Glucoregulatory Physiology in Subjects with Low-Normal, High-Normal, or Impaired Fasting Glucose

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Objective: We determined whether variations in fasting plasma glucose (FPG) within the non-diabetic range represent differences in insulin action or secretion.

Subjects and Methods: Using results of the 75-g oral glucose tolerance test, we classified 39 adults (body mass index range 20–56 kg/m²) as having normal fasting glucose (NFG, <100 mg/dl, n = 24) or impaired fasting glucose (IFG, 100–125 mg/dl, n = 15). The NFG group was subdivided into low-NFG (<90 mg/dl, n = 11) and high-NFG (90–99 mg/dl, n = 13). The 2-h oral glucose tolerance test plasma glucose value was used to assign normal or impaired glucose tolerance (IGT) status. Insulin sensitivity was assessed by hyperinsulinemic euglycemic clamp, quantitative insulin sensitivity check index, and homeostasis model assessment of insulin resistance; β-cell function was assessed by calculating the insulinogenic index, homeostasis model assessment, and the disposition index.

Results: Compared with low-NFG subjects, insulin sensitivity and glucose tolerance were lower among persons with isolated IFG and combined IFG-IGT. The hyperinsulinemic euglycemic clamp (\(\mu\)mol/kg⋅min \(^{-1}\)/pmol/liter) was 0.109 ± 0.011 in low-NFG subjects, 0.088 ± 0.009 in IFG subjects (\(P = 0.04\)), and 0.022 ± 0.003 in IFG-IGT subjects (\(P = 0.0014\)). The spread in FPG from 70–125 mg/dl was associated with a greater than 3-fold difference in insulin sensitivity. Compared with low-NFG subjects, the disposition index decreased by 32.8% in high-NFG subjects (\(P = 0.08\)), by 45.6% in subjects with isolated IFG (\(P = 0.02\)), and by 49.8% (\(P < 0.02\)) in those with combined IFG-IGT.

Conclusion: Compared with persons with low-NFG, those with IFG or combined IFG-IGT have significant alteration of glucoregulatory physiology, whereas high-NFG (pre-prediabetes) status might portend nascent glucoregulatory perturbations. (J Clin Endocrinol Metab 94: 2031–2036, 2009)

The development of type 2 diabetes requires a progressive increase in postprandial and fasting plasma glucose (FPG) levels from previously normal values, through intermediate prediabetic states, to diagnostic hyperglycemic levels. The two currently recognized prediabetic states are impaired glucose tolerance (IGT) and impaired fasting glucose (IFG); subjects with these conditions tend to progress to type 2 diabetes at an annual rate of about 10% (1–3). The definition of IGT originally proposed by the World Health Organization (WHO) has been in use for decades (4). That definition includes a normal FPG level and a 2-h plasma glucose value that lies between 140 and 199 mg/dl during a standard 75-g oral glucose tolerance test (OGTT) (4). Numerous studies based on the WHO definition have established IGT as a prediabetic state that is associated with insulin resistance and impaired insulin secretion (2, 5–7).

In contrast, the criterion for the diagnosis of IFG was first established by the American Diabetes Association (ADA) in 1997 as a FPG value of between 110 and 125 mg/dl (8). In September

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**Abbreviations:** AUC, Area under the curve; BMI, body mass index; DI, disposition index; FPG, fasting plasma glucose; GCRC, General Clinical Research Center; GDR, glucose disposal rate; HOMA, homeostasis model assessment; HOMA-B, homeostasis model assessment of β-cell function; HOMA-IR, homeostasis model assessment of insulin resistance; IFG, impaired fasting glucose; IGT, impaired glucose tolerance; ISI, insulin sensitivity index; NFG, normal fasting glucose; NGT, normal glucose tolerance; OGTT, oral glucose tolerance test; QUICKI, quantitative insulin sensitivity check index.
2003, the ADA revised the definition of normal FPG (NFG) as less than 100 mg/dl (9), which in effect revised the definition of IFG as FPG levels of between 100 and 125 mg/dl. These definitions and redefinitions of NFG and IFG have occurred without a precise physiological or pathophysiological understanding of the gradations in metabolic, glucoregulatory, or cardiovascular risks across FPG thresholds within the nondiabetic range (10). However, few studies have specifically examined glucoregulatory physiology in persons with IFG (11) or normoglycemia (12). In this report, we have used a variety of methodologies to determine whether plasma glucose thresholds for significant alteration of insulin action or secretion are discernible among individuals whose FPG values lie in the low-normal (<90 mg/dl), high-normal (90–99 mg/dl), or IFG (100–125 mg/dl) range. We thus tested the hypothesis that variations in FPG levels within the normal range represent differences in insulin sensitivity and β-cell function.

Subjects and Methods

Subjects

We studied 39 (16 male, 23 female) healthy, nondiabetic adults. The subjects had a mean (±SEM) age of 36.4 ± 1.7 yr and a body mass index (BMI) range of 20–56 kg/m². There were 28 Caucasians (72%) and 11 (28%) African-Americans. None of the subjects had a history of diabetes or current or past use of antidiabetic medications. The study subjects were taking no medication known to affect glucose tolerance or insulin sensitivity, and no subject was enrolled in an active weight loss program. The study protocol was approved by the institutional review board and all subjects gave written informed consent before participation in this study.

OGTT

All participants were screened with a standard 75-g OGTT after an overnight fast. In brief, participants were given written instructions to maintain an adequate carbohydrate intake (>100 g/d) 3 d before the test, refrain from heavy exercise and alcohol consumption for 24 h, fast overnight for about 12 h, and avoid smoking on the morning of the test. On arrival at the General Clinical Research Center (GCRC), study subjects had their vital signs recorded, followed by placement of an indwelling cannula in a forearm vein. A 75-g oral glucose solution was administered over a period of less than 10 min; blood sampling was performed immediately before and 30 min and 2 h after ingestion of glucose.

Assessment of insulin action

Using the fasting glucose and insulin data obtained during the screening OGTT, we calculated the quantitative insulin sensitivity check index (QUICKI) (13) and the homeostasis model assessment of insulin resistance (HOMA-IR) (14) as measures of insulin action.

The formulas used were QUICKI (13) = 1/log [fasting plasma insulin (microunits per milliliter)] + log [FPG (milligrams per deciliter)] and HOMA-IR (14) = [fasting plasma insulin (microunits per milliliter) × FPG (millimoles per liter)]/22.5.

A third measure of insulin action was obtained directly using the hyperinsulinemic euglycemic clamp method (15).

Hyperinsulinemic euglycemic clamp

Study subjects were admitted to the GCRC at about 0730 h after an approximately 12-h overnight fast. Shortly after arrival at the GCRC, indwelling iv lines were placed in an antecubital vein for infusion of insulin and dextrose and in a contralateral hand vein for arterialized blood sampling. Baseline blood specimens were obtained at −30 min and again at 0 min, just before commencement of continuous iv infusion (2.0 mU/kg ⋅ min) of regular human insulin. The insulin infusion was continued for 180 min while blood glucose concentration was maintained at about 100 mg/dl using a variable-rate dextrose (20%) infusion. The dextrose infusion rate was recorded every 20 min. Plasma glucose was measured by the bedside every 5–10 min, and blood specimens were obtained every 20 min for measurement of insulin and C-peptide levels. The rate of total insulin-stimulated glucose disposal (M) was calculated during the steady state between 120 and 180 min of the clamp procedure and corrected for ambient plasma insulin concentration, to obtain the insulin sensitivity index (M/I) (15). At the end of 180 min, the clamp procedure was terminated by stopping the insulin infusion; the dextrose (20%) infusion was continued and gradually tapered to maintain euglycemia while study subjects ate lunch before discharge. The hyperinsulinemic euglycemic clamps were completed within 3 wk of the OGTT date for all except two subjects, who had their clamp studies 30 and 40 d after the OGTT, respectively.

TABLE 1. Characteristics of study subjects by glycemic groups

<table>
<thead>
<tr>
<th></th>
<th>Low-NFG</th>
<th>High-NFG</th>
<th>Pure IFG</th>
<th>IFG-IGT</th>
<th>ANOVA P values</th>
</tr>
</thead>
<tbody>
<tr>
<td>n (male/female)</td>
<td>11 (4/7)</td>
<td>13 (5/8)</td>
<td>8 (4/4)</td>
<td>7 (3/4)</td>
<td></td>
</tr>
<tr>
<td>Age (yr)</td>
<td>32.6 ± 2.61</td>
<td>37.2 ± 2.23</td>
<td>38.0 ± 4.50</td>
<td>35.0 ± 4.21</td>
<td>0.13</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>31.1 ± 2.93</td>
<td>30.7 ± 2.97</td>
<td>30.5 ± 3.27</td>
<td>38.2 ± 3.4</td>
<td>0.07</td>
</tr>
<tr>
<td>FPG (mg/dl)</td>
<td>83.3 ± 1.57</td>
<td>94.7 ± 0.90</td>
<td>108 ± 2.91</td>
<td>114 ± 2.52</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>2-h PG (mg/dl)</td>
<td>101 ± 7.28</td>
<td>113 ± 7.69</td>
<td>118 ± 7.44</td>
<td>178 ± 11.2</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Fasting plasma insulin (μU/ml)</td>
<td>7.0 ± 0.60</td>
<td>9.9 ± 1.5</td>
<td>17.3 ± 4.1</td>
<td>30.6 ± 5.5</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

To convert glucose from milligrams per deciliter to millimoles per liter, multiply by 0.05551. To convert insulin from microunits per milliliter to picomoles per liter, multiply by 6.0.

a P < 0.05 vs. low-NFG.

b P < 0.05 vs. low-NFG, high-NFG, and pure IFG.

c P < 0.0001 vs. low-NFG and significantly different from each other (P < 0.05 to <0.0001).

d P < 0.0001 vs. the other three groups.

P = 0.009 to <0.0001 vs. low-NFG and high-NFG and significantly different from each other (P = 0.0015).
Assessment of insulin secretion

Insulin secretion was assessed by calculating the insulinogenic index (16) and homeostasis model assessment of β-cell function (HOMA-B) (14) from data obtained during the 75-g OGTT performed at baseline. The formulas used were HOMA-B (14) = [20 × fasting plasma insulin (microunits per milliliter)]/[FPG (millimoles per liter) − 3.5] and insulinogenic index (16) = [plasma insulin (30 min) − fasting plasma insulin (microunits per milliliter)]/[plasma glucose (30 min) − FPG (milligrams per deciliter)].

Disposition index (DI)

Under normal glucoregulatory metabolism, changes in insulin sensitivity trigger compensatory changes in the β-cell’s sensitivity to glucose. By convention, DI is measured as the product of the insulin sensitivity index (ISI) and β-cell function as measured by the acute insulin response (17). The acute insulin response is obtained from the frequently sampled iv glucose tolerance test. Because we did not perform the frequently sampled iv glucose tolerance test in the present study, we substituted insulin secretion during the first 30 min of the OGTT (ΔG0–30/ΔI0–30) as evidence of the acute insulin response (18). We thus calculated DI as (ΔG0–30/ΔI0–30) × (ISI-clamp).

Analytical methods

Plasma glucose was measured with a glucose oxidase method (Glucose Analyzer 2; Beckman Instruments, Fullerton, CA) and insulin with a specific RIA (19).

Statistical analysis

Results are expressed as mean ± SEM. Statistical analyses were run on StatView version 5.0.1 for Windows (SAS Institute, Cary, NC). Comparisons of data among subjects in the different groups were done using ANOVA. Correlations between parameters were evaluated using Spearman correlation coefficients. P < 0.05 was accepted as significant for all the data analyses.

Results

Table 1 shows the characteristics of study subjects by glycemic groups. The mean age was higher in subjects with high-normal fasting glucose compared with other groups. On the other hand, the mean BMI was similar among low-NFG, high-NFG, and pure IFG subjects and significantly higher in the combined IFG-IGT group compared with the other groups. Figure 1A shows plasma glucose and insulin levels during OGTT: the subjects with low-NFG had the lowest glucose and insulin excursions after oral glucose challenge. The incremental area under the curve (AUC) for glucose (milligrams per deciliter per minute) during the OGTT (Fig. 1A) increased progressively across the spectrum from low-NFG (AUC 7036 ± 1075), high-NFG (AUC 9722 ± 1202), and pure IFG (AUC 13,578 ± 1,543) to combined IFG-IGT (AUC 18,176 ± 2,102) (ANOVA P = 0.0035).

Insulin sensitivity

The glucose disposal rate (GDR, micromoles per kilogram per minute) at steady state during the hyperinsulinemic euglycemic clamp was 34.7 ± 4.00 in low-NFG, 37.9 ± 4.82 in high-NFG, 29.1 ± 3.56 in IFG, and 13.3 ± 1.41 in IFG-IGT subjects, respectively (ANOVA P = 0.007). The GDR was similar among subjects with low- or high-NFG but was significantly lower in IFG subjects (P = 0.045) and those with combined IFG-IGT (P = 0.0009) compared with subjects with low- or high-NFG. The ISI-clamp (micromoles per kilogram per minute/picomoles per liter), which is GDR corrected for steady-state plasma insulin concentration, was similar among low-NFG and high-NFG subjects (0.109 ± 0.011 vs. 0.117 ± 0.015) but significantly lower by about 20% in IFG subjects (0.088 ± 0.009, P = 0.04) and by about 60–80% among subjects with combined IFG-IGT (0.022 ± 0.003) (P = 0.0014 vs. low-NFG, 0.0027 vs. high-NFG, and 0.0011 vs. pure IFG) (Fig. 2A).

Estimates of insulin action calculated from HOMA-IR and QUICKI were concordant with the clamp-derived data (Table 2 and Fig. 2B). Notably, the HOMA-IR showed a significant stepwise increase in insulin resistance among subjects with low-NFG compared with those with high-NFG (P = 0.04), isolated IFG (P = 0.006), or combined IFG-IGT (P = 0.0001) (Table 2 and Fig. 2B). Similarly, the QUICKI data showed a significant stepwise increase in insulin resistance between subjects with low-NFG compared with those with high-NFG (P = 0.005), isolated IFG (P = 0.003), or combined IFG-IGT (P = 0.0001). The derived indices of insulin sensitivity (HOMA-IR and QUICKI) correlated well with the ISI-clamp (Spearman coefficient = 0.79; P = 0.0001). For the entire cohort, FPG levels were inversely correlated with ISI-clamp (Fig. 2C). The inverse relationship between fasting glucose and ISI-clamp persisted after controlling for BMI (r = −0.49; P = 0.005).
Insulin secretion

Among normoglycemic subjects, basal (fasting) plasma insulin levels were similar between the low-NFG and high-NFG groups. Fasting plasma insulin levels were higher among subjects with IFG (P = 0.009) and combined IFG-IGT (P < 0.0001) compared with normoglycemic subjects (Table 1). There was fairly good agreement between the insulogenic index and HOMA-B (r = 0.46; P = 0.004). Estimates of β-cell function derived from both the HOMA-B (Fig. 2C) and insulogenic index (Table 2) indicated no significant differences in insulin secretion among subjects with low-NFG, high-NFG, or isolated IFG. However, the subjects with combined IFG-IGT had values for HOMA-B and insulogenic index that were higher than values for subjects in the other groups.

Because insulin secretion generally increases in response to insulin resistance, changes in HOMA-B or insulogenic index may not accurately reflect β-cell function. We therefore examined the DI (the product of acute insulin secretory response to glucose and ISI) (Fig. 2D). Compared with the mean value among low-NFG subjects, the DI decreased by 32.8% in high-NFG subjects (P = 0.08), by 45.6% in subjects with isolated IFG (P < 0.02), and by 49.8% (P < 0.02) in those with combined IFG-IGT.

Discussion

Conclusions

Insulin resistance and pancreatic β-cell dysfunction are demonstrable in persons with prediabetes and type 2 diabetes (5). However, the threshold for perturbation of insulin action or secretion during transition from normoglycemia to prediabetes has not been delineated. In this report, we used multiple methodologies to assess glucose tolerance, insulin sensitivity, and β-cell function in healthy subjects with low-normal (<90 mg/dl) or high-normal (90–99 mg/dl) fasting glucose, isolated IFG, or combined IFG-IGT. The groups were fairly well matched in age and adiposity with the exception of the combined IFG-IGT subjects who were significantly more obese. We observed that persons with low-normal FPG levels exhibited lower glycemic and insulinemic excursions during OGGT than did subjects with high-normal FPG or IFG levels.

In general, we found good correlation between the formulaic indices of insulin sensitivity (HOMA-IR and QUICKI) and direct estimate from hyperinsulinemic euglycemic clamp (r = 0.79; P = 0.0001), as has been reported by others (12, 20). However, our ISI-clamp data showed similar values for insulin sensitivity in low-NFG and high-NFG subjects, whereas both the HOMA-IR and QUICKI indicated significant differences between the two glycemic groups. The reasons for the discrepancy are unclear, but it should be noted that the HOMA-IR assesses predominantly hepatic insulin resistance, whereas the clamp method estimates whole-body (predominantly skeletal muscle) glucose disposal (21). Thus, the metabolic data obtained by the two methods are related but not necessarily identical. The HOMA-IR data suggest that the high-NFG subjects had relative hepatic insulin resistance compared with subjects with low NFG. In contrast, the euglycemic clamp data indicate that skeletal muscle glucose uptake under conditions of exogenous hyperinsulinemia is similar between the low-NFG and high-NFG groups. Compared with the normoglycemic groups, subjects with IFG and those with combined IFG-IGT were significantly insulin resistant by all measures (clamp, HOMA-IR, and QUICKI).

Our findings demonstrate that insulin sensitivity is inversely related to glycermia, even within the normal fasting glucose range. Remarkably, increases in FPG levels from 70 to 125 mg/dl are associated with a greater than 3-fold decline in insulin sensitivity (Fig. 1B). When the predefined glycemic groups were considered, subjects with isolated (or pure) IFG had about 20% reduction in insulin sensitivity (ISI-clamp) compared with subjects with NFG levels. Subjects with combined IFG-IGT had the severest degree of insulin resistance by all measures used in the present study (clamp, HOMA-IR, and QUICKI). In a study of Mexican-American offspring of parents with type 2 diabetes, insulin-stimulated glucose disposal was reportedly similar in those with NGT and IFG but was reduced by 42% in subjects with IGT and by 48% in those with combined IFG-IGT (11).

With regard to β-cell function, the IFG group did not show a significant change in basal insulin secretion compared with subjects with NFG levels, whereas the subjects with combined IFG-IGT (who had ~80% decline in insulin sensitivity) had higher basal insulin levels. However, the apparent hyperinsulinemia
among the latter group was associated with significant \(\beta\)-cell dysfunction, as evidenced by about a 50% decline in the DI compared with low-NFG subjects. Thus, a normal or even increased basal insulin secretion does not exclude the presence of \(\beta\)-cell defect in insulin-resistant subjects at risk for type 2 diabetes (22). Our findings are in accord with previous reports indicating impaired acute insulin response to iv glucose in IGT subjects, a finding that predicts progression to diabetes (5, 11, 23).

It must be noted that the HOMA-B and the insulinogenic index that we used for assessing insulin secretion in the present study could be misleading as measures of \(\beta\)-cell function because they are dependent on fasting or early postchallenge plasma insulin levels. Because insulin secretion generally increases in response to insulin resistance, changes in HOMA-B or insulinogenic index may not necessarily equate to improvement or worsening of \(\beta\)-cell function. The DI is a more accurate measure of \(\beta\)-cell function that integrates insulin secretion in relation to the ambient glycermia and the peripheral insulin resistance. When we examined the DI among subjects in our study groups, we observed a downward trend among the high-NFG subjects and a significant decline among the subjects with IFG (FPG 100–125 mg/dl) be reevaluated with a standard IFG-IGT, compared with low-NFG subjects. Strictly, the DI is a measure of acute \(\beta\)-cell function; therefore, our data do not provide information regarding later-phase postchallenge (or postprandial) \(\beta\)-cell function or maximal \(\beta\)-cell secretory reserve across the study groups. Nonetheless, the present findings indicate that effective early insulin secretion decreases as fasting glucose increases within the nondiabetic range.

Our overall data are consistent with recent observations in adolescents (24, 25) and adults (26, 27) showing the presence of a \(\beta\)-cell defect in subjects with isolated IFG or IGT and profound insulin resistance in those with combined IFG-IGT. Focusing on persons with NGT, mild dysglycemia, or prediabetes is justified by the reports of significant cardiometabolic risk among such persons (7, 28). In the EPIC-Norfolk study, a 1% increase in hemoglobin A1c within the normal range was associated with increased 10-yr cardiovascular mortality (28). Our present findings suggest that persons whose FPG levels are less than 90 mg/dl are more glucose tolerant and more insulin sensitive than persons with higher fasting glucose levels, albeit within the normal range.

In conclusion, we have demonstrated a remarkable heterogeneity in insulin sensitivity among healthy subjects with NFG levels. We have also shown that insulin sensitivity is a major determinant of fasting glucose levels among healthy subjects. Extrapolating from previous prospective studies, we speculate that the states of high-NFG (pre-prediabetes), isolated IFG (prediabetes), or IFG-IGT (double prediabetes) may represent gradations in the risks for worsening dysglycemia, type 2 diabetes, and cardiovascular disease (2, 3, 7, 23–25). Our findings argue in favor of a reconsideration of the definition of normal FPG from the current target of less than 100 mg/dl (9) to a lower one of less than 90 mg/dl. However, this suggestion must be tempered by the fact that our division of normoglycemic subjects into low-normal and high-normal categories was rather arbitrary. Further studies using a statistical or an evidence-based approach for the determination of glycemic thresholds in a population-based sample would be needed to support our proposed revision.

Finally, we caution that the use of FPG measurement alone to diagnose IFG can be misleading, given the emerging understanding of the enormous metabolic differences between subjects with isolated IFG and those with combined IFG-IGT, as shown by our data and recent reports (24, 26–28). We therefore recommend that all persons with IFG (FPG 100–125 mg/dl) be reevaluated with a standard 75-g OGTT to separate out the potentially higher-risk subgroup with combined IFG-IGT. Clearly, prospective studies are needed to fully characterize the natural history and cardiometabolic implications of the various prediabetic states (25, 29, 30).

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References


10. Schrieger DL, Lorber B 2004 Lowering the cut point for impaired fasting glucose: where is the evidence? Where is the logic? Diabetes Care 27:592–601


