OBJECTIVE — The aims of the present study were to 1) evaluate autonomic function during the oral glucose tolerance test (OGTT) in pregnant women and 2) investigate whether gestational diabetes mellitus (GDM) modifies autonomic control of heart rate variability.

RESEARCH DESIGN AND METHODS — We prospectively studied 27 pregnant women (15 without GDM, 12 with GDM) during a 100-g OGTT. The maternal electrocardiogram was recorded before and 60 min after glucose ingestion, when peak glucose levels are expected. The time and frequency domains of maternal cardiac intervals were analyzed.

RESULTS — There was a significant decrease in the high-frequency (HF) band in both groups after the ingestion of glucose. The normalized low-frequency (LF) band significantly increased and the normalized HF band significantly decreased after glucose ingestion. The LF-to-HF ratio was significantly higher in the group with GDM at baseline and significantly increased in both groups after glucose ingestion. A regression analysis revealed a significant decrease in the HF band with increasing blood glucose levels.

CONCLUSIONS — Acute elevation of blood glucose levels during the OGTT caused substantial autonomic alterations, including sympathetic activation and parasympathetic withdrawal. Both arms of the autonomic system were affected during the test, thus lending support to the concept that these changes are centrally integrated. The autonomic changes were less pronounced in women with GDM compared with in normal control subjects, suggesting that chronic hyperglycemia and hyperinsulinemia may alter modulation of the autonomic nervous system.

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lucose ingestion has been shown to alter the activity of the autonomic nervous system (1–2). Cardiovascular autonomic function and baroreflex sensitivity are inversely related to blood glucose levels in healthy individuals (3–5). During pregnancy, there are significant physiological alterations in glucose absorption and metabolism that cause an increase in insulin resistance. In addition, profound hemodynamic changes take place, with an increase in cardiac output and a decrease in total peripheral resistance accompanied by an expanded blood volume (6). During the 100-g oral glucose tolerance test (OGTT), performed in pregnant women for the diagnosis of gestational diabetes mellitus (GDM), blood glucose levels are increased within a short time period. These abrupt changes in blood glucose may cause maternal and fetal metabolic and cardiovascular alterations (7–9).

Heart rate variability, modulated by diverse central and peripheral inputs, provides a quantitative marker of autonomic activity. Spectral analysis of heart rate variability is an accepted method for assessing the autonomic nervous system and permits a noninvasive evaluation of the sensitivity of the sinoatrial node to sympathetic and parasympathetic activity (10–11).

The aims of the present study were to 1) evaluate the effects of rapid changes in blood glucose levels during the 100-g OGTT in pregnant women on heart rate variability and 2) investigate whether women diagnosed with GDM react differently to the OGTT compared with euglycemic women.

RESEARCH DESIGN AND METHODS — For this study, 27 healthy, nonsmoking women with normal pregnancies between 24–28 weeks gestation were recruited. All had an abnormal 1-h 50-g glucose challenge test (≥140 mg/dl [≥7.8 mmol/l]) and were therefore referred for the 100-g OGTT. None had hypertension, thyroid disease, obesity, or a family history of diabetes. The study was approved by the Institutional Review Board in accordance with the Helsinki Declaration, and each patient signed an informed consent.

The 100-g OGTT was performed between 8:00 and 9:00 A.M. after the subjects had fasted overnight for at least 8 h. In the 3 preceding days, all women followed an unrestricted diet containing at least 150 g of carbohydrates. After giving a fasting venous blood sample, the women were asked to drink a solution of 100 g of glucose dissolved in 200 ml of water within 10 min. Blood samples were then obtained at 60, 120, and 180 min. Plasma glucose levels were determined by the glucose oxidase method (Hitachi 747). All patients were studied in the supine position. An electrocardiogram (ECG) via bipolar skin electrodes was performed for 10 min before the glucose tolerance test (phase 0) and was repeated at 60 min after glucose ingestion, when peak glucose levels were expected (phase 1). The diagnosis of GDM was established when two or more of the following venous plasma glucose levels were exceeded: fasting, 5.8...
Heart rate variability during hyperglycemia

Table 1—Baseline characteristics of the study groups

<table>
<thead>
<tr>
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<th>Without GDM</th>
<th>With GDM</th>
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<tbody>
<tr>
<td>n</td>
<td>15</td>
<td>12</td>
</tr>
<tr>
<td>Age (years)</td>
<td>30.8 ± 3.7</td>
<td>31.2 ± 4.4</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>70.7 ± 7.1</td>
<td>73 ± 8.5</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>26.7 ± 2.7</td>
<td>26.4 ± 2.2</td>
</tr>
<tr>
<td>Fasting plasma glucose (mg/dl)</td>
<td>77.9 ± 7.7*</td>
<td>89.5 ± 12.1*</td>
</tr>
<tr>
<td>50-g glucose challenge test (mg/dl)</td>
<td>159.1 ± 10.4</td>
<td>172.3 ± 31.5</td>
</tr>
</tbody>
</table>

Data are means ± SD. *P = 0.004.

mmol/l; 1 h, 10.6 mmol/l; 2 h, 9.2 mmol/l; or 3 h, 8.1 mmol/l (12).

**ECG data processing**

The ECG was digitized at 200 samples/s using a 12-bit analog-to-digital converter and stored in a binary format for subsequent analysis. Peak R wave detection was performed using a derivative/amplitude criterion algorithm. The power spectrum of the interbeat intervals was obtained by an autoregressive model, applying an order of 16. Three frequency bands were defined: the very low frequency (VLF) band (<0.03 Hz), related mainly to thermoregulation; the low-frequency (LF) band (0.03–0.15 Hz), related mainly to baroreflex control of arterial blood pressure and modulated by the parasympathetic and sympathetic arms of the autonomic nervous system; and the high-frequency (HF) band (0.15–0.40 Hz), primarily attributed to respiratory sinus arrhythmia and modulated by the parasympathetic system. The area under the respective frequency bands was integrated and expressed in absolute units. Total power (TP) and the LF-to-HF ratio, regarded as an index of sympathovagal balance, were also computed. The LF and HF bands were expressed in normalized units (LF normalized units = LF/[TP – VLF]; HF normalized units = HF/[TP – VLF]). When expressed in normalized units, LF is considered to be a quantitative index of sympathetic activity (10). Time domain measures (heart rate, mean normal-to-normal interval, the standard deviation of all RR intervals, the square root of the mean squared differences between RR intervals, and the standard deviation of successive differences between RR intervals) were also computed.

**Statistical analyses**

Statistical computations were performed using Statgraphics 5 software (Manugistics, San Carlos, CA). A two-way ANOVA was applied to test differences between patients with and without GDM at the two phases of the study (GDM × phase). Regression analyses were performed to assess the relation between blood glucose levels and the different variables studied. P < 0.05 was considered statistically significant. Results are given as means ± SD. Power calculations performed before the initiation of the study indicated that to detect true differences with 80% power at a two-sided significance level of 5% and with the different variables we included in the analyses, a sample size of 14 participants would be adequate.

**RESULTS** — The characteristics of the two study populations are given in Table 1. Women in both groups were of a similar age and had a similar weight and BMI. Their fasting plasma glucose levels differed significantly and were significantly higher in the women who were subsequently diagnosed as having GDM; however, they did not differ by the results of the 1-h 50-g glucose challenge test. Variables derived by analysis in the time and frequency domains in both groups (i.e., with and without GDM) are presented in Table 2. There were no statistically significant changes in the time domain measures between the two groups in the different phases of the study. There was a decrease in the LF and HF bands in both groups after the ingestion of glucose; however, only the change in the HF band reached statistical significance. The magnitude of this decrease was similar in both groups.

The normalized LF band significantly increased and the normalized HF band significantly decreased after glucose in

<table>
<thead>
<tr>
<th></th>
<th>Without GDM</th>
<th>With GDM</th>
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<tbody>
<tr>
<td>Phase 0</td>
<td>15</td>
<td>12</td>
</tr>
<tr>
<td>Mean heart rate</td>
<td>87.5 ± 9.8</td>
<td>91.4 ± 8.4</td>
</tr>
<tr>
<td>Mean normal-to-normal interval</td>
<td>708.7 ± 84.1</td>
<td>660.6 ± 60.1</td>
</tr>
<tr>
<td>SD of all RR intervals</td>
<td>35.0 ± 19.8</td>
<td>31.1 ± 8.0</td>
</tr>
<tr>
<td>Square root of mean squared differences between RR intervals</td>
<td>26.1 ± 29.2</td>
<td>16.2 ± 5.9</td>
</tr>
<tr>
<td>Total power</td>
<td>853.0 ± 144.4</td>
<td>898.6 ± 111.4</td>
</tr>
<tr>
<td>VLF</td>
<td>481.3 ± 165.5</td>
<td>591.2 ± 196.1</td>
</tr>
<tr>
<td>LF</td>
<td>233.4 ± 110.7</td>
<td>202.9 ± 96.2</td>
</tr>
<tr>
<td>HF</td>
<td>140.7 ± 65.8*</td>
<td>104.5 ± 57.4*</td>
</tr>
<tr>
<td>HF (normalized units)</td>
<td>60.9 ± 14.9†</td>
<td>66.8 ± 9.9†</td>
</tr>
<tr>
<td>HF (normalized units)</td>
<td>39.1 ± 14.9†</td>
<td>33.2 ± 9.9†</td>
</tr>
<tr>
<td>LF-to-HF ratio</td>
<td>1.6 ± 0.7†</td>
<td>2.8 ± 2.1†</td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>83.2 ± 20.1§</td>
<td>154.3 ± 23.6§</td>
</tr>
</tbody>
</table>

Data are means ± SD and reflect two-way ANOVA (phase × GDM). *P < 0.01 between phases, NS by diagnosis of GDM; †P < 0.05 between phases, NS by diagnosis of GDM; ‡P < 0.04 between phases, P < 0.02 by diagnosis of GDM; §P < 0.001 between phases and by diagnosis of GDM.
gestion. The two-way ANOVA revealed that these differences were significantly related to the ingestion of the glucose solution (defining the phase) but were unaffected by the presence of GDM. The LF-to-HF ratio was significantly higher in the GDM group at baseline (phase 0) compared with in the euglycemic group, and it significantly increased in both groups after glucose ingestion. It was still substantially higher in the GDM group in phase 1.

Linear regression analyses revealed a significant decrease in the HF band with increasing blood glucose levels (P < 0.02, correlation coefficient -0.317, R² = 10.1%) (Fig. 1). The equation of the fitted model is $Y = 165.6 - (0.425 \times \text{HF})$, where Y is glucose level.

**CONCLUSIONS** — The present study showed that an acute glucose load during an OGTT in pregnant women induces significant changes in the autonomic control of heart rate variability. The changes are characterized by sympathetic dominance and an attenuation of parasympathetic activity. A negative correlation between blood glucose levels and parasympathetic activity, represented by the HF band (both in absolute and normalized units), was demonstrated, and an increase in the LF-to-HF ratio after the glucose load was also observed. This was particularly striking in the non-GDM group, thus emphasizing the effect of acute hyperglycemia in normal pregnant women. However, the LF-to-HF ratio, a measure of sympathovagal balance, was significantly higher in women with GDM in the fasting state and after glucose ingestion, thereby implicating the influence of chronic hyperglycemia on the autonomic nervous system (Table 2). The decrease in spectral density was more prominent in the HF band, which is a marker of vagal activity (13). The observed changes suggest that women with GDM have an increased sympathetic activation at baseline compared with non-GDM women and that the sympathetic balance is further shifted after glucose ingestion. This supports the concept that women with GDM are more insulin resistant and have higher baseline glucose and insulin levels. It has been previously reported that impaired autonomic control of heart rate occurs early in diabetes (14) and that even modest elevations of fasting plasma glucose or insulin can impair autonomic cardiac control (4). Our study demonstrated that impaired autonomic control of heart rate variability also occurs in pregnant women who are diagnosed with GDM.

Only a small number of studies on the effect of induced hyperglycemia on the autonomic nervous system in nonpregnant healthy subjects have been published, and data for pregnant women are even more scarce (15). Paolisso and colleagues (1–2) reported that glucose ingestion alters cardiac autonomic functions in healthy, nonpregnant patients and that the LF-to-HF ratio correlates with the amount of body fat. In our study, the BMI did not significantly differ between the two groups, suggesting that the increase in the LF-to-HF ratio probably reflects acutely elevated blood glucose levels. Indeed, the LF-to-HF ratio has also been reported to increase after a 75-g OGTT in healthy older subjects (2). It appears that an acute glucose load universally modifies the modulation of heart rate variability by the autonomic nervous system.

During a 100-g OGTT, a rapid increase in blood glucose levels occurs within a relatively short time, and an increase in insulin secretion rapidly follows. In healthy humans, insulin levels rise shortly after food intake in accordance with the rise in plasma glucose, and the increase is directly proportional to the quantity of carbohydrate ingested (16–17). Acute hyperglycemia also causes systemic hemodynamic changes in healthy and type 2 diabetic subjects; this effect is independent of endogenous insulin secretion as it persists even when insulin excretion is suppressed (18–19). These changes consist of an increase in heart rate and blood pressure, suggesting a vasoconstrictive effect due to alterations in baroreflex activity. Insulin itself exerts a potent vasodilator effect that has been reported to stimulate sympathetic activity (20). These autonomic and hemodynamic alterations become even more complex in pregnancy, during which substantial cardiovascular and hemodynamic changes normally occur.

During pregnancy, an increase in insulin resistance normally occurs, and it even becomes more striking in women with GDM (21). Whether the effects of glucose on the autonomic nervous system are direct or mediated by plasma insulin has yet to be determined. Several studies support the role of insulin rather than plasma glucose in mediating the stimulatory effects on the sympathetic nervous system after a glucose load (22–24). Others hypothesize that hyperglycemia may increase the production of free radicals that mediate the hemodynamic effects (19).

In summary, acute short-term elevations of blood glucose during the OGTT in pregnant women are associated with sympathetic activation and parasympathetic withdrawal. The observation that both arms of the autonomic system are reciprocally affected supports the concept that these changes are centrally integrated. Chronic exposure to higher blood glucose and insulin levels, as are seen in pregnant women with GDM, may amplify the...
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changes observed in the autonomic control over maternal heart rate variability.

References


