High-fat hypocaloric diet modifies carbohydrate utilization of obese rats during weight loss

MING C. CHA, JULIA A. JOHNSON, CHANG-YUN HSU, AND CAROL N. BOOZER

High-fat hypocaloric diet modifies carbohydrate utilization of obese rats during weight loss. 

The effects of fat content in the hypocaloric diet on whole body glucose oxidation and adipocyte glucose transport were investigated in two animal-feeding experiments. Diet-induced obese rats were food restricted to 75% of their previous energy intakes with either a high (45% by calorie) or a low (12% by calorie) corn oil diet for 9 wk (experiment 1) or 10 days (experiment 2). The losses of body weight (P < 0.05) and adipose depot weight (P < 0.05) were less in the 45% compared with the 12% fat group. During the dynamic phase of weight loss (day 10 of food restriction), plasma glucose and insulin concentrations were higher (P < 0.05) in the 45% than those in the 12% fat group. Whole body carbohydrate oxidation rate in response to an oral load of glucose was increased (P < 0.001) by food restriction in both dietary groups; however, carbohydrate oxidation rates were lower (P < 0.01) in the 45% than in the 12% fat-fed rats during the weight loss period. Adipocyte glucose transport was greater (P < 0.02) in the 45% than in the 12% fat group in an intra-abdominal adipose depot but not in subcutaneous fat. These data suggest that dietary fat content modifies whole body glucose oxidation and intra-abdominal adipocyte glucose uptake during weight loss.

energy restriction; glucose transport

HIGH DIETARY FAT INTAKE has been shown to impair glucose tolerance and decrease insulin sensitivity in both humans and animals (25, 34). High-fat diet feeding was also reported to decrease insulin suppressibility of hepatic glucose production (29) and whole body glucose disposal rate (19, 29), which is accompanied by impaired pancreatic β-cell function (21) and reduced insulin-stimulated skeletal muscle glucose transport (17, 24). Decreased adipocyte glucose transport with consumption of a high-fat diet was reported by some investigators (18, 28) but not by others (19, 36). The fatty acid composition of dietary fat may determine its effect on glucose metabolism and insulin action. Saturated fat intake has been shown to be associated with insulin resistance, whereas some of the adverse effects of high-fat diets on insulin sensitivity can be ameliorated by substitution of n-3 fatty acids (25, 35).

Energy restriction has been shown to enhance insulin sensitivity. Food restriction was reported to decrease hyperglycemia (7), reverse hepatic insulin resistance (2), and increase insulin-stimulated glucose transport in skeletal muscle (15) as well as in adipocytes (9). Improved insulin sensitivity by energy restriction has been suggested to be related to decreased visceral fat (2), although this may not be the only mechanism by which food restriction affects glucose tolerance.

It is generally accepted that a high-fat diet promotes obesity under ad libitum feeding conditions (5). Moreover, a reduction in fat intake without restriction of energy intake produced weight loss in obese subjects (1). However, there is no consensus regarding the role of quantitative dietary fat in energy deficit weight reduction diets. Although some authors have reported that a hypocaloric low-fat diet did not produce increased body weight loss compared with a high-fat calorie-restricted diet in obese humans (16, 31), data from animal studies reported by us (3) and others (4) showed a significant effect of dietary fat content on body weight and body fat loss during energy restriction. In a study with diet-induced obese rats that were food restricted isocalorically to diets containing graded levels of fat, body weight loss was lower in rats fed the high-fat diets, and body fat mass was proportional to the level of fat in the diet (3). The mechanism responsible remains to be elucidated.

It has been shown that a high-fat diet with a mixture of soybean, sunflower, and coconut oils as the oil source induced insulin resistance in skeletal muscle but not in adipocytes (36). A high-fat diet increased the sensitivity of insulin to stimulation of adipocyte glucose uptake in a mouse strain that is genetically sensitive to developing dietary obesity but decreased sensitivity in a strain resistant to diet-induced obesity (13). This evidence suggests that increased adipocyte insulin-related glucose transport could be responsible for lipid...
accretion in adipose tissue. We hypothesized that high content of fat in the energy-restricted diet may increase adipocyte glucose uptake and thereby contribute to maintenance of body adiposity during caloric restriction.

The purpose of this study was to investigate the effect of fat content in a hypocaloric diet on whole body ability to oxidize glucose and on adipocyte responsiveness to insulin of glucose transport in obese animals during weight loss. The relationship between adipocyte uptake of glucose and the size of adipose depot mass was particularly interesting.

MATERIALS AND METHODS

Animals and diet. Two experiments were conducted in this study. Male Sprague-Dawley rats purchased from Charles River Laboratories (Raleigh, NC) were used for both experiments. Eighteen animals (428 ± 7.0 g, 4 mo old) were used in experiment 1, and twelve animals (219.5 ± 6.6 g, 9 wk old) were used in experiment 2. Rats were housed individually in a temperature- (23 ± 2°C) and humidity-controlled (50% humidity) room with a 12:12-h light-dark cycle. Body weights were recorded three times per week throughout the experiments.

After habituation to a commercial rat chow (Lab Diet, PMI Nutrition International, St. Louis, MO) for 1 wk, rats were fed ad libitum a high-fat diet (45% fat, 35% carbohydrate, and 20% protein by energy, consisting of Crisco vegetable shortening, powered rodent chow, and casein) for 14 (experiment 1) or 20 wk (experiment 2) to gain weight (Fig. 1). Daily food intake for each rat, corrected for spillage, during the final 2 wk was used to calculate the amount of food to be given for the following food restriction period. At the end of the ad libitum high-fat-feeding period, rats in each experiment were weight matched into two dietary groups: nine in the ad libitum high-fat-feeding period, rats in each experiment were food restricted (Food res) to 75% of their previous energy intake by 10.2 ± 0.3 kcal/day for 9 wk. In experiment 1, 12 rats were fed the 45% fat diet for 9 wk to gain weight. Rats were then energy restricted to 75% (n = 9) of their previous intakes by 45 (n = 9) or 12% (n = 9) energy as fat for 9 wk. In experiment 2, 12 rats were fed the 45% fat diet for 20 wk to gain weight. Rats were then energy restricted to 75% of their previous intakes by 45 (n = 6) or 12% (n = 6) fat diet for 10 days.

Fig. 1. Experimental design. In experiment 1, 18 rats were fed ad libitum a diet with 45% energy as fat for 14 wk to gain weight. Rats were then food restricted (Food res) to 75% of their previous energy intakes by diets containing either 45 (n = 9) or 12% (n = 9) energy as fat for 9 wk. In experiment 2, 12 rats were fed the 45% fat diet for 20 wk to gain weight. Rats were then energy restricted to 75% of their previous intakes by 45 (n = 6) or 12% (n = 6) fat diet for 10 days.

Energy expenditure and carbohydrate oxidation. Energy expenditure and carbohydrate oxidation were examined in experiment 1 by indirect calorimetry. The calorimeter system consisted of a Magnos IV oxygen analyzer and a Uras 3G carbon dioxide analyzer (Hartmann Braun, Germany) and six Plexiglas metabolic chambers (School of Engineering, Columbia University) maintained in an environmental room with constant temperature, humidity, and a 12:12-h light-dark cycle. Rats were housed in chambers starting at 5 PM without food supply. Oxygen consumption rates were recorded from 9 PM to 9 AM for the fasting energy expenditure calculation. Animals were then given 10 ml of 25% glucose solution in food jars. The solution was entirely consumed by all of the animals within 20 min. Oxygen consumption and carbon dioxide production were recorded for the following 4 h consecutively to determine the carbohydrate oxidation rate. Energy expenditure was calculated using a standard formula (6). Carbohydrate oxidation was determined by formulas described previously (20). Fasting energy expenditure and carbohydrate oxidation were measured three times: on the day before food restriction (day 0), on the 10th day of food restriction (day 10) and in the week before animals were killed (week 9).

Adipocyte glucose transport and adipocyte size determination. In experiment 2, adipocytes were isolated from inguinal and mesenteric adipose tissue by collagenase digestion (12). Mean adipocyte size or weight (μg lipid/cell) was determined as previously described (11, 12). Rates of basal and insulin-
stimulated glucose uptake were measured in isolated inguinal and mesenteric adipocytes as described (12, 22), using porcine insulin (0, 25, 50, 100, and 400 mU). Data are expressed as glucose clearance rate in femtoliters per cell per second. Responsiveness of insulin to stimulate glucose transport was calculated as the absolute difference between the rate of glucose transport at a given concentration of insulin and the rate of glucose transport in the basal condition (when no insulin was present).

Other analyses. Plasma glucose concentration was analyzed with a Beckman Glucose Analyzer (Beckman Coulter, Fullerton, CA). Plasma insulin and leptin concentrations were determined by insulin and leptin radioimmunoassay kits for rat (Linco Research, St. Charles, MO). All analyses were conducted in duplicate. For the lepin assay, the sensitivity is 0.1 ng/ml, the limit of positivity is 10 ng/ml (100-μl sample size), and the CV is 5.3%.

Statistical methods. Data are presented as means ± SE. The effect of dietary fat was determined by one-way ANOVA using the SPSS general linear model program (SPSS, Chicago, IL). Analysis of covariance procedures were applied to the body weight and adipocyte glucose transport data to control for potential confounding factors or variations within treatment groups. The initial body weight was used as the covariate for the body weight analysis, and the sum of abdominal fat depot weight was employed as the covariate for the analysis of adipocyte glucose transport. Differences between means were considered to be significant at P < 0.05.

RESULTS

Body weight decreased in response to food restriction in both dietary groups. In experiment 1, body weight loss was greater (P < 0.03) in the 12% than in the 45% fat group after 2 wk of food restriction (45.7 ± 2.2 vs. 31.9 ± 1.9 g in the 12% vs. 45% fat group, respectively; Fig. 2A). This significant difference between the 12 and 45% fat groups persisted for the remainder of the dietary restriction period. Body weight loss on the 10th day of energy restriction was not statistically different between the two dietary groups in either experiment (35.1 ± 3.3 vs. 23.2 ± 3.0 g in the 12 vs. 45% fat group, respectively; Table 2). Epididymal and retroperitoneal fat pads were heavier (P < 0.05) in the 45% than those in the 12% fat-fed animals after 10 days of food restriction (experiment 2). This difference was not observed in other fat depots examined. However, after 9 wk of food restriction (experiment 1), all adipose depots investigated were heavier (P < 0.01) in the 45% compared with the 12% fat groups. Inguinal and mesenteric adipocyte sizes were not different between the two dietary groups after 10 days of food restriction (experiment 2).

Table 2. Body and tissue weights and adipocyte size of rats food restricted to 75% of their previous ad libitum intakes by 12 or 45% fat diets isocalorically for 9 wk (experiment 1) or 10 days (experiment 2)

<table>
<thead>
<tr>
<th></th>
<th>12% Fat</th>
<th>45% Fat</th>
<th>12% Fat</th>
<th>45% Fat</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial body weight</td>
<td>549.3 ± 13.5</td>
<td>546.7 ± 12.1</td>
<td>597.7 ± 30.24</td>
<td>602.0 ± 18.3</td>
</tr>
<tr>
<td>Day 10 body weight</td>
<td>514.2 ± 12.0</td>
<td>525.3 ± 11.7</td>
<td>551.2 ± 20.7</td>
<td>575.5 ± 18.6</td>
</tr>
<tr>
<td>Final body weight</td>
<td>486.4 ± 11.3a</td>
<td>509.5 ± 11.4†</td>
<td>551.2 ± 20.7</td>
<td>575.5 ± 18.6</td>
</tr>
<tr>
<td>Liver</td>
<td>14.9 ± 0.7</td>
<td>13.7 ± 0.6</td>
<td>17.9 ± 1.2</td>
<td>16.5 ± 0.8</td>
</tr>
<tr>
<td>Soleus muscle</td>
<td>0.17 ± 0.006</td>
<td>0.16 ± 0.007</td>
<td>0.22 ± 0.01</td>
<td>0.21 ± 0.01</td>
</tr>
<tr>
<td>Epididymal fat</td>
<td>2.57 ± 0.15†</td>
<td>3.52 ± 0.24†</td>
<td>6.09 ± 0.80*</td>
<td>9.02 ± 0.59†</td>
</tr>
<tr>
<td>Retroperitoneal fat</td>
<td>1.82 ± 0.10a</td>
<td>3.16 ± 0.28†</td>
<td>8.15 ± 1.33†</td>
<td>12.7 ± 1.43†</td>
</tr>
<tr>
<td>Omental fat</td>
<td>0.67 ± 0.05*</td>
<td>1.12 ± 0.12†</td>
<td>1.65 ± 0.43</td>
<td>2.10 ± 0.23</td>
</tr>
<tr>
<td>Mesenteric fat</td>
<td>2.13 ± 0.15†</td>
<td>3.87 ± 0.35†</td>
<td>9.02 ± 1.55</td>
<td>9.84 ± 1.29</td>
</tr>
<tr>
<td>Sum of intra-abdominal fat depots</td>
<td>7.59 ± 0.36a</td>
<td>11.8 ± 0.9†</td>
<td>23.7 ± 3.1†</td>
<td>33.5 ± 2.9†</td>
</tr>
<tr>
<td>Inguinal fat</td>
<td>1.36 ± 0.11a</td>
<td>2.05 ± 0.21†</td>
<td>5.29 ± 1.01</td>
<td>5.67 ± 0.57</td>
</tr>
<tr>
<td>Adipocyte size (μg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inguinal</td>
<td>‡</td>
<td></td>
<td>0.50 ± 0.09</td>
<td>0.44 ± 0.04</td>
</tr>
<tr>
<td>Mesenteric</td>
<td></td>
<td></td>
<td>0.36 ± 0.05</td>
<td>0.38 ± 0.04</td>
</tr>
</tbody>
</table>

Values are means ± SE in grams. For each parameter within the same experiment, means with different superscripts are significantly different. ‡Not measured.
Fasting glucose, insulin, and leptin concentrations are shown in Fig. 3. Glucose levels on day 10 were higher (P < 0.02) in the 45% (131.2 ± 4.7 and 120.4 ± 4.3 mg/dl for animals in experiments 1 and 2, respectively) than those in the 12% groups (112.4 ± 3.2 and 100.2 ± 3.4 mg/dl). Similarly, insulin concentrations were higher (P < 0.05) in the 45% (2.26 ± 0.52 and 3.51 ± 0.60 ng/ml for animals in experiments 1 and 2, respectively) than in the 12% fat-fed animals (0.86 ± 0.08 and 2.44 ± 0.54 ng/ml, respectively). Glucose concentrations decreased (P < 0.05) from day 10 to week 9 (115.6 mg/dl) in the 45 but not in the 12% group in experiment 1. However, insulin concentrations were increased (P < 0.05) in the 12% group but decreased (P < 0.05) in the 45% fat-fed animals, from day 10 to week 9 (1.48 ± 0.20 and 1.87 ± 0.19 ng/ml for 12 and 45% fat groups at week 9, respectively). In experiment 1, plasma leptin concentrations on day 10 were 150% higher (P < 0.03) in the 45% group (7.56 ± 1.60 ng/ml) than in the 12% fat (3.06 ± 0.67 ng/ml)-fed animals. Leptin levels decreased (P < 0.05) from day 10 to week 9 in the 45% but not in the 12% fat group. However, leptin concentrations remained higher (P < 0.01) at week 9 in the 45% group (4.02 ± 0.56 ng/ml) compared with the 12% fat-fed (2.14 ± 0.25 ng/ml) animals. In experiment 2, the difference in leptin levels between the two dietary groups on day 10 was not statistically significant.

Twelve-hour fasting energy expenditure and carbohydrate (CHO) oxidation rate after an oral glucose load in rats food restricted to 75% of their previous ad libitum energy intakes with either a 12 or a 45% fat diet for 9 wk. Values are means ± SE. Means associated with different letters are significantly different.

Fig. 4. Data collected from experiment 1. Twelve-hour fasting energy expenditure and carbohydrate (CHO) oxidation after an oral glucose load in rats food restricted to 75% of their previous ad libitum energy intakes with either a 12 or a 45% fat diet for 9 wk. Values are means ± SE. Means associated with different letters are significantly different.
and 31.2 ± 4.3 fl·cell⁻¹·s⁻¹ for the 45 and 12% fat groups, respectively) and insulin-stimulated glucose transport at different insulin concentrations were higher (P < 0.02) in the 45% group compared with those in the 12% fat group in the mesenteric depot. These differences in glucose transport in mesenteric adipocytes persisted when data were expressed in terms of cell surface area (data not shown). However, these differences between diet groups were not observed in inguinal adipocytes. Responsiveness of insulin to stimulate glucose transport in mesenteric and inguinal adipocytes treated with different concentrations of insulin. Adipocytes served in inguinal adipocytes. Responsiveness of insulin to stimulate glucose transport in mesenteric and inguinal adipose depots was not statistically different (P > 0.08) between the two dietary groups at 25, 50, and 100 pM insulin (data not shown); however, responsiveness at 400 pM insulin was higher in the 12% group (27.3 ± 9.2 and 7.7 ± 3.3 fl·cell⁻¹·s⁻¹ in mesenteric and inguinal, respectively) compared with that in the 45% fat group (23.7 ± 3.7 and 6.3 ± 1.8 fl·cell⁻¹·s⁻¹, respectively).

**DISCUSSION**

Weight reduction in obesity has been shown to improve insulin sensitivity (14). The present study demonstrated that a high content of dietary fat rich in n-6 fatty acids had an adverse effect on the improvement of whole body ability to oxidize glucose in obesity in response to energy restriction. Despite relative peripheral insulin resistance, adipocyte glucose transport in intra-abdominal adipose tissue of animals food restricted to the high-fat diet was greater than that of those fed the low-fat diet. Increased partitioning of glucose into the fuel storage pathways in visceral adipose tissue may contribute to the limited loss of intra-abdominal fat mass associated with consumption of the high-fat hypocaloric diet.

Intake of dietary fat is closely associated with the development of obesity (1, 5). We (3) have previously demonstrated that the amount of fat in an energy-restricted diet was inversely related to the loss of body weight and body fat in diet-induced obese animals. In the present study, the dietary obese animal model in the two experiments differed in the age of the animals and in the length of time of feeding the high-fat diet to induce obesity. Body weight and fat mass loss corresponded to the initial body weights of rats in the two experiments, but in both experiments these losses were less in the 45% than in the 12% fat group, consistent with our previous findings. The differences in body weight loss occurred during the first 3 wk of food restriction, the dynamic phase of weight loss. In experiment 1, body weight decreased ~40 g in the 45% fat group and 60 g in the 12% fat group during the dynamic weight reduction period. Body weight of both groups then remained constant for the remaining weeks of food restriction, suggesting that decreased energy expenditure had matched the reduced energy intake. Energy balance was thus achieved at a lower level of energy intake.

It has been reported that, in rats, chronic high-fat feeding reduced whole body glucose disposal rate and impaired skeletal muscle glucose metabolism (29). In contrast, energy restriction enhanced insulin-stimulated glucose transport by skeletal muscle, resulting in increased whole body insulin sensitivity (15). In the present study, whole body insulin sensitivity was examined by testing the carbohydrate oxidation rate in response to an oral glucose challenge. Because in rodents >80% of glucose is oxidized in muscle (10), this finding mainly reflects skeletal muscle insulin sensitivity. The carbohydrate oxidation rate was lower in both diet groups before food restriction (day 0), indicating that insulin resistance was induced by long-term high-fat feeding. Insulin sensitivity was improved by energy restriction in both dietary groups, as shown both by increased whole body ability to oxidize glucose and by decreased circulating glucose and insulin levels. However, the glucose oxidation rate was transiently lower, and glucose and insulin concentrations were correspondingly higher, in the 45% fat group during the dynamic phase of weight loss, suggesting that the high-fat diet delayed the improvement in insulin sensitivity with energy deficit.

Rats fed the hypocaloric high-fat diet had more intra-abdominal fat and, to a lesser extent, more subcutaneous fat, compared with animals fed the low-fat diet. Intra-abdominal and subcutaneous adipose tissues are known to display important metabolic differences. It has been shown in obese rats that exercise reduced the activity and mRNA levels of acyl-CoA synthetase and the mRNA level of lipoprotein lipase (LPL) in mesenteric fat but did not affect these parameters in subcutaneous adipose tissue (33). Ventromedial hypothalamic lesions, which produce obesity in rats, increased acyl-CoA synthetase activity in mesenteric but not subcutaneous fat (32). Consistent with these previous findings, in this study, basal and insulin-stimulated adipocyte glucose transport were affected by the high-fat diet only in mesenteric but not in...
inguinal (subcutaneous) fat. It appears that the high-fat hypocaloric diet shunts glucose toward fuel storage pathways in visceral adipose tissue, favoring fat preservation. It has been shown that the magnitude of reduction of visceral fat is closely related to the improvement in whole body insulin sensitivity by food restriction (2). The limited loss of visceral fat could be responsible for the slowed improvement of whole body glucose oxidation in the energy-restricted high-fat-fed animals. A recent study demonstrating that selective insulin resistance in muscle promoted redistribution of substrates to adipose tissue, thereby contributing to increased adiposity and development of insulin resistance (23), supports our interpretation. We (3) have previously demonstrated that rats fed a restricted amount of high-fat vs. low-fat diet had a higher LPL activity in their abdominal (retroperitoneal) adipose tissue. Present results provide an additional mechanism to explain our findings of diminished body fat loss in animals food restricted to a high-fat diet.

Dietary fatty acid composition has been shown to be important for glucose metabolism and insulin sensitivity. Saturated fatty acids, compared with monounsaturated or polyunsaturated fat, appear to be more deleterious with respect to dietary fat-induced insulin insensitivity in humans (35). Feeding diets high in n-3 fatty acids corrected the hyperinsulinemia in insulin-resistant rats (26) and resulted in a significant decline in both basal and insulin-stimulated glucose uptake in adipocytes (27). In another study, glucose transport and subsequent utilization were greater in epididymal fat cells of rats fed beef tallow compared with those of animals fed corn oil or menhaden oil. There were no differences in these measurements between cells from corn oil- or menhaden oil-fed rats (30). In the present study, the oil source for both diets was corn oil, which is rich in n-6 fatty acids (mainly linoleic acid). Because the amount of n-3 fatty acids (mainly linolenic acid in the corn oil) in those diets is minimal (about 2 g/kg in the 45% fat diet and 0.5 g/kg in the 12% fat diet), the present data indicate that a diet high in n-6 fatty acids delayed the improvement of whole body glucose metabolism and the reduction of intra-abdominal adipose depot mass in response to energy restriction.

A previous study demonstrated that weight reduction improved insulin sensitivity in proportion to the degree of weight loss (14). In the present study, longer-term food restriction abolished the differences in circulating glucose, insulin, and whole body glucose oxidation between the two dietary groups despite persistent differences in body weight and fat mass size. It has been shown that energy restriction diluted the changes related to dietary fat type in membrane phospholipid fatty acid composition (8). Compared with the differences in body composition or in dietary fat intakes, the effect of prolonged food restriction may be a more potent effector of whole body insulin sensitivity. It is also possible that a negative energy balance may be needed to show the difference in insulin sensitivity as a function of different extent of body weight and fat mass loss.

Although adipocyte glucose metabolism was not examined beyond day 10, we assume that increased glucose transport in visceral fat continued, at least during the rest of the weight loss period, as evidenced by increased differences in both body weight and fat mass loss between the two dietary groups. Glucose transport in inguinal adipose tissue at day 10 was not influenced by the high-fat diet; however, inguinal fat mass weight after 9 wk of food restriction was 51% heavier in the 45% group than in the 12% fat-fed animals. Although less than the diet effect on mesenteric fat (82% heavier in the 45 vs. the 12% fat group), consistent with our previous report of increased central distribution of fat with a hypocaloric high-fat diet (3), the increased retention of fat in inguinal depots here is significant. Potential mechanisms for a diet-induced effect on peripheral adipose depots include alterations in LPL activity, hormone-sensitive lipase activity, or ß-adrenergic agonist-induced lipolysis. The exact mechanism remains to be elucidated.

In conclusion, dietary intake of a moderately high amount of fat (mainly n-6 fatty acids) delayed the improvement of whole body insulin sensitivity in obesity in response to energy restriction. Increased glucose uptake in visceral adipocytes may contribute to the limited loss of visceral adipose tissue, which is, in turn, related to the slowed recovery of whole body insulin sensitivity associated with consumption of the high-fat hypocaloric diet. Results from the present study thus suggest the importance of decreasing the content of n-6 fatty acid-enriched fat in hypocaloric weight reduction diets.

The technical support of Lina Basilio and Yim Dam at St. Luke’s-Roosevelt Hospital is greatly appreciated.

This research was supported by National Institute of Diabetes and Digestive and Kidney Diseases Grants DK-49853 and P30 DK-26687.

REFERENCES

13. Eberhart GP, West DB, Boozer CN, and Atkinson RL.
15. Ferrannini E and Camastra S.
17. Han DH, Hansen PA, Host HH, and Holloszy JO.