# Inflammation Markers and Metabolic Characteristics of Subjects With 1-h Plasma Glucose Levels

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**OBJECTIVE** — To assess the association of 1-h plasma glucose (1hPG) and inflammation with normal glucose tolerance (NGT) and pre-diabetes.

**RESEARCH DESIGN AND METHODS** — A cohort of 1,062 subjects was enrolled. After oral glucose load (oral glucose tolerance test), we compared subjects with NGT and pre-diabetes above and below the 1hPG cut point (155 mg/dl). Fibrinogen and leukocytes count (white blood cells [WBCs]) for subclinical inflammation, lipid ratios, insulin sensitivity (Matsuda index) were determined.

**RESULTS** — Patients with NGT and pre-diabetes (1hPG >155 mg/dl) showed a significant increase of inflammatory markers and lipid ratios (for all, $P < 0.05$). In age-, sex-, and BMI-adjusted analysis, 1hPG was associated with a significantly higher WBC count and fibrinogen ($P < 0.05$). Patients with elevated 1hPG showed a highly significant lower insulin sensitivity than subjects <1hPG ($P < 0.01$).

**CONCLUSIONS** — Elevated 1hPG in subjects with NGT and pre-diabetes is associated with subclinical inflammation, high lipid ratios, and insulin resistance. Therefore, 1hPG >155 mg/dl could be considered a new "marker" for cardiovascular risk.

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Pre-diabetes identifies subjects with impaired fasting glucose (IFG) and/or impaired glucose tolerance (IGT) at high risk for type 2 diabetes; moreover, it is associated with insulin resistance, subclinical inflammation, and cardiovascular diseases (CVDs) (1–4). Recently, 1-h hyperglycemia (1hPG) during an oral glucose tolerance test (OGTT) with a cut point of 155 mg/dl has been indicated as a further risk factor for type 2 diabetes (5,6) and showed early carotid atherosclerosis (7). The aim of this study is to evaluate the metabolic characteristics and inflammation markers in subjects with normal glucose tolerance (NGT) and pre-diabetes with or without 1hPG >155 mg/dl.

## RESEARCH DESIGN AND METHODS

— We examined a consecutive series of 1,062 subjects with no history of diabetes, CVD, with any malignant disease, liver or chronic kidney failure or inflammatory diseases, and with any drugs interfering with glucose or lipid metabolism. All subjects gave their written informed consent before study participation. Anthropometrical data and blood pressure were measured. After overnight fasting, a 75-g OGTT with blood samples at 0, 30, 60, 90, and 120 min was performed. Plasma glucose level, triglycerides, total cholesterol HDL cholesterol, and serum uric acid were automatically measured (Beckman Instruments, Brea, CA), as well as fibrinogen and leukocytes (white blood cell [WBC]) count as subclinical inflammation markers. Plasma insulin was determined by a standard assay (Roche Diagnostics, Mannheim, Germany). Insulin sensitivity was evaluated by the Matsuda index (8), calculated as 10,000 per square root of (fasting glucose [mg/dl] × fasting insulin [μU/ml]) × (mean glucose × mean insulin during OGTT). Lipid ratios as triglycerides to HDL cholesterol >3.5 and total cholesterol to HDL cholesterol >5 were considered as predictors of CVD risk (9).

According to American Diabetes Association criteria (10), we considered NGT and pre-diabetes categories; patients with diagnosis of type 2 diabetes were excluded. The cut point of 1hPG during OGTT >155 mg/dl was applied, subdividing all patients into four groups, below and above the 1hPG cutoff (NGT high, NGT low, pre-diabetes high, and pre-diabetes low). Diagnosis of metabolic syndrome was performed according to National Cholesterol Education Program Adult Treatment Panel III criteria (11). Statistical analysis was performed using SPSS 15.0 software. We used ANCOVA to compare differences between selected groups in means and the Bonferroni test to assess differences between selected groups. Adjustment for age and sex was made in all analyses. Statistical significance was considered with $P < 0.05$.

## RESULTS

— Of 1,062 patients studied, 507 (47.7%) had NGT and 555 (52.3%) were pre-diabetic. Among subjects with NGT, 122 (24.0%) had 1hPG >155 mg/dl during OGTT, while 433 (78.0%) of pre-diabetic patients showed elevated 1hPG. Glucose 30′ and 120′ were significantly higher in NGT high and pre-diabetic high versus NGT low and pre-diabetic low patients ($P < 0.05$), while glucose 30′, 60′, and 120′ were highly significantly elevated in subjects with 1hPG >155 mg/dl versus NGT low and pre-diabetes low ($P < 0.01$). NGT high and pre-diabetic high patients showed a significant increase ($P < 0.05$) of fibrinogen level and WBC count with respect to NGT low and pre-diabetic low subjects; all subjects with any history of CVD were excluded from the analysis (Table 1). NGT high and pre-diabetic high subjects were older, female, and had higher BMI in comparison to NGT low and pre-diabetic low patients; therefore, a logistic regression analysis adjusted for age, sex, and BMI was applied. After ad-

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**Table 1—*Demographic and clinical characteristics of patients enrolled***

| | NGT low | NGT high | Pre-diabetes low | Pre-diabetes high |
|---|---|---|---|---|
| Male/female | 136/249 | 56/66 | 52/70 | 206/227 |
| Age (years) | 37.8 ± 14.1 | 45.3 ± 13.4* | 45.7 ± 12.4† | 49.7 ± 12.5‡§ |
| BMI (kg/m$^2$) | 33.0 ± 6.6 | 34.1 ± 7.5 | 34.1 ± 6.6† | 36.2 ± 7.3‡§ |
| Waist circumference (cm) | 104.4 ± 13.9 | 107.9 ± 14.7 | 104.6 ± 13.3 | 111.2 ± 13.6‡§ |
| Glucose | | | | |
| 0′ (mg/dl) | 89.0 ± 6.3 | 92.1 ± 5.2* | 102.9 ± 8.0 | 106.4 ± 9.0‖ |
| 30′ (mg/dl) | 130.1 ± 22.3 | 157.6 ± 19.3* | 146.5 ± 23.0 | 172.9 ± 23.8‖ |
| 60′ (mg/dl) | 118.2 ± 21.1 | 174.0 ± 17.9¶ | 132.8 ± 16.2 | 194.0 ± 27.6‖ |
| 120′ (mg/dl) | 99.2 ± 19.4 | 112.9 ± 19.5* | 119.5 ± 27.2 | 146.0 ± 29.4‖ |
| Systolic blood pressure (mmHg) | 135.4 ± 19.0 | 140.9 ± 20.4* | 141.9 ± 21.0† | 146.5 ± 21.9§** |
| Diastolic blood pressure (mmHg) | 83.2 ± 11.2 | 85.8 ± 11.4 | 86.8 ± 12.5† | 89.0 ± 12.6§** |
| Uric acid (mg/dl) | 4.0 ± 1.0 | 4.5 ± 1.2* | 4.2 ± 0.9 | 4.8 ± 1.3‡§ |
| Fibrinogen (mg/dl) | 347.7 ± 85.3 | 360.8 ± 80.1* | 369.3 ± 71.5†‡††** | 379.0 ± 77.0‡§** |
| WBC count ($\times 10^9$/l) | 6.0 ± 1.2 | 6.8 ± 1.4* | 7.1 ± 1.3†‡ | 7.8 ± 1.5‡§** |
| Insulin sensitivity | 4.8 ± 2.5 | 3.2 ± 1.7‖ | 3.8 ± 1.9‡‡ | 2.7 ± 1.3‡§§ |
| Triglyceride–to–HDL cholesterol ratio | 2.6 ± 2.2 | 3.4 ± 2.7* | 3.5 ± 3.0† | 4.3 ± 3.6§** |
| Total cholesterol–to–HDL cholesterol ratio | 3.9 ± 1.4 | 4.6 ± 1.4* | 4.9 ± 1.5† | 5.3 ± 1.5‡§** |

Data are means ± SD. *$P < 0.05$ vs. NGT low; †$P < 0.05$ vs. NGT low; ‡$P < 0.05$ vs. pre-diabetes low; §$P < 0.05$ vs. NGT low; ‖$P < 0.01$ vs. pre-diabetes low; ¶$P < 0.01$ vs. NGT low; **$P < 0.05$ vs. NGT high; ††$P < 0.05$ vs. NGT high; ‡‡$P < 0.01$ vs. NGT low; §§$P < 0.01$ vs. NGT low.

justed analysis, fibrinogen concentration and WBC count remained significantly associated with sex ($P < 0.001$), age ($P < 0.001$), BMI ($P < 0.05$), and 1hPG ($P < 0.001$). Triglyceride–to–HDL cholesterol ratio was significantly increased in NGT high versus NGT low subjects, in pre-diabetic low versus NGT low individuals, in pre-diabetic high versus NGT low subjects, and between pre-diabetic high and NGT high patients (for all, $P < 0.05$). Higher significant levels of total cholesterol–to–HDL cholesterol ratio were found in NGT high versus NGT low subjects, in pre-diabetic low versus NGT low individuals, in pre-diabetic high versus NGT high patients, and in NGT low and NGT high subjects (for all, $P < 0.05$). Significant increased concentrations of uric acid were observed in NGT high than NGT low subjects ($P < 0.05$), in pre-diabetic high versus pre-diabetic low patients, and NGT low and NGT high patients (for all, $P < 0.05$). A highly significant lower insulin sensitivity was found between pre-diabetic high versus pre-diabetic low and between NGT high versus NGT low subjects ($P < 0.01$). Overall metabolic syndrome prevalence was 43.5%. Considering those subjects with 1hPG >155 mg/dl, 100% fulfilled metabolic syndrome criteria, but 31.0% of patients without metabolic syndrome diagnosis revealed 1hPG >155 mg/dl.

**CONCLUSIONS** — Pre-diabetes is associated with a high risk for type 2 diabetes, subclinical inflammation, early atherosclerosis, and CVD. Moreover, it was shown that subjects with NGT with 1hPG >155 mg/dl had a fivefold type 2 diabetes risk than subjects with NGT with 1hPG below the cutoff of 155 mg/dl (5). In this study, NGT high subjects showed increased WBC count and fibrinogen levels, signs of subclinical inflammation, as patients with pre-diabetes and a significant worsening of lipid profile than NGT low patients. The mechanism that links elevated 1hPG to subclinical inflammation is probably due to hyperglycemia that acutely increases circulating cytokine concentrations by oxidative mechanisms, and this effect is more pronounced in patients with impaired glucose regulation (3).

Subjects with 1hPG >155 mg/dl showed significantly lower insulin sensitivity (Matsuda index). As previously observed, metabolic syndrome is strongly associated with decreased insulin sensitivity (12); therefore, the link observed between elevated 1hPG and insulin resistance could be explained by the high prevalence of metabolic syndrome in patients with elevated 1hPG.

We decided to measure inflammation levels by fibrinogen and WBC count without considering the measure of C-reactive protein. It is important to underline that the measurement of C-reactive protein (CRP) concentration requires a specific high-sensitivity assay method that is not available in all laboratories; moreover, several data have shown that elevated levels of CRP were associated to increased CVD, but recently, analysis of genetic polymorphisms in the CRP gene in Caucasian subjects showed that these polymorphisms were not associated with an increased risk of ischemic vascular disease (13), increasing the doubts about the reliability of such a measurement in clinical practice. A high WBC count is associated with increased CVD-related morbidity and mortality in several populations and clinical settings; it may turn out to be a less expensive and more readily available risk CVD marker than other currently available risk factors (14,15).

In conclusion, our data show the relevance of 1hPG >155 mg/dl as an important cutoff related to subclinical inflammation, lipid disorders, and insulin resistance; therefore, this threshold could be seriously taken into consideration to identify subjects at high CVD risk.

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**References**

1. Nathan DM, Davidson MB, DeFronzo RA, Heine RJ, Henry RR, Pratley R, Zinman B, the American Diabetes Association. Impaired fasting glucose and impaired glucose tolerance: implications for care. Diabetes Care 2007;30:753–759

2. Haffner SM. Insulin resistance, inflammation, and the prediabetic state. Am J Cardiol 2003;92:18J–26J
3. Esposito K, Nappo F, Marfella R, Giugliano G, Giugliano F, Ciotola M, Quagliaro L, Ceriello A, Giugliano D. Inflammatory cytokine concentrations are acutely increased by hyperglycemia in humans: role of oxidative stress. Circulation 2002;106:2067–2072
4. Ohshita K, Yamane K, Hanafusa M, Mori H, Mito K, Okubo M, Hara H, Kohno N. Elevated white blood cell count in subjects with impaired glucose tolerance. Diabetes Care 2004;27:491–496
5. Abdul-Ghani MA, Abdul-Ghani T, Ali N, Defronzo RA. One-hour plasma glucose concentration and the metabolic syndrome identify subjects at high risk for future type 2 diabetes. Diabetes Care 2008;31:1650–1655
6. Abdul-Ghani MA, Lyssenko V, Tuomi T, DeFronzo RA, Groop L. Fasting versus postload plasma glucose concentration and the risk for future type 2 diabetes: results from the Botnia Study. Diabetes Care 2009;32:281–286
7. Succurro E, Marini MA, Arturi F, Grembiale A, Lugarà M, Andreozzi F, Sciacqua A, Lauro R, Hribal ML, Perticone F, Sesti G. Elevated one-hour post-load plasma glucose levels identifies subjects with normal glucose tolerance but early carotid atherosclerosis. Atherosclerosis 2009;207:245–249
8. Matsuda M, DeFronzo RA. Insulin sensitivity indices obtained from oral glucose tolerance testing. Diabetes Care 1999;22:1462–1470
9. Kannel WB, Vasan RS, Keyes MJ, Sullivan LM, Robins SJ. Usefulness of the triglyceride-high-density lipoprotein versus the cholesterol-high-density lipoprotein ratio for predicting insulin resistance and cardiometabolic risk (from the Framingham Offspring Cohort). Am J Cardiol 2008;101:497–501
10. Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Follow-up report on the diagnosis of diabetes mellitus. Diabetes Care 2003;26:3160–3167
11. Grundy SM, Cleeman JI, Daniels SR, Donato KA, Eckel RH, Franklin BA, Gordon DJ, Krauss RM, Savage PJ, Smith SC Jr, Spertus JA, Costa F, the American Heart Association, the National Heart, Lung, and Blood Institute. Diagnosis and management of the metabolic syndrome: an American Heart Association/National Heart, Lung, and Blood Institute scientific statement. Circulation 2005;112:2735–2752
12. Haffner SM, D'Agostino R Jr, Festa A, Bergman RN, Mykkänen L, Karter A, Saad MF, Wagenknecht LE. Low insulin sensitivity (S(i) = 0) in diabetic and nondiabetic subjects in the Insulin Resistance Atherosclerosis Study: is it associated with components of the metabolic syndrome and nontraditional risk factors? Diabetes Care 2003;26:2796–2803
13. Zacho J, Tybjaerg-Hansen A, Jensen JS, Grande P, Sillesen H, Nordestgaard BG. Genetically elevated C-reactive protein and ischemic vascular disease. N Engl J Med 2008;359:1897–1908
14. Madjid M, Awan I, Willerson JT, Casscells SW. Leukocyte count and coronary heart disease: implications for risk assessment. J Am Coll Cardiol 2004;44:1945–1956
15. Ikonomidis I, Stamatelopoulos K, Lekakis J, Vamvakou GD, Kremastinos DT. Inflammatory and non-invasive vascular markers: the multimarker approach for risk stratification in coronary artery disease. Atherosclerosis 2008;199:3–11