Maternal high-fat diet consumption results in fetal malprogramming predisposing to the onset of metabolic syndrome-like phenotype in adulthood

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Srinivasan, Malathi, Subhash D. Katewa, Arivazhagan Palaniyappan, Jignesh D. Pandya, and Mulchand S. Patel. Maternal high-fat diet consumption results in fetal malprogramming predisposing to the onset of metabolic syndrome-like phenotype in adulthood. Am J Physiol Endocrinol Metab 291: E792–E799, 2006. First published May 23, 2006; doi:10.1152/ajpendo.00078.2006.—Chronic consumption of a high-fat (HF) diet by female rats in their postweaning period resulted in significant increases in body weight and plasma levels of insulin, glucose, and triglycerides during pregnancy compared with female rats consuming a standard rodent laboratory chow (LC). On gestational day 21, plasma insulin levels and the insulin secretory response of islets to various secretagogues were significantly increased in HF fetuses. The HF male progeny weaned onto LC (HF/LC) demonstrated increases in body weight from postnatal day 60 onward. In adulthood, HF/LC male rats were significantly heavier than controls, had increased plasma levels of insulin, glucose, free fatty acids, and triglycerides, and demonstrated glucose intolerance. HF/LC male islets secreted increased amounts of insulin in response to low glucose concentrations, but their response to a high glucose concentration was similar to that of LC/LC islets. In another set of experiments, when the male progeny of HF female rats were weaned onto a high-sucrose diet (HF/HSu), their metabolic profile was further worsened. These results indicate that chronic consumption of a HF diet by female rats malprograms the male progeny for glucose intolerance and development of increased body weight in adulthood. The long-term high-fat feeding to female rats employed in this study bears resemblance to the dietary habits in Western societies. The results of this study implicate dietary practices of women in the etiology of the present epidemic of human obesity and related disorders.

fat-enriched diet; intrauterine environment; fetal hyperinsulinemia; adult-onset obesity; glucose intolerance

OVER THE PAST TWO TO THREE DECADES, the prevalence of obesity has increased steadily and has now reached epidemic proportions in developed countries. In the US alone, more than two-thirds of the adult population have been classified as overweight, with about one-half of them being obese (27). Obesity is a risk factor for the onset of metabolic diseases in adulthood, including type 2 diabetes and cardiovascular diseases (13). More than 80% of people with diabetes are overweight or obese, indicating a close correlation between being overweight and having diabetes (35). It is now recognized that genetics alone cannot explain the unprecedented increase in the number of overweight/obese individuals worldwide. Several environmental factors have been implicated in the etiology of obesity. Hales and Barker (17) coined the term “fetal programming,” which was based on data from several epidemiological studies, to demonstrate that metabolic diseases have their origin in early life nutritional experience during gestation and lactation. A nutritional stress/stimulus occurring during the period of fetal development results in adaptive responses in the fetus (metabolic programming) that are advantageous for its survival in a less than optimal environment. In the long term, such adaptations result in unfavorable outcomes due to malprogramming of many aspects of anatomy, physiology, and metabolism predisposing the fetuses for the onset of metabolic syndrome in adulthood (6). Results from animal models where an altered nutritional condition in the female during pregnancy and/or lactation (low-protein diet, total caloric restriction, and gestational diabetes) was used showed similarities to the observations obtained from human epidemiological reports (7, 18, 28, 30, 39).

Although studies on energy balance have revealed that body weight is tightly regulated, when animals or humans consistently consume a diet enriched in fat-derived calories (but otherwise nutritionally adequate), the amount of stored fat that they maintain progressively increases. These results demonstrate a positive correlation between increased percentage of fat-derived calories in the diet and the incidence of obesity (9). In the Westernized world, there is an increase in the abundance and accessibility of fat- (largely saturated fats) and carbohydrate-dense (largely sucrose and purified starch) foods. Such foods, present in increasingly processed and palatable forms, have induced alterations in the feeding habits of people, contributing to the present prevalence of obesity and its related disorders. Such changes in feeding habits could have long-term consequences not only for these individuals (same generation) but also for subsequent generations due to the altered intrauterine environment encountered by the fetuses in the overweight/obese females during child-bearing age.

Several animal studies (40, 41) have demonstrated that prolonged feeding of a high-fat (HF) diet to normal rats results in increased body weight, hyperinsulinemia, and insulin resistance in their adulthood. The long-term consequences on the adult progeny due to consumption of a HF diet by female rats include abnormalities such as impaired glucose homeostasis, cardiovascular dysfunction, and alterations in hypothalamic energy circuitry and liver lipid metabolism (11, 15, 21). In those studies, HF diet feeding to female rats was limited to the period of gestation and, in some cases, to both gestation and

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lactation, and the effects were demonstrated in only the adult progeny (11, 15, 21). Although the prolonged consumption of a HF diet by female rats bears similarities to the human situation, especially in Western societies, the immediate and long-term consequences for the progeny due to such a dietary practice have not been investigated in the rat. Therefore, in the present study we have investigated 1) the consequences of prolonged consumption (beginning from the immediate postweaning period) of a HF diet in female rats and 2) the malprogramming effects in both term fetuses and adult males to assess the immediate and long-term effects due to the altered intrauterine environment in the HF female. Our results demonstrate that prolonged consumption of a HF diet by female rats results in an adverse maternal intrauterine environment, predisposing the fetuses to metabolic malprogramming. These early fetal maladaptations eventually predispose them in their adult life to the metabolic syndrome-like phenotype (increased body adiposity, chronic hyperinsulinemia, glucose intolerance, and hyperlipidemia). These observations suggest that mother’s health during both prepregnancy and pregnancy may be a contributing factor to the rapidly developing obesity epidemic.

**MATERIALS AND METHODS**

**Maternal HF dietary treatment.** The Institutional Animal Care and Use Committee approved all animal protocols. Twenty-four-day-old Sprague-Dawley female rats were obtained from Zivic Miller (Zellenpole, PA). They were randomly divided into two groups and were fed ad libitum either a HF diet [Product no. F3282; BioServ, Frenchtown, NJ (designated as HF female rats)] or standard rodent laboratory chow (LC) [16% protein rodent diet; Harlan Teklad, Madison, WI (designated as LC female rats)]. The macronutrient composition and caloric distribution in the HF and LC diets are detailed in Table 1. In the HF diet, 59.5 and 24.4% of the total calories were derived from fat and carbohydrates, respectively. The LC diet contained 10.9 and 70% of total calories from fat and carbohydrate, respectively. In the HF diet, there was a decrease in polysaccharide content with a concomitant increase in the availability of simple sugars and only negligible amounts of fiber. The increase in the fat content in the HF diet was characterized by substantial increases in the availability of palmitic and oleic acids without marked changes in the content of linoleic and linolenic acids. The energy availability per gram of the diet was 5.3 kcal for HF and 3.2 kcal for LC.

The HF and LC female rats were weighed on postnatal day 24, and their body weights were monitored every 10 days from postnatal day 30 onward. Tail blood was collected every 20 days between 9 and 10 AM from postnatal day 40 for plasma insulin measurements.

**Fetal studies.** HF and LC female rats (6–8 rats/group) were bred with normal Sprague-Dawley male rats (Zivic Miller) on approximately postnatal day 120. Male rats used for breeding were fed a standard rodent LC from postnatal day 24. The presence of a vaginal plug confirmed pregnancy. Food intake for the pregnant HF and LC rats was monitored from gestational days 14 to 21. For fetal studies, pregnant HF and LC female rats were killed on gestational day 21. The number of pups in each litter and their body weights were recorded. Trunk blood was collected in heparinized tubes and centrifuged, and the plasma was stored at −80°C for hormone and substrate analyses. Fetal blood from all the fetuses in the same litter was pooled, and their plasma was stored as indicated above. To determine the fetal pancreatic insulin content, the pancreas from one fetus of each mother was weighed and homogenized in 500 μl of acid-ethanol solution (126 mM ethanol, 0.005 N HCl). The pancreatic extracts were centrifuged and the supernatants stored at −20°C until assayed for insulin.

For studies on insulin secretion by islets isolated from pancreata of HF and LC fetuses, the protocol described by Cherif et al. (12), with some modifications, was used. Briefly, RPMI 1640 supplemented with 11 mM glucose, 10% heat-inactivated fetal bovine serum and antibiotics (2,000 U/ml penicillin, 0.3 g/l streptomycin) was used for the isolation and culturing of fetal islets, and all steps were carried out under aseptic conditions. Pancreata from fetuses of the same mother were pooled, minced, and digested with collagenase (Sigma type V, 1.5 mg in 3 ml of RPMI 1640) at 37°C in a shaking water bath at 120 rpm for 6–8 min. The enzyme reaction was stopped by the addition of ice-cold RPMI 1640 medium. The tissue digestate was washed three times with ice-cold medium, resuspended in 10 ml of medium, and stirred at low speed at room temperature for 30 min. After a brief centrifugation at low speed, the islets were resuspended in 10 ml of RPMI 1640 medium and distributed into 60-mm culture dishes and cultured for ≤7 days in a humidified atmosphere of 5% CO2 in air at 37°C. After the first 48 h, the medium was replaced every 24 h. The islets were hand picked using an inverted stereomicroscope, and the insulin secretory response of these islets to 5.5 (basal) and 16.7 mM (high) glucose and 5.5 mM glucose plus either 10 mM arginine or 10 mM leucine at 60 min was determined as described by Xia and Laychock (42).

**Characterization of the progeny.** For studies on the progeny, HF and LC female rats (6–8 rats/group) were bred with normal male rats. After delivery, the litter size was adjusted to 11 pups/dam within 24 h. On postnatal day 24, 2–3 male rats from each litter were weighed and weaned onto standard rodent LC. The progeny of LC and HF females weaned onto standard rat chow were referred to as LC/LC and HF/LC, respectively. Their body weights were recorded every 10 days from postnatal day 30 onward. Tail blood was collected every 20 days between 9 and 10 AM from postnatal day 40 onward for determination of plasma insulin levels. A glucose tolerance test (GTT) was carried out in 90-day-old HF/LC and age-matched LC/LC male progeny, as described earlier (8). Rats were fasted overnight, and glucose (2 g/kg body wt) was injected intraperitoneally in awake rats. Blood glucose levels were monitored in tail blood samples using a glucometer (Ascencia Elite; Bayer, Mishawaka, IN) before and 15, 30, 45, 60, 90, 120, and 180 min after glucose injection. Blood samples were also collected from the tail vein sequentially prior to and 15 and 60 min after the injection of glucose for measurement of plasma insulin levels. On postnatal day 120, HF/LC and LC/LC rats were killed and trunk blood collected in heparinized tubes. Plasma was separated by centrifugation and stored at −80°C until assayed for insulin, glucose, free fatty acids (FFAs), and triglycerides. Pancreatic islets were isolated from 120-day-old HF/LC and LC/LC male progeny by collagenase (Sigma, St. Louis, MO) digestion, as described previously (1, 42). Islets were hand picked under a stereomicroscope and used for studies on their insulin secretory response to 1, 5.5, or

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**Table 1. Macronutrient composition of a standard rodent LC and a HF diet**

<table>
<thead>
<tr>
<th>Macronutrient Composition</th>
<th>Standard Rodent, g/kg</th>
<th>HF, g/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carbohydrate,</td>
<td>609 (70)*</td>
<td>327 (24.4)</td>
</tr>
<tr>
<td>Polysaccharides</td>
<td>451</td>
<td>196</td>
</tr>
<tr>
<td>Mono- and disaccharides</td>
<td>50</td>
<td>149</td>
</tr>
<tr>
<td>Fat,</td>
<td>42 (10.9)</td>
<td>355 (59.5)</td>
</tr>
<tr>
<td>Palmitic acid</td>
<td>6.1</td>
<td>90</td>
</tr>
<tr>
<td>Oleic acid</td>
<td>8.9</td>
<td>162</td>
</tr>
<tr>
<td>Linoleic acid</td>
<td>23.1</td>
<td>35.5</td>
</tr>
<tr>
<td>Linolenic acid</td>
<td>1.5</td>
<td>0.36</td>
</tr>
<tr>
<td>Protein,</td>
<td>167 (19.2)</td>
<td>200 (16.2)</td>
</tr>
<tr>
<td>Fiber,</td>
<td>39</td>
<td>1</td>
</tr>
<tr>
<td>kcal/g</td>
<td>3.2</td>
<td>5.3</td>
</tr>
</tbody>
</table>

LC, laboratory chow; HF, high fat; kcal/g, total amount of calories obtained per gram of each diet. *Numbers in parentheses indicate %calorie content in the diets.
16.7 mM glucose. Aliquots of the incubation medium were collected at 10 and 60 min to determine the early- and late-phase insulin secretory responses. The results are expressed as insulin secreted per 10 islets at 10 and 60 min.

To investigate whether a hypercaloric diet from the time of weaning will worsen the phenotype, 2–3 male progeny of HF and LC female rats were weaned onto a high sucrose (HSu) diet (67% carbohydrate, 7% fat, and 18% protein, Product no. F4439; BioServ) and were designated as HF/HSu and LC/HSu. A HSu diet was selected to avoid the HF diet-mediated responses and to provide a diet that mimics the sugar-enriched foods that are consumed in large proportions, especially in Western societies. For these studies, body weights were recorded in the postweaning period as described above for chow-fed progeny rats. GTT was carried out in 90-day-old rats as described above. On postnatal day 120, these rats were killed and measurements similar to those described above for chow-fed progeny were carried out.

**Plasma parameters.** Insulin assay was carried out using the radioimmunoassay kit (Linco, St. Charles, MO). The within- and between-assay coefficients of variation for the insulin assay were 4 and 10%, respectively. Plasma levels of glucose (Thermo Trace, Arlington, TX), FFAs (Roche, Indianapolis, IN), and triglycerides (Thermo Trace) were carried out according to manufacturers’ recommendations.

**Statistical analyses.** The results are expressed as means ± SE of the indicated number of animals for each experiment. For multiple comparisons, one-way analysis of variance (ANOVA), followed by post hoc analysis using the Student-Newman-Keuls test, was used to compare the significance of the difference in the means. For studies on the progeny, only the following comparisons are reported for evaluating the significance in the difference between the means: HF/LC vs. LC/LC; HF/HSu vs. LC/LC; HF/HSu vs. LC/HSu; and HF/HSu vs. HF/LC. Whenever only two groups were being compared, Student’s t-test was used for analyses of the significance of the difference in the means.

**RESULTS**

**Maternal parameters.** The average body weights of the 24-day-old LC and HF female rats were not significantly different (60.8 ± 0.5 g for LC and 62.7 ± 0.2 g for HF, n = 15). Up to postnatal day 60, the body weights were similar for the HF and LC female rats. From postnatal day 70 onward, the HF female rats were significantly heavier than the age-matched LC female rats (Fig. 1A). The differences in the body weights between the two groups of rats were larger with the increasing age of the rats (Fig. 1A). Although the plasma insulin levels in the HF female rats were similar to those of the LC female rats on postnatal day 60, significant increases were observed in plasma insulin levels in the HF female rats on postnatal days 80 and 100 (Fig. 1B).

On postnatal day 100 (prepregnancy), body weight and plasma insulin levels were significantly higher in HF female rats, without changes in plasma levels of glucose, FFAs, and triglycerides compared with age-matched LC female rats in the fed state (blood sampled between 9 and 10 AM; Table 2); HF and LC female rats were bred with normal male rats when they were ~4 mo-old and killed on gestational day 21. HF female rats were ~20% heavier than age-matched LC rats on gestational day 21 (Table 2). Plasma glucose and triglyceride levels showed significant increases of ~34 and 68%, respectively, in HF female rats on gestational day 21 (Table 2). There were no significant changes in plasma FFAs on gestational day 21. The average weights of the placenta from HF and LC female rats were similar on gestational day 21 (0.53 ± 0.02 g for HF and 0.53 ± 0.01 g for LC female rats; n = 6 mothers).

**Fetal parameters.** The average litter size was not significantly different between the two groups of rats (14.8 ± 0.9 for LC group and 16 ± 1 for HF group). Average fetal body weights were not significantly different between the two groups of rats on gestational day 21 (Table 2). There was a nearly twofold increase in the plasma insulin levels in HF fetuses compared with LC fetuses (Table 2). An ~43% increase in pancreatic insulin content was observed in HF fetuses (Table 2). Plasma glucose, FFA, and triglyceride levels were not significantly different in HF fetuses compared with LC fetuses on gestational day 21 (Table 2).
To investigate whether fetal islets from HF fetuses possess an altered insulin secretory capacity, the insulin secretory response of these islets to various secretagogues was studied (Fig. 2). For this purpose, the insulin secretory responses to 5.5 (basal glucose) and 16.7 mM (high glucose) glucose and 5.5 mM glucose plus either 10 mM arginine or 10 mM leucine by islets isolated from fetuses of HF and LC female rats on gestational day 21 were determined. In the presence of 5.5 mM glucose, both groups of islets secreted very low amounts of insulin, but HF fetal islets secreted significantly more insulin than LC fetal islets (0.255 ± 0.03 pmol·10 islets⁻¹·60 min⁻¹ for LC fetal islets and 0.400 ± 0.03 pmol·10 islets⁻¹·60 min⁻¹ for HF fetal islets; Fig. 2). In the presence of 16.7 mM glucose, islets from LC and HF groups demonstrated an increased response compared with their corresponding responses at 5.5 mM glucose. However, HF fetal islets secreted significantly increased amounts of insulin in the presence of 16.7 mM glucose compared with LC fetal islets. In response to 5.5 mM glucose plus either 10 mM arginine or leucine, HF fetal islets secreted significantly higher amounts of insulin compared with LC fetal islets (Fig. 2).

Characteristics of the progeny born to HF female rats. In another set of experiments, pregnant HF and LC females were allowed to deliver their pups. Pups were reared by their natural dams (continuing on their assigned diets) after adjustment of litter size to 11 pups/dam. On postnatal day 24, only the male progeny (2–3 from each dam) of the HF female rats were weaned onto either laboratory rat chow (HF/LC; average body wt 79.5 ± 2.5 g, n = 16) or high sucrose (HF/HSu; average body wt 76 ± 2.1 g, n = 16), and all female rats were removed from the experiment. The controls were the male progeny of LF female rats weaned onto either laboratory rat chow (LC/LC; average body wt 75.4 ± 3.7 g, n = 16) or HSu (LC/HSu; average body wt 72.2 ± 1.2 g, n = 16). The body weights of the HF and HF offspring weaned onto laboratory rat chow (LC) or HSu are shown in Fig. 3A. Compared with the LC/LC male progeny, the HF/LC and HF/HSu male progeny demonstrated significant increases in the body weights beginning from postnatal day 30 onward (see insert for body weights from postnatal days 24 to 40). The body weights of HF/HSu rats were similar to those of LC/HSu rats up to postnatal day 60 and then after there were significant differences in the body weights between the two groups of rats. The plasma insulin levels in the HF/LC rats weaned onto lab chow were significantly higher than those of the LC/LC rats from postnatal day 40 onward (Fig. 3B). The plasma insulin levels in the progeny weaned onto high sucrose were not determined.

The ability of the HF and LC progeny to dispose of a glucose load was determined by performing an intraperitoneal GTT in these rats on postnatal day 90. The blood glucose levels in the HF/LC progeny were significantly higher from 0 to 180 min than the LC/LC progeny (Fig. 4A). Similarly, the ability to dispose of a glucose load was impaired in the HF/HSu rats compared with both the LC/LC and LC/HSu rats (Fig. 4A). Plasma insulin levels, measured at 0, 15, and 60 min, were significantly higher in the HF/LC rats than the LC/LC rats (Fig. 4B). In LC/LC rats, the plasma insulin levels peaked at 15 min and returned to basal levels at the end of 60 min. In the case of the HF/LC rats, the plasma insulin levels continued to remain high even at the end of 60 min. The plasma insulin levels during the GTT were significantly higher in the HF/HSu progeny than in the LC/LC rats. However, the plasma insulin levels were not significantly different between the LC/HSu and HF/HSu rats (Fig. 4B).

The biochemical characteristics of the HF/LC and HF/HSu progeny on postnatal day 120, as seen in Table 3, indicated significant increases in body weight and in plasma levels of insulin, glucose, FFAs, and triglycerides compared with the values of age-matched LC/LC progeny. In the case of the HF progeny weaned onto a HSu diet, body weights and plasma

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**Table 2. Biochemical characteristics of LC and HF female rats and fetuses**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Prepregnancy (postnatal day 100)</th>
<th>Pregnancy (postnatal day 150 and gestational day 21)</th>
<th>Term Fetuses (gestational day 21)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>LC</td>
<td>HF</td>
<td>LC</td>
</tr>
<tr>
<td>Body weight, g</td>
<td>328 ± 10</td>
<td>389 ± 12*</td>
<td>502 ± 20</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>5.38 ± 0.19</td>
</tr>
<tr>
<td>Plasma insulin, pM</td>
<td>251 ± 57</td>
<td>442 ± 30*</td>
<td>250 ± 13</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>362 ± 27</td>
</tr>
<tr>
<td>Plasma glucose, mg/dl</td>
<td>117 ± 9</td>
<td>111 ± 4</td>
<td>95 ± 4</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>58 ± 3</td>
</tr>
<tr>
<td>Plasma FFA, μM</td>
<td>345 ± 86</td>
<td>375 ± 43</td>
<td>876 ± 60</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>16.9 ± 1.2</td>
</tr>
<tr>
<td>Plasma triglycerides, mM</td>
<td>1.25 ± 0.27</td>
<td>1.08 ± 0.08</td>
<td>2.19 ± 0.3</td>
</tr>
<tr>
<td>Pancreatic insulin content, μg/g tissue</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>18.4 ± 1.5</td>
</tr>
</tbody>
</table>

Results are means ± SE; n = 6–8. FFA, free fatty acids; ND, not determined. Student’s t-test was used to determine significance of the differences in the means between the groups. ∗P ≤ 0.05 HF compared with LC.
insulin levels were significantly increased without any variations in the plasma levels of glucose, FFAs, and triglycerides compared with LC/HSu rats (Table 3).

The insulin secretory responses at 10 and 60 min of islets isolated from 120-day-old male progeny of HF and LC female rats are depicted in Fig. 5. Islets isolated from LC/LC rats depicted a normal secretory pattern by secreting increasing amounts of insulin in response to 1, 5.5, and 16.7 mM glucose at both 10 and 60 min (Fig. 5). In response to 1 and 5.5 mM glucose, HF/LC rat islets secreted significantly higher amounts of insulin at 10 and 60 min than the LC/LC islets (Fig. 5). However, in the presence of high glucose (16.7 mM), the
insulin secretion response of HF/LC islets was not significantly different from the response of LC/LC islets at both 10 and 60 min (Fig. 5). HF/HSu islets secreted significantly increased amounts of insulin compared with LC/HSu islets at all three glucose concentrations at 10 min (Fig. 5). At 60 min, there were no significant changes in the insulin secretory response of HF/HSu islets to 1, 5.5, and 16.7 mM glucose compared with LC/HSu islets.

DISCUSSION

The nutritional status during critical periods of early life is an important determinant for proper development of the organism and maturation of metabolic and endocrine systems. An aberration in the quality and/or quantity of maternal nutrition during pregnancy resembling gestational diabetes in humans (19). Cerf et al. (11) demonstrated that, in dams fed a cafeteria-type diet during gestation and lactation, in both the nonpregnant and pregnant rats there were no changes in plasma glucose levels. However, in response to an intravenous glucose tolerance test, the rats fed the cafeteria-type diet demonstrated glucose intolerance during pregnancy resembling gestational diabetes in humans (19). Cerf et al. (11) demonstrated that, in dams fed a HF diet for 3 wk during gestation, there were significant increases in food intake, body weight gain, and serum insulin levels at the end of 2 wk of gestation without any change in plasma glucose concentration. In rats that developed diet-induced obesity, Levin and Govek (26) showed that, on a high-energy diet, these rats had increased body weights and

Table 3. Biochemical characteristics of the male progeny of LC and HF rats weaned onto either a standard rodent LC or a HSu diet on postnatal day 120

<table>
<thead>
<tr>
<th>Parameter</th>
<th>LC/LC</th>
<th>HF/LC</th>
<th>LC/HSu</th>
<th>HF/HSu</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight, g</td>
<td>637±23</td>
<td>849±44*</td>
<td>725±17</td>
<td>882±23†‡</td>
</tr>
<tr>
<td>Plasma insulin, pM</td>
<td>452±16</td>
<td>788±59*</td>
<td>792±75</td>
<td>1012±42‡</td>
</tr>
<tr>
<td>Plasma glucose, mg/dl</td>
<td>148±4</td>
<td>162±7*</td>
<td>170±4</td>
<td>168±5*</td>
</tr>
<tr>
<td>Plasma FFA, μM</td>
<td>268±18</td>
<td>348±14*</td>
<td>419±31</td>
<td>462±16†‡</td>
</tr>
<tr>
<td>Plasma triglycerides, mM</td>
<td>1.82±0.10</td>
<td>2.50±0.16*</td>
<td>2.35±0.15</td>
<td>2.40±0.10*</td>
</tr>
</tbody>
</table>

Values are means ± SE; n = 6–12 for each group. HSu, high sucrose; LC/LC, male progeny of LC female rats weaned on to a standard rodent laboratory chow; HF/LC, male progeny of HF female rats weaned on to a standard rodent laboratory chow; LC/HSu, male progeny of LC female rats weaned on to HSu diet; HF/HSu, male progeny of HF female rats weaned on to a HSu diet. Multiple-group comparisons by ANOVA for body weight, plasma insulin, plasma glucose, plasma FFA, and plasma triglycerides × progeny group indicated F3,39 = 22.12, P = 0.0001; F3,48 = 21.04, P = 0.0001; F3,27 = 4.36, P = 0.01; F3,28 = 15.28, P = 0.001; and F3,20 = 5.63, P = 0.006, respectively. Post hoc analysis was carried out by Student-Newman-Keuls test. *P ≤ 0.05 vs. LC/LC; †P ≤ 0.05 vs. LC/HSu; ‡P ≤ 0.05 vs. HF/LC.
plasma insulin and leptin levels at the end of 2 wk of gestation without changes in plasma glucose levels. The differences observed in the metabolic profiles of dams in those studies and the HF female rats in the present study could be due to the longer duration of HF diet feeding in our study. Additionally, variations in the amount and source of fat in the diets used in these different studies could also contribute to the observed differences. Fetal development in the hyperinsulinemic/obese HF female intrauterine environment results in fetal hyperinsulinemia without significant changes in their plasma glucose levels (Table 2). Similar to the results obtained from a mild diabetic pregnancy (2), the increase in fetal plasma insulin levels could be attributed to the increased plasma glucose levels in the HF female rat during pregnancy. Pancreatic adaptations, including increase in pancreatic insulin content and the amplified insulin secretory response of the HF fetal islets to various secretagogues, were most likely contributing factors for the observed fetal hyperinsulinemia (Table 2 and Fig. 2). Fetal pancreatic changes in response to an altered maternal intrauterine environment have been demonstrated in other animal models. In the high-carbohydrate rat model, fetuses of high-carbohydrate female rats (which were artificially reared on the high-carbohydrate milk formula for 3 wk in their immediate postnatal period) demonstrated hyperinsulinemia, increased pancreatic insulin content, and an altered insulin secretory response by fetal islets (37). Islets from fetuses of dams fed a low-protein diet demonstrated an impairment of insulin secretory capacity, reductions in islet size, insulin content, and vascularization (12, 36). A mild diabetic pregnancy in the rat was shown to induce an increase in pancreatic insulin content and an exaggerated response to a glucose stimulus by fetal islets (39). There are not many reports on fetal islet adaptations due to a HF dietary modification in the dam. Cerf et al. (11) demonstrated that feeding a HF diet to female rats throughout gestation resulted in significant decreases in β-cell volume and number and converse changes in α-cells, resulting in hyperglycemia in 1-day-old newborn rat pups without changes in serum insulin concentrations. In the present study, we did not perform morphometric analyses of fetal islets in the pancreata. However, the presence of fetal hyperinsulinemia in the HF term fetus is not consistent with the findings of Cerf et al. (11). Again, variations in the length of the HF dietary treatment and quality and quantity of fat in the diet between these two studies could be the reason for the observed differences.

Insulin is potent modulator of the central nervous system development. When occurring in elevated concentrations during critical periods of development, it can lead to malprogramming of central regulators of body weight and metabolism (31, 32). Perinatal hyperinsulinism has been attributed to be responsible for the observed obesity and diabetes-prone trait in offspring of diabetic mothers (31–33). Kozak and colleagues (23, 24) demonstrated that a HF diet during gestation and lactation affected body weight regulation in the adult progeny via alterations in the functioning of neuropeptide Y. Exposure of fetal brain to excess insulin during development in the hyperinsulinemic HF maternal environment may result in an abnormal development of the energy homeostasis circuitry, predisposing to increased body weight gain in adulthood. The observed fetal hyperinsulinism in the present study could be a contributing factor for the phenotype of the adult progeny of the HF female rats.

Chronic hyperinsulinemia and insulin resistance (as suggested by the observed glucose intolerance) may be the basis for the observed increase in body weight in the HF progeny rats. By postnatal day 120, the HF/LC rats were markedly heavier and had significantly increased plasma levels of insulin, glucose, FFAs, and triglycerides and increased insulin secretory response to basal glucose, suggesting impairments in carbohydrate as well as lipid metabolism (Table 3 and Fig. 5). The malprogramming effects observed in the HF/LC male progeny were further amplified in the HF/HSu male progeny rats due to the combined effects of fetal development in the HF mother and consumption of a HSu (energy-dense) diet in the postweaning period.

There are several reports on the consequences of a HF diet (during gestation only or both gestation and lactation) on the adult progeny. Some of the observed consequences include abnormal glucose homeostasis, reduced whole body insulin sensitivity, impaired β-cell insulin secretion and changes in the structure of pancreas (16, 38), defective mesenteric artery endothelial function (21), hypertension (22, 25), alterations in the conduit artery and renal functions (3), increased body adiposity (16, 21), deranged blood lipid profile (16, 20, 22), hyperleptinemia (38), and proatherogenic lesions (29).

Additionally, Levin and Govek (26) demonstrated that, in contrast to the progeny of diet-resistant rats on a high-energy diet, only the progeny of diet-induced obesity rats on the high-energy diet became heavy, hyperphagic with increased food intake and weight gain, and developed insulin resistance (as suggested by the observed glucose intolerance) may be the basis for the observed differences. This work was supported by National Institute of Diabetes and Digestive and Kidney Diseases Grant DK-61518.

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GRANTS

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REFERENCES


