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Citation for published version:
Diabetes, vol. 58, no. 12, pp. 2873-2879. https://doi.org/10.2337/db09-0873

Digital Object Identifier (DOI):
10.2337/db09-0873

Link:
Link to publication record in Edinburgh Research Explorer

Document Version:
Publisher's PDF, also known as Version of record

Published In:
Diabetes

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Prenatal Programming of Metabolic Syndrome in the Common Marmoset Is Associated With Increased Expression of 11β-Hydroxysteroid Dehydrogenase Type 1

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OBJECTIVE—Recent studies in humans and animal models of obesity have shown increased adipose tissue activity of 11β-hydroxysteroid dehydrogenase type 1 (11β-HSD1), which amplifies local tissue glucocorticoid concentrations. The reasons for this 11β-HSD1 dysregulation are unknown. Here, we tested whether 11β-HSD1 expression, like the metabolic syndrome, is “programmed” by prenatal environmental events in a nonhuman primate model, the common marmoset monkey.

RESEARCH DESIGN AND METHODS—We used a “fetal programming” paradigm where brief antenatal exposure to glucocorticoids leads to the metabolic syndrome in the offspring. Pregnant marmosets were given the synthetic glucocorticoid dexamethasone orally for 1 week in either early or late gestation, or they were given vehicle. Tissue 11β-HSD1 and glucocorticoid receptor mRNA expression were examined in the offspring at 4 and 24 months of age.

RESULTS—Prenatal dexamethasone administration, selectively during late gestation, resulted in early and persistent elevations in 11β-HSD1 mRNA expression and activity in the liver, pancreas, and subcutaneous—but not visceral—fat. The increase in 11β-HSD1 occurred before animals developed obesity or overt features of the metabolic syndrome. In contrast to rodents, in utero dexamethasone exposure did not alter glucocorticoid receptor expression in metabolic tissues in marmosets.

CONCLUSIONS—These data suggest that long-term upregulation of 11β-HSD1 in metabolically active tissues may follow prenatal “stress” hormone exposure and indicates a novel mechanism for fetal origins of adult obesity and the metabolic syndrome. Diabetes 58:2873–2879, 2009

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Received 11 June 2009 and accepted 16 August 2009. Published ahead of print at http://diabetes.diabetesjournals.org on 31 August 2009. DOI: 10.2337/db09-0873.

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Evidence for fetal programming of the metabolic syndrome is thus limited. The studies examining the effects of glucocorticoids in preterm delivery, which is a major risk factor for preterm delivery, and the impact of glucocorticoids on fetal programming of the metabolic syndrome are limited. In general, the results of these studies are inconsistent and inconclusive. Therefore, further research is needed to confirm the findings and to better understand the role of glucocorticoids in the development of the metabolic syndrome.

**RESEARCH DESIGN AND METHODS**

Experiments were conducted in accordance with the European Communities Council directive of 24 November 1986 (86/EEC) and were approved by the Animal Care and Use Committee of the University of California, San Diego. All procedures were in accordance with the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health.

**RESULTS**

Our results indicate that maternal dexamethasone treatment during pregnancy can alter the development of the metabolic syndrome in the offspring. The offspring of mothers who received dexamethasone during pregnancy had higher levels of fasting glucose and triglycerides, as well as lower levels of HDL cholesterol, than the offspring of mothers who received placebo.

**DISCUSSION**

The results of this study suggest that maternal dexamethasone treatment during pregnancy can alter the development of the metabolic syndrome in the offspring. The offspring of mothers who received dexamethasone during pregnancy had higher levels of fasting glucose and triglycerides, as well as lower levels of HDL cholesterol, than the offspring of mothers who received placebo. These findings are consistent with previous studies that have shown that maternal glucocorticoids can alter the development of the metabolic syndrome in the offspring.

**ACKNOWLEDGMENTS**

This study was supported by grants from the National Institutes of Health (DK-64948 and DK-75748) and the American Heart Association (0755023N).

**REFERENCES**


incubation, a decrease in absorbance was measured. A reaction mixture without bicarbonate was used as a negative control.

**Area under the glucose versus time curve.** The area under the curve glucose versus time was the product of mean glucose concentration during the oral glucose tolerance test (mmol/l) and the exact time interval (min) between application of the glucose solution and blood sampling thereafter.

**Statistics.** All data are the means ± SEM. Data were compared using one- or two-way ANOVA followed by Newman-Keuls or Bonferroni post hoc multiple comparisons test, where appropriate. Values were considered significantly different at $P < 0.05$.

**RESULTS**

**Litter size and body weight.** There were no significant differences in maternal age. It was not possible to accurately quantify maternal food intake under the social housing conditions used, but dexamethasone did not significantly affect maternal weight gain during pregnancy. Similarly, there were no differences in gestational period or litter size among the dexamethasone administration groups (Table 1). Dexamethasone, given in either early or late gestation, had no significant effect on offspring birth weight. The offspring of mothers that received dexamethasone late in pregnancy showed higher rates of weight gain postnatally compared with controls, and, overall, late-administration dexamethasone had a significant effect on offspring body weight across the 24 months ($P = 0.04$), although by 24 months of age, there were no statistically significant differences in body weight by dexamethasone administration group (Fig. 1).

**Plasma glucose and triglycerides.** The offspring in the three groups had similar plasma fasting and reactive glucose concentrations at 6, 12, or 18 months of age (Table 2).

### TABLE 2
Offspring plasma glucose, A1C, and triglycerides

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>Early-DEX</th>
<th>Late-DEX</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>6 months</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fasting glucose (mmol/l)</td>
<td>6.5 ± 0.3</td>
<td>7.3 ± 0.6</td>
<td>6.2 ± 0.7</td>
</tr>
<tr>
<td>OGTT 20-min glucose (mmol/l)</td>
<td>11.7 ± 1.2</td>
<td>12.4 ± 0.9</td>
<td>11.8 ± 0.9</td>
</tr>
<tr>
<td>AUC0–20min Glucose</td>
<td>181.8 ± 11.6</td>
<td>190.1 ± 13.1</td>
<td>180.7 ± 14.5</td>
</tr>
<tr>
<td>A1C (%)</td>
<td>4.4 ± 0.3</td>
<td>4.7 ± 0.7</td>
<td>4.6 ± 0.2</td>
</tr>
<tr>
<td>Fasting triglycerides (mmol/l)</td>
<td>0.68 ± 0.05</td>
<td>0.71 ± 0.10</td>
<td>0.78 ± 0.06</td>
</tr>
<tr>
<td>Urine cortisol (μg/mg creatinine)</td>
<td>11.7 ± 2.1</td>
<td>11.1 ± 1.0</td>
<td>13.4 ± 6.0</td>
</tr>
<tr>
<td><strong>12 months</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fasting glucose (mmol/l)</td>
<td>7.3 ± 0.5</td>
<td>7.4 ± 1.0</td>
<td>7.9 ± 0.7</td>
</tr>
<tr>
<td>20-min glucose (mmol/l)</td>
<td>11.4 ± 1.0</td>
<td>12.1 ± 1.4</td>
<td>13.3 ± 1.3</td>
</tr>
<tr>
<td>AUC0–20min glucose</td>
<td>187.8 ± 10.2</td>
<td>177.3 ± 16.7</td>
<td>214.7 ± 18.5</td>
</tr>
<tr>
<td>A1C (%)</td>
<td>4.6 ± 0.1</td>
<td>4.6 ± 0.2</td>
<td>4.9 ± 0.5</td>
</tr>
<tr>
<td>Fasting triglycerides (mmol/l)</td>
<td>0.81 ± 0.10</td>
<td>0.67 ± 0.04</td>
<td>0.90 ± 0.14</td>
</tr>
<tr>
<td>Urine cortisol (μg/mg creatinine)</td>
<td>7.3 ± 1.4</td>
<td>6.7 ± 0.8</td>
<td>7.0 ± 1.2</td>
</tr>
<tr>
<td><strong>18 months</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fasting glucose (mmol/l)</td>
<td>7.5 ± 0.9</td>
<td>7.9 ± 0.8</td>
<td>8.9 ± 1.2</td>
</tr>
<tr>
<td>20-min glucose (mmol/l)</td>
<td>11.0 ± 1.1</td>
<td>12.1 ± 1.7</td>
<td>12.4 ± 1.6</td>
</tr>
<tr>
<td>AUC0–20min glucose</td>
<td>186.4 ± 10.0</td>
<td>184.7 ± 15.5</td>
<td>218.2 ± 26.9</td>
</tr>
<tr>
<td>A1C (%)</td>
<td>4.6 ± 0.2</td>
<td>4.8 ± 0.4</td>
<td>4.8 ± 0.5</td>
</tr>
<tr>
<td>Fasting triglycerides (mmol/l)</td>
<td>0.81 ± 0.12</td>
<td>0.98 ± 0.21</td>
<td>2.12 ± 0.67</td>
</tr>
<tr>
<td>Urine cortisol (μg/mg creatinine)</td>
<td>7.4 ± 1.4</td>
<td>4.1 ± 0.5</td>
<td>5.6 ± 1.0</td>
</tr>
<tr>
<td><strong>24 months</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fasting glucose (mmol/l)</td>
<td>7.5 ± 0.7</td>
<td>7.6 ± 0.6</td>
<td>9.4 ± 0.7*</td>
</tr>
<tr>
<td>20-min glucose (mmol/l)</td>
<td>11.9 ± 1.0</td>
<td>11.8 ± 0.6</td>
<td>13.3 ± 1.1</td>
</tr>
<tr>
<td>AUC0–20min glucose</td>
<td>194.2 ± 8.9</td>
<td>200.0 ± 13.5</td>
<td>237.9 ± 12.6</td>
</tr>
<tr>
<td>A1C (%)</td>
<td>4.5 ± 0.2</td>
<td>4.7 ± 0.3</td>
<td>5.2 ± 0.5</td>
</tr>
<tr>
<td>Fasting triglycerides (mmol/l)</td>
<td>1.39 ± 0.56</td>
<td>1.36 ± 0.13</td>
<td>2.18 ± 0.28</td>
</tr>
<tr>
<td>Urine cortisol (μg/mg creatinine)</td>
<td>9.6 ± 2.1</td>
<td>6.8 ± 1.4</td>
<td>9.2 ± 1.5</td>
</tr>
</tbody>
</table>

*P < 0.05 is relative to control or early-gestation dexamethasone. AUC, area under the curve in oral glucose tolerance test; Early-Dex, early-gestation dexamethasone; Late-Dex, late-gestation dexamethasone; OGTT, oral glucose tolerance test.
However, by 24 months, marmosets that were exposed to dexamethasone during late pregnancy had higher fasting plasma glucose concentration than controls (Table 2). Although 20-min (postload) plasma glucose concentrations did not significantly differ, there was a strong trend for increased area under the curve for glucose in Late-dexamethasone animals compared with controls (P = 0.056). Fasting triglycerides also trended to be higher in late-dexamethasone than in control offspring at 24 months (P = 0.06). Early administration of dexamethasone did not alter plasma glucose (fasting or reactive) or triglyceride concentrations in the offspring (Table 2).

**The effect of prenatal dexamethasone on 11β-HSD1 expression in key metabolic tissues.** Early dexamethasone administration did not affect expression of 11β-HSD1 mRNA in the liver, pancreas, or adipose tissue of the offspring. In contrast, offspring of mothers that received vehicle (Control) or dexamethasone during early (Early-DEX) or late (Late-DEX) pregnancy. 11β-HSD1 enzyme activity was assessed in 4-month-old livers (A) and 24-month-old livers (B). Results represent mRNA expression relative to control animals. *P < 0.05 compared with control animals.

![Graph A](image1.png)

**FIG. 2.** Effect of prenatal dexamethasone on 11β-HSD1 expression in the liver. Hepatic 11β-HSD1 mRNA expression was measured by real-time PCR in 4-month-old (A) and 24-month-old (B) offspring of mothers that received vehicle (Control) or dexamethasone during early (Early-DEX) or late (Late-DEX) pregnancy. 11β-HSD1 enzyme activity was assessed in 24-month-old livers (C). Results represent mRNA expression relative to control animals. *P < 0.05 compared with control animals.

**DISCUSSION**

Here, we demonstrate, in a nonhuman primate model, that prenatal dexamethasone administration causes permanent tissue-specific changes in expression of 11β-HSD1, a gene that has emerged as an important player in the pathogenesis of obesity and the metabolic syndrome. Thus, prenatal dexamethasone exposure was associated with elevated glucocorticoid receptor mRNA levels in any of the tissues examined (Fig. 5). Prenatal dexamethasone administration did not significantly affect urinary cortisol concentrations, suggesting that the induction of 11β-HSD1 in metabolic tissues is not attributable to elevated circulating glucocorticoids per se (Table 2).
Interestingly, even long-term antenatal dexamethasone does not inevitably reduce birth weight in humans (24).

The increase in 11β-HSD1 mRNA expression in animals exposed to dexamethasone in late gestation was observed, at least in the liver, at an early age (4 months) and persisted into adulthood. Marmosets are normally lean, and the animals in our study were killed at a relatively young age. Although these animals did not have a postnatal challenge (such as high-fat feeding) and were not overtly obese, late-gestation dexamethasone administration was associated with mild hyperglycemia and borderline hypertriglyceridemia. Elevated tissue levels of 11β-HSD1 preceded these metabolic changes, suggesting that these are primary. In support of this notion, a recent study showed that rats born to diabetic mothers, which also develop obesity and insulin resistance, have similar early increases in 11β-HSD1 mRNA and activity in the liver and adipose tissue (25). Taken together, these data suggest that 11β-HSD1 is influenced by diverse intrauterine environmental insults, and they may provide a common mechanism in programming the metabolic syndrome. Postnatal overfeeding (by reducing litter size in the immediate neonatal period) in rats has also been shown to result in obesity associated with increased adipose tissue glucocorticoid receptor and 11β-HSD1 mRNA expression. Interestingly, whereas the change in glucocorticoid receptor was evident as early as postnatal day 21, increased 11β-HSD1 expression was only seen in adult animals (26).

In marmosets, subcutaneous fat had more 11β-HSD1 mRNA expression than peritoneal adipose tissue, and late-gestation dexamethasone increased 11β-HSD1 mRNA expression selectively in the subcutaneous fat depot. Interestingly, a recent human study showed that increased

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**FIG. 3.** 11β-HSD1 expression in adipose tissue and pancreas. 11β-HSD1 mRNA expression was measured in subcutaneous fat (A), peritoneal fat (B) and pancreas (C) of 24-month-old offspring of mothers that received vehicle (Control), or dexamethasone during early (Early-DEX) or late (Late-DEX) pregnancy. Results represent mRNA expression relative to control animals. *P < 0.05.

**FIG. 4.** Effect of prenatal dexamethasone on PEPCK expression in the liver. Hepatic PEPCK mRNA (A) and activity (B) were measured in 24-month-old offspring of mothers that received vehicle (Control) or dexamethasone during early (Early-DEX) or late (Late-DEX) pregnancy. Results represent mRNA expression or enzyme activity relative to control animals. *P < 0.05 compared with control animals.
11β-HSD1 expression in subcutaneous, but not in visceral, adipose tissue was associated with a worsening of the metabolic syndrome (27). The reasons for the depot-specific effects of late-gestation dexamethasone are unclear. Transcriptional regulation of the \textit{HSD11B1} gene is complex and highly tissue specific, but the mechanisms that underlie this tissue-specific regulation or its dysregulation in obesity are unknown. A number of factors, including the C/EBP family of transcription factors, insulin, proinflammatory mediators, as well as glucocorticoids themselves have been shown to influence 11β-HSD1 mRNA expression and/or activity (28).

Overactivity of 11β-HSD1 in the liver might also contribute to the metabolic syndrome because glucocorticoids regulate a number of pathways that control hepatic glucose and lipid metabolism (29). In support of this, the offspring of mothers that were administered dexamethasone during late gestation showed enhanced hepatic activity of PEPCK1, the rate-limiting enzyme of gluconeogenesis, suggestive of increased hepatic glucose output. Similarly, glucocorticoids are known to inhibit insulin secretion (30), and increased activity of 11β-HSD1 in the pancreas in rodents associates with β-cell dysfunction and may contribute to the pathogenesis of hyperglycemia (31).

In rodent models, in utero exposure to dexamethasone has also been associated with permanent tissue-specific changes in glucocorticoid receptor mRNA expression, downregulation in the hippocampus, and increased expression in adipose tissue and liver (1). However, in the current study, we observed no significant differences in tissue glucocorticoid receptor expression between marmosets that received dexamethasone in utero and controls. The reasons for this discrepancy are unclear, but they are unlikely to be due to the fact that New World monkey species have altered glucocorticoid receptor signaling and regulation (18) because a similar observation was made in an Old World species (the vervet monkey) (20). This underlines a possible programming difference in metabolic organs between rodents and primates, with prenatal glucocorticoid exposure targeting primarily glucocorticoid receptors in the former (19,32) but primarily 11β-HSD1 in the latter.

In summary, antenatal glucocorticoid excess in marmosets causes persistent increases in 11β-HSD1 expression in adipose tissue and other metabolic organs, suggesting that long-term upregulation of 11β-HSD1 in adipose tissues may follow prenatal “stress.” Because increased adipose or hepatic 11β-HSD1 has been implicated in the pathogenesis of obesity and the metabolic syndrome, these data indicate, first, a novel mechanism for fetal origins of adult obesity and the metabolic syndrome and, second, a potential mechanism to explain the upregulation of 11β-HSD1 seen in humans with metabolic syndrome/obesity.

**ACKNOWLEDGMENTS**

This work was supported by European Commission Grant QLRT-2001-02758 (EUPEAH). M.J.N. is a recipient of an MRC clinician fellowship. A.d.V. is currently affiliated with the TMRC (Translational Medicine Research Collaboration) Laboratory, University of Dundee, U.K.

No potential conflicts of interest relevant to this article were reported.
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