathological states in body weight homeostasis.

We originally hypothesized that the F344 rat would be less prone than the S-D rat to body fat accumulation because the F344 rat has been studied in the past as a fat-independent model of aging-induced insulin resistance (14, 35). We have found, however, that, compared with the S-D rat, F344 rats fed both normal and high-fat diets over 6 wk gained markedly more body fat, measured both as the absolute amount (grams of fat) and as a fractional gain. Not only did F344 rats ingest more calories per gram body weight, but the F344 rats were more efficient than S-D rats in storing calories as weight and converted much more of the ingested calories into fat. A couple of previous investigations (4, 13) have found that F344 rats are a relatively thin strain. The reasons for the discrepancies in percent body fat in F344 animals may be related to the technique for measuring body fat or the source of the supplier of F344 animals. McDonald et al. (36) found body fat compositions and epididymal fat depots in F344 in amounts that were in close agreement with those in our study. Although we do not fully understand the wide variations in body fat in F344 obtained by different laboratories, we believe that caution should be used when studying F344 animals as a model for aging-induced insulin resistance. First, it may be difficult to differentiate obesity-induced insulin resistance from the effects of aging on insulin resistance. Second, the F344 animals demonstrate insulin resistance at even a young age. At 2 mo, F344 animals have at least threefold higher fasting insulin concentrations compared with age- and body fat-matched S-D animals despite comparable glucose concentrations. Third, F344 animals may be different when obtained from different suppliers.

In conclusion, not only are F344 animals insulin resistant throughout their lifespan, but also they appear to be resistant to the actions of leptin. When infused intravenously or into the third ventricles, leptin has been shown to reduce food ingestion in several animal models (43, 45, 58). Despite higher fasting serum leptin levels and acute absolute leptin responses to intravenous glucose, F344 animals ingested more calories per gram body weight compared with S-D animals. In addition, leptin has been shown to prevent decreases in energy expenditure in food-restricted animals (19), perhaps by inducing uncoupling proteins.
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(18, 48) or by other mechanisms mediated by the autonomic nervous system (23). One would predict, therefore, that animals that secrete more leptin would be less efficient in storing excess calories. However, despite higher fasting and meal-induced leptin concentrations, the F344 animals were more efficient in storing calories as weight and fat compared with S-D animals. Therefore, F344 animals appear to be resistant to the two main biological responses of leptin, in inhibiting appetite and in enhancing energy expenditure. The findings of insulin resistance and leptin resistance in a particular animal are not unusual and may have a common pathophysiology. Leptin has been shown to have salutary effects on glucose metabolism (5, 37, 42, 48). In addition, infusion of leptin has been demonstrated to improve insulin resistance in the leptin-deficient ob/ob (21) and lipodystrophic transgenic mice (54). It would be interesting to determine if supraphysiological concentrations of leptin administered to F344 animals would result in weight loss and improved insulin sensitivity compared with pair-fed F344 animals not given leptin.

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REFERENCES


