# Plasma Adiponectin Is an Independent Predictor of Type 2 Diabetes in Asian Indians

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**OBJECTIVE** — Adiponectin, secreted by fat cells, has regulatory functions on energy metabolism. Its low levels are predictive of future development of diabetes. Because no studies on the regulatory role of adiponectin in glucose homeostasis in Asian Indians exist, this analysis was performed to determine the prospective association of adiponectin and diabetes in subjects with impaired glucose tolerance (IGT).

**RESEARCH DESIGN AND METHODS** — Baseline values of plasma adiponectin, results of anthropometry, fasting and 2-h plasma glucose levels, HbA$_{1c}$, results of lipid profile, and insulin resistance were analyzed in 91 subjects with IGT (53 men and 38 women) in a primary prevention study. Reassessment of glucose tolerance was performed during 1-year review. The predictive nature of adiponectin for development of diabetes was assessed using univariate and multiple logistic regression analyses. A control group comprising healthy, normoglycemic individuals was used for comparison.

**RESULTS** — At follow-up, diabetes had developed in 25 of the 91 study subjects. The mean baseline adiponectin level was lower in the diabetic subjects than in the nondiabetic subjects (11.3 ± 5.5 vs. 16.7 ± 7.6 μg/ml, $P = 0.0017$). Low adiponectin level was a strong predictor of future development of diabetes, and HbA$_{1c}$ also showed a positive predictive association. Women had higher adiponectin levels (16.4 ± 6.1 μg/ml) than men (13.9 ± 7.6 μg/ml) ($P = 0.035$).

**CONCLUSIONS** — In Asian Indians, low plasma adiponectin level was predictive of future development of diabetes.

*Diabetes Care* 26:3226–3229, 2003

**A**diponectin is a hormone produced solely by adipocytes and is a regulator of glucose and energy homeostasis (1,2). Low plasma adiponectin concentration is associated with a decrease in whole-body insulin sensitivity in humans (3–5) and has been shown to be predictive of future development of diabetes in a few studies (6–8).

Asian Indians have a high risk of diabetes and have an obesity phenotype characterized by lean BMI, central obesity, and high body fat percentage. In addition, Asian Indians also have high degree of insulin resistance (9,10). The role of adiponectin in regulating the complex mechanisms of insulin action and glucose homeostasis has not been studied in this population.

Subjects with persistent impaired glucose tolerance (IGT) on two oral glucose tolerance tests (OGTTs) were identified in a population screening to prospectively study the feasibility of primary prevention of type 2 diabetes and to assess the efficacy of lifestyle modification and treatment with metformin as preventive strategies (11). This cohort of urban Asian Indians are being followed regularly to look for the end points, viz., occurrence of diabetes or cardiovascular diseases. Glycemic status had been reassessed after 1 year by repeat OGTT. The baseline data of the subjects who had nonpharmacological intervention and had undergone review OGTT at 1 year were used in this analysis. The aims of study were 1) to determine the plasma adiponectin concentration in healthy Asian Indians and 2) to determine whether a prospective association existed between plasma adiponectin and type 2 diabetes in this population.

## RESEARCH DESIGN AND METHODS — The study group was

selected from a cohort of subjects with IGT being followed for 3 years in the Indian Diabetes Prevention Program (IDPP), which studies the effect of intervention measures to prevent deterioration of glucose intolerance (11). A total of 531 subjects with IGT were randomized to four groups: group 1 received standard care, group 2 was instructed in a diet modification and exercise program, group 3 received metformin, and group 4 received advice on lifestyle modification in addition to metformin. The study protocol was approved by the ethical committee of the institution, and written informed consent was obtained from each participant. All participants are being followed regularly with annual OGTTs. The 91 subjects (53 men and 38 women) selected for the study were from groups 1 and 2 only, i.e., they did not have any pharmacologic intervention. Glucose tolerance status was reassessed by repeat OGTT at 1 year, based on the World

**Table 1—***Baseline anthropometric and biochemical parameters in the group that developed diabetes in comparison with the nondiabetic group*

| Parameters | NGT + IGT group | Diabetic group |
|---|---|---|
| $n$ | 66 | 25 |
| Age (years) | 45.1 ± 5.4 | 45.5 ± 5.4 |
| BMI (kg/m$^2$) | 26.1 ± 2.7 | 26.8 ± 5.1 |
| Waist (cm) | | |
| Men | 89.1 ± 7.1 | 92.8 ± 8.9* |
| Women | 86.3 ± 7.9 | 89.2 ± 9.5 |
| Fat (%) | | |
| Men | 30.6 ± 3.9 | 31.8 ± 5.6 |
| Women | 43.7 ± 6.0 | 43.2 ± 8.7 |
| Plasma glucose (mmol/l) | | |
| Fasting | 5.5 ± 0.6 | 5.4 ± 0.9 |
| 2 h | 8.5 ± 0.7 | 8.7 ± 0.8 |
| HbA$_{1c}$ (%) | 6.1 ± 0.54 | 6.33 ± 0.54 |
| Cholesterol (mmol/l) | 5.2 ± 0.9 | 5.0 ± 0.5 |
| Triglycerides (mmol/l) | 1.9 ± 0.9 | 2.0 ± 1.3 |
| HDL cholesterol (mmol/l) | 1.1 ± 0.2 | 1.0 ± 0.2 |
| Homeostasis model assessment of insulin resistance | 5.0 ± 2.1 | 5.45 ± 3.1 |
| Adiponectin (μg/ml) | 16.7 ± 7.6 | 11.3 ± 5.5† |

Data are means ± SD. *$P = 0.04$; †$P = 0.002$.

Health Organization (WHO) criteria (12). During the repeat OGTT at 1 year, 34 subjects had normal glucose tolerance (NGT), 32 subjects had IGT, and 25 had diabetes. Hitherto, subjects are referred to as the NGT, IGT, or diabetic subgroups. The baseline parameters in the subgroups were analyzed. The mean age was 45.2 ± 5.3 years, and mean BMI was 26.3 ± 3.5 kg/m$^2$. A group of age- and BMI-matched, apparently healthy subjects having NGT on OGTT were selected as the control group ($n = 50$; 20 men and 30 women; mean age 45.7 ± 11.3 years and mean BMI 24.5 ± 5.4 kg/m$^2$).

All study subjects had baseline records of age, BMI, waist circumference, and percentage of body fat (bioimpedance method). Measurement of HbA$_{1c}$ (immunoturbidimetric method), fasting and 2-h plasma glucose levels, and plasma lipid levels was performed using standard enzymatic procedures (9). In the total group of 91 subjects with IGT, fasting plasma insulin concentrations were measured by radioimmunoassay. Reagent kits supplied by Diasorin (Saluggia, Italy) were used (assay sensitivity <4 μg/ml at 95% CI). Intra-assay and interassay coefficients of variation were <10%. Insulin resistance was calculated using homeostasis model assessment (HOMA) (13). Baseline fasting plasma adiponectin

levels were measured in the control and IGT subjects by radioimmunoassay using reagent kits provided by Linco Research (St. Louis, MO). This assay uses $^{125}$I-labeled adiponectin and an anti-adiponectin rabbit antiserum to measure adiponectin concentrations by the double-antibody/polyethylene glycol (PEG) technique. Standards over the range of 1–200 ng/ml were prepared using recombinant human adiponectin. All plasma samples were diluted 1:200, yielding an effective range of 0.2–40 μg/ml. The intra-assay and interassay coefficients of variation at adiponectin concentrations in the range of 3–15 μg/ml are 2.0–6.8 and 7.1–9.6%, respectively.

EDTA blood samples were stored in ice, and plasma aliquots were separated within 3 h and stored frozen at –70°C until the assay had been completed. Repeated freezing and thawing was avoided.

**Statistical analysis**
The values are reported as means and SDs. Unpaired Student's $t$ test was used for group comparisons. $P < 0.05$ was considered significant. Adiponectin values in all subjects with IGT ($n = 91$) were divided into tertiles. Percentage distribution of adiponectin in the NGT, IGT, and diabetic subgroups in the tertiles was calculated.

$\chi^2$ analysis was performed to assess

the differences in proportions between groups. Multiple logistic regression analysis with forward stepwise addition was used to determine the association of variables, including adiponectin level, with future development of diabetes. Data were analyzed using the SPSS 4.0.1 statistical package (SPSS, Chicago, IL).

**RESULTS** — Plasma adiponectin levels were similar in the control subjects (14.9 ± 5.9 μg/ml) and those with IGT (15.2 ± 7.5 μg/ml). In the control group, women had higher adiponectin levels (16 ± 4.8 μg/ml) than men (13.4 ± 7.1 μg/ml), but the difference did not reach statistical significance. Because the values were similar in the control and IGT groups, sex difference in adiponectin was assessed in the total group. Women had higher mean values (16.4 ± 6.1 μg/ml; $n = 68$) than men (13.9 ± 7.6 μg/ml; $n = 73$) ($P = 0.035$).

Comparison of the baseline variables in the diabetic and nondiabetic (NGT + IGT) subgroups is shown in Table 1. Because adiponectin levels were similar in the NGT and IGT subgroups (16.9 ± 8.2 and 16.6 ± 7.1 μg/ml, respectively), these subgroups were combined and shown as the nondiabetic group. All parameters were similar in nondiabetic and diabetic groups, except for waist circumference. Diabetic men had higher waist circumference than nondiabetic men. Adiponectin levels were significantly lower in subjects in whom diabetes developed (11.3 ± 5.5 vs. 16.7 ± 7.6 μg/ml, $P = 0.002$).

There was no significant sex difference in the rate of conversion to diabetes. The percentage of men in the prospective diabetic and nondiabetic groups was 64 and 56%, respectively ($\chi^2 = 0.2$, $P = 0.65$). The percentage distribution of the diabetic and nondiabetic groups in tertiles of adiponectin is shown in Fig. 1. The tertile values of adiponectin (μg/ml) were as follows: tertile 1 = 3.0–9.9; tertile 2 = 10.0–18.7; tertile 3 = 18.8–43.7.

A higher percentage of prospective diabetic subjects were in tertile 1 (56%) compared with the nondiabetic subjects (24.2%) ($\lambda^2 = 6.9$, $P = 0.009$).

In the multiple logistic regression analysis (Table 2), the factors predictive of future development of diabetes were baseline HbA$_{1c}$ and low plasma adiponectin concentration.
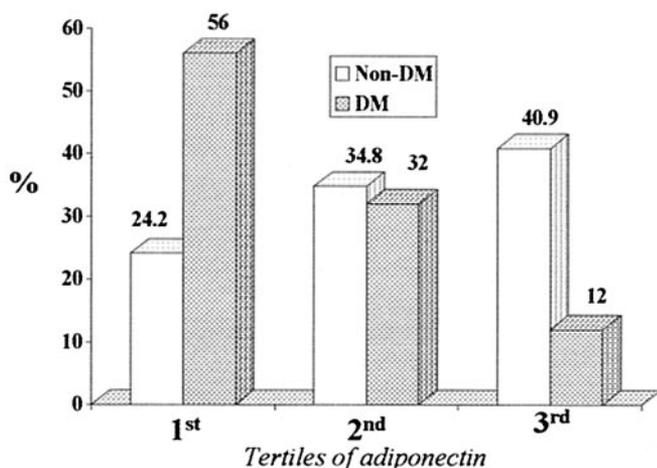
**Figure 1—***Comparison of the percentage distribution of subjects in whom diabetes developed and nondiabetic subjects in the tertiles of adiponectin (μg/ml): tertile 1 = 3.0–9.9; tertile 2 = 10.0–18.7; tertile 3 = 18.8–43.7. Baseline values of adiponectin were analyzed.*

**CONCLUSIONS** — The most important finding of this study was that low adiponectin concentration was predictive of prospective diabetes. We had studied a cohort of subjects with IGT who were at high risk for diabetes by virtue of increased 2-h plasma glucose level. Among these subjects, low adiponectin level was found to be a strong independent predictor of diabetes. Insulin resistance was not significantly different between the prospective diabetic and nondiabetic groups. This is the first study of the predictive value of low adiponectin level in a non-white population. The predictive nature of low concentrations of adiponectin for diabetes has been demonstrated in Pima Indians (7) and in the European Prospective Investigation into Cancer and Nutrition (EPIC) study in Germany (8). Lower concentrations of adiponectin found in the above studies, when compared with the values in our study group, may be partly related to the differences in BMI. As mentioned earlier, there may also be population-based differences in the adiponectin concentrations.

Our study shows that adiponectin concentrations in Asian Indians were similar to the values in the Native Canadian population (14). Both populations are experiencing a rapid increase in diabetes and cardiovascular disease. Valsamakis et al. (15) found that obese Indo-Asians in the U.K. had lower median levels of adiponectin than a matched group of Caucasians. The difference in the values between this study and the present study may be possibly related to the higher BMI of subjects in the U.K. study. Cnop et al. (16) reported lower levels in Americans with BMI comparable to the subjects in our study group. It is likely that concentrations of adiponectin may also vary in different populations.

Although we did not find an independent association between the abdominal obesity indicated by increased waist circumference, the possibility that adiponectin is closely related to intra-abdominal fat and plays an important role in the relationship between visceral adiposity and insulin resistance was not excluded. Elegant studies by Cnop et al. (16) in healthy subjects have shown evidence of such a relationship between intra-abdominal fat and adiponectin.

We observed a sex dimorphism in adiponectin in Asian Indians: women had higher levels than men. Similar reports have been published by some (4,16–18), whereas a few others have failed to observe the sex difference (19,20). The impact of female sex on adiponectin concentrations was shown to be independent of age, BMI, abdominal fat, and insulin sensitivity in healthy subjects (16).

In summary, we have observed that low adiponectin concentration is a strong independent predictor of future development of diabetes in subjects with IGT. Adiponectin may have a central role in the pathogenesis of type 2 diabetes. More detailed studies in Asian Indians are warranted to understand the complex relation between insulin sensitivity, body fat distribution, and adiponectin concentration.

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**Table 2—***Results of multiple logistic regression analysis (diabetic versus nondiabetic group)*

| Significant variables | β | SEβ | *P* | OR | 95% CI |
|---|---|---|---|---|---|
| HbA$_{1c}$ (%) | 1.0013 | 0.484 | 0.038 | 2.72 | 1.05–7.03 |
| Adiponectin (μg/ml) | −0.139 | 0.046 | 0.003 | 0.87 | 0.79–0.95 |
| Constant | −5.28 | 2.99 | | | |

Not significant: age, sex, BMI, waist circumference, fat percentage, homeostasis model assessment of insulin resistance, fasting plasma glucose level, and 2-h plasma glucose level.

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