IGF-I Bioactivity in an Elderly Population

Relation to Insulin Sensitivity, Insulin Levels, and the Metabolic Syndrome

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OBJECTIVE—There is a complex relationship between IGF-I, IGF binding proteins, growth hormone, and insulin. The IGF-I kinase receptor activation assay (KIRA) is a novel method for measuring IGF-I bioactivity in human serum. We speculated that determination of IGF-I bioactivity might broaden our understanding of the IGF-I system in subjects with the metabolic syndrome. The purpose of our study was to investigate whether IGF-I bioactivity was related to insulin sensitivity and the metabolic syndrome.

RESEARCH DESIGN AND METHODS—We conducted a cross-sectional study embedded in a random sample (1,036 elderly subjects) of a prospective population-based cohort study. IGF-I bioactivity was determined by the IGF-I KIRA. Categories of glucose (in)tolerance were defined by the 2003 American Diabetes Association criteria. Insulin sensitivity was assessed by homeostasis model assessment. The Adult Treatment Panel III definition of the metabolic syndrome was used.

RESULTS—In subjects with normal fasting glucose and impaired fasting glucose, IGF-I bioactivity progressively increased with increasing insulin resistance, peaked at fasting glucose levels just below 7.0 mmol/l, and dropped at higher glucose levels. Mean IGF-I bioactivity peaked when three criteria of the metabolic syndrome were present and then declined significantly when five criteria of the metabolic syndrome were present.

CONCLUSIONS—We observed that IGF-I bioactivity was related to insulin sensitivity, insulin levels, and the metabolic syndrome. Our study suggests that there exists an inverse U-shaped relationship between IGF-I bioactivity and number of components of the metabolic syndrome. This observation contrasts with previous results reporting an inverse relationship between total IGF-I and components of the metabolic syndrome.
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A complex relationship exists between IGF-I, IGF binding proteins (IGFBPs), growth hormone (GH), and insulin. GH exerts potent effects on intermediary metabolism by antagonizing insulin action (1). IGF-I directly inhibits insulin secretion and increases insulin sensitivity (2). Insulin appears to be necessary for normal liver GH responsiveness, probably by maintaining liver GH receptor levels (3). However, chronic hyperinsulinemia reduces GH receptor expression and signaling in the liver (4,5). The majority of circulating IGF-I is bound to six IGFBPs that are important in determining IGF-I availability and activity (6).

Recently, the IGF-I–specific kinase receptor activation assay (KIRA) was developed to determine circulating IGF bioactivity (7,8). This bioassay determines IGF-I bioactivity by measuring intracellular receptor auto-phosphorylation upon IGF-I binding. Unlike IGF-I immunoassays, the IGF-I KIRA takes into account the modifying effects of IGFBPs on the interaction between IGF-I and the IGF-I receptor (IGF-IR) (9). The aim of the present study was to investigate whether IGF-I bioactivity was related to insulin sensitivity, insulin levels, and the metabolic syndrome in an elderly population-based cohort.

RESEARCH DESIGN AND METHODS

The Rotterdam Study is a prospective population-based cohort study investigating incidence and risk factors of cardiovascular diseases in elderly people (10). The ethics committee of Erasmus MC approved the study, and written informed consent was obtained from all participants.

Fasting blood samples were obtained after an overnight fast in participants without known diabetes. In subjects with diabetes, blood was drawn prior to the intake of antidiabetic agents. Fasting plasma glucose (FPG) was determined using the hexokinase method (Boehringer Mannheim, Mannheim, Germany). American Diabetes Association 2003 criteria were used to classify glucose tolerance as normal fasting glucose (NFG), FPG <5.6 mmol/l; impaired fasting glucose (IFG), FPG 5.6–6.9 mmol/l; and diabetes, FPG ≥ 7 mmol/l (11). Serum total and HDL cholesterol were determined using an automatic enzymatic procedure (Boehringer Mannheim). Serum triglycerides were analyzed by Triglyceride GPO-PAP (Roche Diagnostics, Mannheim, Germany). Insulin was measured with an electrochemiluminescence immunoassay (Roche Diagnostics; intra- and interassay coefficient of variation [CV] $<3.7\%$, with cross-reactivity of 20% with des-31, 32-proinsulin, per the manufacturer). Sitting blood pressure (mean of two readings) was measured using a random-zero sphygmomanometer.

IGF-I bioactivity was measured using an IGF-I kinase receptor activation assay (KIRA) (intra- and interassay CVs of 5.2 and 12.2%, respectively; cross-reactivity of 15% for IGF-II) (8,12). IGF-I bioactivity was measured in 1,050 blood samples randomly selected from 3,792 individuals who participated in the survey. Fourteen subjects were excluded from analysis because their measurements of IGF-I bioactivity repeatedly did not pass defined acceptance criteria (intra-assay CV $<10\%$). In 904 of 1,036 subjects (87%) all parameters of the metabolic syndrome were available; 132 subjects had one or

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more missing values and were therefore excluded from statistical analyses concerning the metabolic syndrome.

The updated homeostasis model assessment (HOMA-2) was used to assess insulin sensitivity (%S), insulin resistance (HOMA-IR), which is the reciprocal of %S, and β-cell function from pairs of fasting glucose and insulin levels (13). The Adult Treatment Panel III (ATP-III) criteria were used to diagnose the metabolic syndrome (14).

Statistical analysis. Pearson's correlation coefficients were calculated to assess (age-adjusted) associations between variables. Univariate ANCOVA were used to compare variables stratified by deciles of HOMA-IR, adjusted for age, sex, BMI, and antidiabetic therapy when appropriate. In 904 of 1,036 subjects (87%) all parameters of the metabolic syndrome were available; 132 subjects had one or more missing values and were therefore excluded from statistical analyses concerning the metabolic syndrome.

A two-sided probability of $P < 0.05$ was considered statistically significant. All analyses were performed with SPSS for Windows version 15 (SPSS, Chicago, IL).

RESULTS

The mean age of the study population of 1,036 subjects (451 male) was 72.4 years (95% CI 62.8–86.3), and the mean BMI was 26.9 kg/m^2 (21.5–33.9). Based on 2003 American Diabetes Association criteria, 697 subjects (69.7%) had NFG, 165 subjects (16.3%) had IFG, and 153 subjects (15.1%) had diabetes. A total of 62 subjects (6%) had known diabetes, of which 19 (1.8%) were treated with diet only and 39 (3.8%) were treated with oral antidiabetic agents.

Mean ± SE IGF-I bioactivity was significantly lower in men (168.9 ± 2.5 pmol/l) than in women (187.0 ± 3.1 pmol/l, $P < 0.001$ after adjustment for age and BMI). There was no association between IGF-I bioactivity and fasting insulin ($r = 0.049$, $P = 0.206$). However, when analysis was confined to subjects with NFG, IGF-I bioactivity was significantly and positively related to fasting insulin ($r = 0.096$, age-adjusted, $P = 0.036$). Mean ± SE IGF-I bioactivity did not differ between diabetic subjects with (169 ± 8.7 pmol/l) and without (179 ± 1.8 pmol/l, $P = 0.47$) oral antidiabetic agents.

After adjustment for age, overall IGF-I bioactivity was inversely related to insulin sensitivity ($r = -0.113$, $P = 0.003$), while the relationship between IGF-I bioactivity and β-cell function was borderline significant ($r = 0.065$, $P = 0.088$). When subjects were stratified to deciles of insulin resistance (HOMA-IR), mean FPG and insulin levels progressively increased with increasing insulin resistance. For mean IGF-I bioactivity there was a progressive increase with increasing insulin resistance up to and including the 9th decile (P for trend = 0.003) (Fig. 1). However, when mean FPG levels met diagnostic criteria for diabetes in the 10th decile, IGF-I bioactivity dropped significantly compared with the 9th decile ($P = 0.005$) (Fig. 1). When analyses were repeated after excluding diabetic subjects on medical therapy, the outcome was unaffected—IGF-I bioactivity increased significantly up to and including the 9th decile of HOMA-IR (P for trend = 0.006) and decreased significantly in the 10th decile ($P = 0.010$ for the 9th vs. 10th decile).

Mean HOMA-IR increased significantly per increasing number of components of the metabolic syndrome (P for trend < 0.001, adjusted for age and sex). As expected, mean FPG levels also increased progressively with an increasing number of components of the metabolic syndrome. Mean fasting insulin levels were significantly higher in subjects with five components of the metabolic syndrome than in subjects with four or fewer components of the metabolic syndrome. Up to a point, IGF-I bioactivity was also directly related to number of components of the metabolic syndrome, peaking when three components were present and the diagnosis of the metabolic syndrome could be made. However, in subjects with more than three components of the metabolic syndrome, IGF-I bioactivity decreased significantly (mean IGF-I bioactivity in subjects with three vs. five components $P = 0.04$, adjusted for age and sex) (Fig. 2).

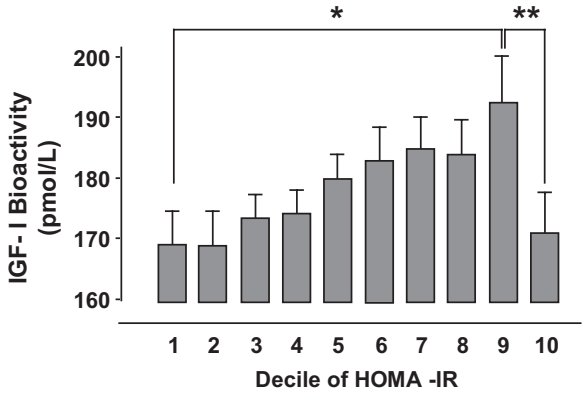

FIG. 1. Mean ± SEM IGF-I bioactivity stratified per decile of HOMA-IR. Statistical differences between deciles were calculated after adjustment for age, BMI, and sex. P for trend < 0.001, **$P = 0.005$.

DISCUSSION

The main finding of our study is that circulating IGF-I bioactivity progressively increases with increasing severity of insulin resistance and hyperinsulinemia. At FPG levels just below 7.0 mmol/l, mean IGF-I bioactivity reached a plateau. However, in subjects with diabetes, mean IGF-I bioactivity was significantly lower than in subjects with NFG and IFG.

Strong evidence exists that insulin is essential for GH stimulation of hepatic IGF-I production (3). Insulin deficiency results in a decreased liver GH receptor expression, which can be reversed by insulin administration (3,15). On the other hand, patients with type 2 diabetes often exhibit reduced circulating total IGF-I levels (16). One explanation is that chronic hyperinsulinemia induces GH receptor resistance (4). Under experimental conditions, hyperinsulinemia reduces not only GH receptor expression but also GH signaling both at receptor and postreceptor levels (4,5). Although the direction of cause and effect may be difficult to assess in a cross-sectional study, this opens the possibility that the observed decline in IGF-I bioactivity in

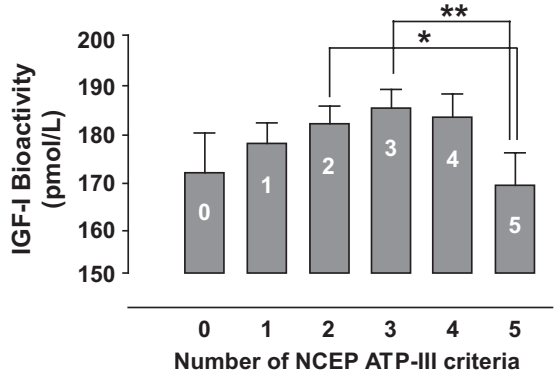

FIG. 2. Relationship between the number of components of the metabolic syndrome and mean ± SEM IGF-I bioactivity. *$P = 0.03$, **$P = 0.04$.

diabetic subjects was related to hyperinsulinemia-induced hepatic GH resistance.

Several epidemiological studies have linked low circulating total IGF-I, particularly when coupled with low levels of the potentially inhibitory IGFBP-1, to an increased risk of the metabolic syndrome (17–21). An increased ratio of total IGF-I to IGFBP-1 has been proposed as a surrogate marker of increased IGF-I bioactivity (22). This is probably a compensatory mechanism to maintain normal glucose tolerance, since raised IGF-I bioactivity improves insulin sensitivity (23). High IGF-I bioactivity may suppresses GH secretion as part of a negative feedback system, thereby inducing lower total IGF-I levels (22). This may explain why low total IGF-I levels have been previously reported to be associated with the metabolic syndrome (20).

Our study demonstrates that IGF-I bioactivity declines during progression of the metabolic syndrome. Because individuals with the metabolic syndrome are chronically exposed to high circulating insulin levels, the increase in IGF-I bioactivity in subjects with one to three components of the metabolic syndrome may be due to an insulin-mediated suppression of IGFBP-1 levels (24). However, in subjects with more than three components of the metabolic syndrome, IGF-I bioactivity significantly declined. The lower IGF-I bioactivity in these latter individuals may be a direct consequence of the development of hepatic insulin resistance (manifested by a relative increase of IGFBP-1) and hyperinsulinemia-induced GH resistance.

A limitation of our study is that no comparisons were made between circulating IGF-I bioactivity and total IGF-I measurements. However, we previously found a weak correlation between circulating total IGF-I levels and IGF-I bioactivity, suggesting that the IGF-I KIRA produces information about the IGF-I system that essentially differs from that obtained by IGF-I immunoassays (12).

Another shortcoming of this study is that we did not perform oral glucose tolerance tests. Therefore, it is not clear how many subjects had IGT or even diabetes that was not assessed by fasting glucose alone. In addition, HOMA-IR does not always reflect insulin sensitivity accurately in subjects with differing β-cell function.

In conclusion, our study suggests that an individual's insulin sensitivity and circulating insulin levels directly modulate IGF-I bioactivity. When insulin resistance becomes apparent, but glucose levels are still within the normal range, there is an initial rise in circulating IGF-I bioactivity. However, when glucose tolerance rises to the point where IFG occurs, IGF-I bioactivity reaches a plateau. Finally, when blood glucose levels have risen into overt type 2 diabetes, circulating IGF-I bioactivity progressively declines. IGF-I bioactivity seems also to be related to the metabolic syndrome. Our study suggests that there exists an inverse U-shaped relationship between IGF-I bioactivity and number of components of the metabolic syndrome.

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

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