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Circulating Adiponectin Levels Are Associated with Better Glycemic Control, More Favorable Lipid Profile, and Reduced Inflammation in Women with Type 2 Diabetes

Christos S. Mantzoros, Tricia Li, JoAnn E. Manson, James B. Meigs, and Frank B. Hu

Division of Endocrinology, Diabetes, and Metabolism, Department of Medicine, Beth Israel Deaconess Medical Center, Harvard Medical School (C.S.M.); Departments of Nutrition (T.L., F.B.H.) and Epidemiology (J.E.M., F.B.H.), Harvard School of Public Health; Channing Laboratory (J.E.M., J.B.M., F.B.H.) and Division of Preventive Medicine (J.E.M.), Department of Medicine, Brigham and Women's Hospital and Harvard Medical School, Boston, Massachusetts 02215; and General Medicine Division (J.B.M.), Department of Medicine, Massachusetts General Hospital and Harvard Medical School, Boston, Massachusetts 02114

Context: Low adiponectin levels, by regulating insulin resistance and metabolic profile, may contribute to the markedly increased risk of atherosclerosis in diabetic subjects.

Objective: The complex interrelationships between adiponectin and metabolic abnormalities have not yet been fully assessed in diabetic women.

Design/Setting/Patients: We performed a cross-sectional evaluation of the association between circulating adiponectin and glycemia, lipid-lipoprotein levels, and inflammatory markers in 925 women with type 2 diabetes enrolled in the Nurses' Health Study.

Results: Circulating adiponectin levels were significantly and positively associated with high-density lipoprotein (HDL) cholesterol and physical activity levels, and inversely with body mass index and plasma concentrations of hemoglobin A_{1c} ($HgbA_{1c}$), triglycerides, non-HDL cholesterol, apolipoprotein B-100, C-reactive protein, fibrinogen, soluble E-selectin, and soluble intercellular adhesion molecule-1. The above associations were not appreciably altered after adjusting for lifestyle factors, existing medical conditions, obesity, and body fat distribution, with the exception of $HgbA_{1c}$ and soluble intercellular adhesion molecule-1 (which became nonsignificant). Associations between adiponectin and inflammatory markers persisted after control for the potential confounding effects of $HgbA_{1C}$ and HDL cholesterol, suggesting that the antiinflammatory properties of adiponectin are not mediated by its effect on glycemia and lipidemia. With the exception of the associations with triglycerides and apolipoprotein B_{100}, which were significant only in subjects with body mass index less than 30, all other associations observed herein were consistent among obese and nonobese diabetic women.

Conclusions: In summary, higher adiponectin levels are associated with better glycemic control, more favorable lipid profile, and reduced inflammation in diabetic women. **(*J Clin Endocrinol Metab* 90: 4542–4548, 2005)**

ADIPONECTIN, AN ADIPOCYTE-derived hormone, is thought to play an important role in regulating glycemia, lipidemia, endothelial dysfunction, and proinflammatory mechanisms in humans, all of which may contribute to the markedly increased risk of atherosclerosis (1–4). Adiponectin levels circulate in high concentrations (5) and are inversely associated with obesity, especially central obesity, as well as with hyperlipidemia and insulin resistance (6, 7). In addition, adiponectin seems to have substantial antiinflammatory properties *in vitro* and in animal studies *in vivo* (4) and has been shown to be a predictor of the development of type 2 diabetes and cardiovascular events in diabetics (8–10).

Previous studies have not yet fully evaluated potential associations between adiponectin levels and circulating inflammatory markers and/or lipoprotein abnormalities among diabetic women. More specifically, previously reported associations with fibrinogen, apolipoprotein B ($apoB_{100}$) (11), or TNF-α levels (12) in diabetic men have not yet been studied in women. It also remains unknown whether previously reported associations are independent of potentially confounding lifestyle factors, anthropometric measures, and glycemia in men and women. Furthermore, it remains unknown whether adiponectin levels are associated with soluble E-selectin (sE-selectin) and lipoprotein(a) [Lp(a)] levels in men or women. Thus, in the context of the prospective cohort Nurses' Health Study (NHS), we have examined potential independent associations between circulating adiponectin levels and hemoglobin A_{1c} ($HgbA_{1c}$), blood lipid and lipoprotein levels, as well as several inflammatory markers in 925 diabetic women.

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Abbreviations: $apoB_{100}$, Apolipoprotein B-100; BMI, body mass index; CRP, C-reactive protein; CV, coefficient of variation; CVD, cardiovascular disease; HDL, high-density lipoprotein; $HgbA_{1c}$, hemoglobin A_{1c}; LDL, low-density lipoprotein; Lp(a), lipoprotein(a); Q, quartile; sICAM, soluble intercellular adhesion molecule; sTNFR, soluble TNF receptor; WHR, waist to hip ratio.

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Subjects and Methods

Study population

The NHS was initiated in 1976 with the enrollment of 121,700 U.S. nurses, aged 30–55 yr. This prospective cohort study involves biannually mailed questionnaires related to lifestyle factors and health outcomes. In 1989–1990, 32,826 study participants provided blood samples

by overnight courier. The present study included 925 women with a confirmed diagnosis of type 2 diabetes and without prevalent or incident malignancy, 780 of whom did not report a prior diagnosis of myocardial infarction, coronary revascularization, or stroke at the time of blood drawing.

Definition of diabetes

Cases of diabetes were reported by the respondents on the biannual questionnaires. A supplementary questionnaire was mailed to all women reporting a diagnosis of diabetes to obtain additional information about the date of diagnosis, symptoms, diagnostic tests, and treatment. In accordance with the criteria of the National Diabetes Data Group, confirmation of diabetes required at least one of the following self-reports on the supplementary questionnaire: 1) an elevated plasma glucose concentration (fasting plasma glucose ≥140 mg/dl, random plasma glucose ≥200 mg/dl, and/or plasma glucose, ≥200 mg/dl after ≥2 h during an oral glucose tolerance test) plus at least one classic symptom (excessive thirst, polyuria, weight loss, or hunger); 2) no symptoms, but at least two elevated plasma glucose concentrations (by the above criteria) on different occasions; or 3) treatment with hypoglycemic medication (insulin or oral hypoglycemic agent). We used the National Diabetes Data Group criteria to define diabetes because all our subjects were diagnosed before the American Diabetes Association released their criteria in 1997. The validity of self-reported diagnosis of type 2 diabetes by our supplementary questionnaire has been established by a separate and independent validation study through medical record reviews (13).

Blood collection and processing

Blood was drawn in 1989–1990. Participants were sent a blood-set kit that included supplies (blood tubes, tourniquet, needles, bandage, and coolant pack) and instructions. Participants arranged for the blood to be drawn and sent the samples back by prepaid overnight courier. Most samples arrived within 24 h of the blood drawing. After arrival in the laboratory, samples were centrifuged and aliquoted into cryotubes as plasma, buffy coat, and red blood cells. Cryotubes were stored in liquid-nitrogen freezers at −130 C or lower.

Adiponectin was assayed by RIA [Linco Research, Inc., St. Charles, MO; sensitivity, 2 μg/ml; intraassay coefficients of variation (CV), 1.78–6.21%] as previously described (14). Plasma was assayed for the presence of soluble TNF-α receptor II (sTNF-αRII) using the human sTNF-αRII ELISA kit (R&D Systems, Minneapolis, MN). The minimum detectable range of this assay is 0.6 pg/ml for a sample size of 200 μl (diluted 1:10), with a range up to 500 pg/ml. Intraassay precision was determined using three samples of known concentration tested 20 times on one plate; the CV range was 2.6–4.8%. Interassay precision was determined from three samples of known concentration tested on 20 separate plates; the CV range was 3.5–5.1%. Plasma C-reactive protein (CRP) was measured using the US CRP ELISA kit (Diagnostic Systems Laboratories, Inc., Webster, TX) with a CV range of 2.8–5.1%. Because the CRP levels from the current assay consistently read higher CRP levels compared with an assay previously used in our studies (15, 16), we performed a cross-validation study using 204 samples obtained from the diabetic women cohort, in which the CRP levels were measured by both methods. The correlation coefficient between the two methods was 0.97, suggesting that the results reported using this assay should be comparable with those of assays previously reported. Soluble intercellular adhesion molecules (sICAM-1) were assayed in plasma using the human sICAM-1 ELISA kit (R&D Systems) with a CV range of 3.3–4.8%. Plasma levels of sE-selectin were assayed using the human sE-selectin ELISA kit (R&D Systems) with a CV range of 5.7–8.8%. Concentrations of glycosylated hemoglobin ($HgbA_{1c}$) were based on turbidimetric immunoinhibition with hemolyzed whole blood or packed red cells with a CV of less than 3.0%. Fibrinogen was measured on a Hitachi 911 analyzer (Tokyo, Japan) using reagents and calibrators from Kamiya Biomedical Co. (Seattle, WA) with a CV of 1.16%. The concentrations of total cholesterol, high-density lipoprotein (HDL) cholesterol, and triglycerides were measured simultaneously on the Hitachi 911 analyzer with reagents and calibrators from Roche (Indianapolis, IN); the CVs for these measurements were less than 1.8%. Concentrations of low-density lipoprotein (LDL) cholesterol were measured using a homogenous direct method from Genzyme (Cambridge, MA) with a CV less than 3.1%. Concentrations of $apoB_{100}$ were measured in an immunonephelometric assay using reagents and calibrators from Wako Chemicals (Richmond, VA) with a CV less than 5%.

Assessment of lifestyle exposures

We calculated body mass index (BMI) as the ratio of weight (in kilograms) to the height squared (in meters squared). Physical activity was computed as hours per week using the duration of moderate or vigorous forms of exercise per week (17). History of hypertension and family history of myocardial infarction were determined from self-reports before blood collection. Alcohol intake was estimated with a dietary questionnaire in 1990.

Statistical analysis

Spearman correlations and scatter plots were used to evaluate bivariate relationships between plasma levels of adiponectin and levels of lipoproteins or inflammatory markers. Multivariate linear regression analyses with robust variance were performed to evaluate the associations between adiponectin and biomarkers without the need for normal distribution assumptions (18). We adjusted for age, BMI, physical activity (quartiles of metabolic equivalents), smoking (never, past, and current), aspirin use, history of cardiovascular disease, history of high blood pressure, history of high blood cholesterol, baseline and fasting status, alcohol intake (0.0, 0.1–4.9, 5.0–9.9, 10.0–14.9, and ≥15.0 g/d), insulin use, postmenopausal status, and hormone use as indicated in Table 3. In separate models, we also adjusted for waist to hip ratio (WHR; Table 3; in the 565 women who had WHR measurements) as well as $HgbA_{1C}$, triglycerides, and/or HDL as appropriate (data not shown). We then tested for effect modifications by insulin use or obesity status, with adjustment for potential confounders. Because fasting triglyceride, lipid, and leptin levels were available for only 615 subjects, we conducted secondary analyses among these samples. Finally, we performed separate analyses of the entire sample (n = 925) and of the subgroup of women free of cardiovascular disease (CVD) at baseline (n = 722) or nonusers of insulin (n = 631). All statistical analyses were performed using SAS statistical software version 8 (SAS Institute, Inc., Cary, NC).

Results

In this study of 925 diabetic women enrolled in the NHS, higher plasma adiponectin levels were associated with lower BMI and greater physical activity, whereas smoking and alcohol consumption were not clearly related to adiponectin levels (Table 1). Women with higher adiponectin levels were more likely to use insulin and less likely to have a history of hypertension. Estrogen use tended to be associated with higher adiponectin levels ($P = 0.076$). Adiponectin levels were not associated with LDL or total cholesterol, whereas they were strongly and positively associated with HDL and Lp(a) and strongly inversely associated with non-HDL, triglycerides, and $apoB_{100.}$ Adiponectin levels were also inversely and strongly associated with fibrinogen, sICAM-1, sE-selectin, and CRP. Half of the subjects reported aspirin use and/or a family history of diabetes, and approximately 25% had a family history of myocardial infarction, but adiponectin levels were not associated with either one by bivariate analysis. With the exception of nonfasting leptin levels as well as levels of sTNF-αRII, $HgbA_{1c}$ and LDL, which were not associated with adiponectin, as well as Lp(a) and sICAM-1, for which the association was not monotonically increasing, all other biomarkers increased or decreased monotonically with increasing adiponectin levels.

To assess the observed associations, we then calculated Spearman correlation coefficients (Table 2). Adiponectin was positively and strongly associated with HDL (r = 0.52) and

TABLE 1. Characteristics by quartiles of adiponectin in 925 diabetic women

Variable	Quartiles of adiponectin				*P* for trend
	Q1 0.8–3.65	Q2 3.66–5.83	Q3 5.84–9.58	Q4 9.59–42.07	
Adiponectin (μg/ml)	2.7 ± 0.7	4.7 ± 0.6	7.3 ± 1.0	16.9 ± 7.2	
Age (yr)	58.3 ± 6.7	60.4 ± 5.9	60.3 ± 6.5	59.1 ± 6.7	0.83
BMI (kg/m^2)	31.4 ± 6.0	30.9 ± 6.0	30.0 ± 6.3	26.4 ± 5.7	<0.001
Physical activity (h/wk)	1.14 ± 1.84	1.55 ± 2.59	1.66 ± 2.67	2.46 ± 3.60	<0.001
Alcohol consumption (g/d)	2.6 ± 8.7	2.4 ± 7.9	2.1 ± 5.6	3.1 ± 7.1	0.37
Waist circumference (in)	24.5 ± 18.3	25.6 ± 17.9	26.6 ± 16.7	24.8 ± 14.5	0.95
WHR	0.88 ± 0.22	0.85 ± 0.07	0.85 ± 0.10	0.80 ± 0.10	<0.001
Current smokers (%)	15.7	14.4	10.3	13.9	0.38
Aspirin use (%)	53.5	47.8	48.0	48.5	0.56
Insulin use (%)	20.8	26.0	32.3	48.1	<0.001
Hormone use in postmenopausal women	23.8	29.0	32.3	36.2	0.076
History of hypertension (%)	74.5	67.1	68.1	54.6	<0.001
Baseline CVD[a] (%)	19.1	15.6	15.5	12.6	0.30
History of high cholesterol (%)	62.3	60.6	59.5	54.6	0.36
Family history of diabetes (%)	55.4	55.4	58.6	48.9	0.20
Family history of MI (%)	23	28	23	29	0.34
Fasting (%)	68.8	72.7	64.2	60.2	0.03
Total cholesterol (mg/dl)	221.4 ± 46.5	232.0 ± 42.7	233.8 ± 45.0	230.5 ± 46.6	0.13
HDL (mg/dl)	42.8 ± 10.0	47.2 ± 12.1	51.9 ± 13.6	64.6 ± 17.6	<0.001
LDL (mg/dl)	133.6 ± 42.0	143.7 ± 36.9	143.9 ± 39.3	139.8 ± 39.9	0.28
Non-HDL (mg/dl)	178.6 ± 46.6	184.8 ± 42.2	181.9 ± 45.3	166.0 ± 47.7	<0.001
Fasting triglycerides (mg/dl)	250.2 ± 210.2	228.8 ± 183.4	203.4 ± 154.3	152.6 ± 139.2	<0.001
$ApoB_{100}$ (mg/dl)	106.0 ± 25.0	108.7 ± 23.6	105.0 ± 24.5	94.3 ± 25.8	<0.001
Fibrinogen (mg/dl)	401.7 ± 95.8	390.2 ± 112.1	378.6 ± 101.6	351.2 ± 89.8	<0.001
Fasting leptin (ng/ml)	50.0 ± 25.4	49.6 ± 28.8	50.5 ± 30.0	43.3 ± 35.7	<0.001
Lp(a) (mg/dl)	17.6 ± 23.1	18.2 ± 23.3	20.8 ± 27.2	19.9 ± 25.6	0.02
$HgbA_{1c}$ (%)	7.2 ± 1.6	7.2 ± 1.7	6.9 ± 1.7	7.1 ± 1.9	0.08
sICAM-1 (ng/ml)	329.9 ± 98.1	322.9 ± 117.3	293.9 ± 82.9	296.3 ± 94.7	<0.001
sTNF-α RII (pg/ml)	2637.4 ± 829.8	2625.6 ± 789.7	2662.5 ± 864.6	2606.9 ± 985.0	0.38
sE-Selectin (ng/ml)	77.8 ± 39.4	69.3 ± 44.5	60.6 ± 32.6	54.6 ± 34.8	<0.001
CRP (mg/liter)	7.3 ± 6.7	5.9 ± 5.8	5.4 ± 5.0	4.0 ± 5.3	<0.001

Values shown are the mean ± SD for the respective quartiles (crude values).
[a] Self-reported CVD at baseline, MI, myocardial infarction.

inversely with triglycerides (r = −0.30). Inverse associations were found with CRP (r = −0.32) and sE-selectin (r = −0.28). Significant and inverse associations were also found with $apoB_{100}$, fibrinogen, and sICAM-1, but the corresponding correlation coefficients were relatively weak (r ≅ −0.20).

Cholesterol fractions and triglyceride levels were significantly interrelated, as expected. Both leptin, an indicator of overall fat mass, and glycemia, as expressed by $HgbA_{1c}$, were positively and significantly associated with inflammatory markers (all r ≥ 0.19, except for sICAM-1 with leptin r = 0.10)

We then used multivariate linear regression to estimate the changes in blood lipids and inflammatory markers corresponding to a change in adiponectin levels by 10 μg/ml before and after adjusting for age, physical activity, smoking, and other covariates, including BMI and/or WHR, as indicated in Table 3. No significant associations with $HgbA_{1c}$ were detected, whereas a 10 μg/ml increase in adiponectin levels was associated with a significant decrease in triglyceride levels by 47.4 mg/dl (approximately −25% compared with the mean level), an 11.4 mg/dl increase in HDL (22%), and significant reductions in apoB100 (−5.68 mg/dl; 6%), fibrinogen (−19.4 mg/dl; 5%), sE-selectin (−7.76 ng/ml; 12%), and CRP (−1.02 mg/liter; 20%). These associations remained essentially unchanged after adjusting for WHR as an indicator of central obesity. Importantly, the weak but significant and inverse association between adiponectin levels and $HgbA_{1c}$ in diabetic women revealed by bivariate analysis became nonsignificant after adjustment for body fat distribution.

The association between adiponectin and Lp(a) remained nonsignificant after controlling for all the variables shown in Table 3 and triglycerides or HDL and $HgbA_{1c}$, respectively. Adiponectin remained significantly associated with $apoB_{100}$, CRP, fibrinogen, sE-selectin, and sTNF-αRII I after additional adjustment for triglycerides or HDL and $HgbA_{1c}$, suggesting that the associations reported in this study were particularly robust.

We obtained similar data when we studied all study subjects or only subjects who had fasting triglyceride and leptin levels (data not shown). We also evaluated whether the observed associations were modified by baseline history of CVD. We thus repeated the above analysis in women free of cardiovascular disease at baseline (n = 722) and obtained similar results (data not shown). Lastly, we studied whether the reported associations were relatively consistent across obesity strata, and this was the case for most variables, *i.e.* interaction terms were nonsignificant for the following variables: CRP (P = 0.80), sE-selectin (P = 0.61), sTNF-αRII (P = 0.08), sICAM-1 (P = 0.052), fibrinogen (P = 0.19), Lp(a) (P = 0.61), and HDL (P = 0.057). In contrast, interaction terms were significant for apoB ($P < 0.001$) and triglycerides ($P <$ 0.001). More specifically, stratification for BMI less than 30 *vs.* BMI of 30 or more revealed that the associations between adiponectin and both apoB and triglycerides were negative

TABLE 2. Spearman correlation between adiponectin and blood lipids, $HgbA_{1c}$, and inflammatory markers in 925 diabetic women

	Adiponectin	Total cholesterol	Fasting triglycerides	HDL cholesterol	LDL cholesterol	Non-HDL cholesterol	$ApoB_{100}$	Fibrinogen
Adiponectin	1	0.07[a]	−0.30[a]	0.52[a]	0.045	−0.11[a]	−0.19[a]	−0.20[a]
Total cholesterol		1	0.36[a]	0.13[a]	0.86[a]	0.92[a]	0.84[a]	0.12[a]
Fasting triglycerides			1	−0.52[a]	0.16[a]	0.54[a]	0.55[a]	0.17[a]
HDL cholesterol				1	0.09[a]	−0.20[a]	−0.27[a]	−0.15[a]
LDL cholesterol					1	0.83[a]	0.82[a]	0.10[a]
Non-HDL cholesterol						1	0.94[a]	0.17[a]
$ApoB_{100}$							1	0.20[a]
Fibrinogen								1
Fasting leptin								
Lp(a)								
$HgbA_{1c}$ (%)								
sICAM-1								
sE-Selectin								
sTNF-α RII								
CRP								

	Fasting leptin	Lp(a)	$HgbA_{1c}$ (%)	sICAM-1	sE-Selectin	sTNF-α RII	CRP
Adiponectin	−0.11[a]	0.09[a]	−0.08[a]	−0.16[a]	−0.28[a]	−0.03	−0.32[a]
Total cholesterol	0.08	0.06	0.11[a]	−0.087[a]	−0.04	−0.07[a]	0.05
Fasting triglycerides	0.11[a]	−0.13[a]	0.19[a]	0.07	0.18[a]	0.13	0.26[a]
HDL cholesterol	−0.10[a]	0.12[a]	−0.13[a]	−0.25[a]	−0.30[a]	−0.22[a]	−0.23[a]
LDL cholesterol	0.08[a]	0.13[a]	0.10[a]	−0.04	−0.02	−0.08[a]	0.01
Non-HDL cholesterol	0.11[a]	0.016	0.14[a]	−0.008	0.06	0.008	0.12[a]
$ApoB_{100}$	0.11	0.04	0.16[a]	0.06	0.13[a]	0.02	0.18[a]
Fibrinogen	0.24[a]	0.06	0.18[a]	0.11[a]	0.14[a]	0.22[a]	0.34[a]
Fasting leptin	1	−0.025	0.04	0.10[a]	0.19[a]	0.30[a]	0.34[a]
Lp(a)		1	−0.02	−0.04	−0.07[a]	−0.005	−0.02
$HgbA_{1c}$ (%)			1	0.24[a]	0.33[a]	0.19[a]	0.21[a]
sICAM-1				1	0.52[a]	0.38[a]	0.29[a]
sE-Selectin					1	0.22[a]	0.28[a]
sTNF-α RII						1	0.31[a]
CRP							1

[a] $P < 0.05$.

and significant only for the BMI less than 30 stratum [quartile 1 (Q1) = 11.96, Q2 = 9.71, Q3 = 7.71, Q4 = 7.80 ($P < 0.001$); and Q1 = 12.04, Q2 = 8.56, Q3 = 8.18, Q4 = 5.76 ($P < 0.001$), respectively]. In contrast, associations between adiponectin and apoB or triglycerides were null in the stratum of diabetic women with BMI of 30 or more. Similar to obesity, stratification for insulin use revealed results that were not significant for CRP ($P = 0.24$); sTNF-α RII ($P = 0.70$), sICAM-1 ($P = 0.78$), sE-selectin ($P = 0.53$), Lp(a) ($P = 0.08$), or fibrinogen ($P = 0.21$), but were significant for triglycerides ($P = 0.008$)

TABLE 3. Parameter estimates and *P* values for a 10 μg/dl increase in adiponectin levels in relation to $HgbA_{1c}$, blood lipids, and inflammatory makers in 925 diabetic women

Biomarker	Age adjusted		Multivariate adjusted[a]		Multivariate[b] adjusted	
	Estimate	*P*	Estimate	*P*	Estimate	*P*
$HgbA_{1c}$ (%)	−0.04	0.68	−0.12	0.14	−0.11	0.22
Total cholesterol	0.04	0.98	5.71	0.015	5.67	0.017
Fasting triglycerides	−62.20	<0.001	−47.40	<0.001	−42.50	<0.001
HDL cholesterol	12.80	<0.001	11.40	<0.001	10.90	<0.001
LDL cholesterol	−1.81	0.35	2.49	0.24	2.46	0.24
Non-HDL cholesterol	−12.80	<0.001	−5.74	0.009	−5.20	0.02
$ApoB_{100}$	−9.60	<0.001	−5.68	<0.001	−5.14	<0.001
Fibrinogen	−30.00	<0.001	−19.40	<0.001	−20.80	<0.001
Fasting leptin	−4.82	0.056	2.58	0.23	2.33	0.31
Lp(a)	1.66	0.21	1.12	0.41	1.27	0.37
sICAM-1	−11.00	0.034	−5.03	0.34	−3.46	0.52
sTNF-αRII	23.20	0.61	140.00	0.002	136.10	0.003
sE-Selectin	−10.98	<0.001	−7.76	<0.001	−6.13	0.002
CRP	−1.88	<0.001	−1.02	<0.001	−1.07	<0.001

[a] Multivariate adjusted: age at blood draw, BMI at the year of blood draw, smoking status [never smoked, former smoker, or current smoker (1–14, 15–24, or 25 or more cigarettes/d)], alcohol consumption (none, 0.1–4.9, 5.0–14.9, or 15 g or more/d), postmenopausal status, and hormone use (never used hormones, used them in past, or use them currently), level of physical activity (four categories), aspirin use at baseline, fasting status at blood drawing, baseline hypertension, baseline cardiovascular disease, baseline high blood cholesterol, and use of insulin.

[b] Except for variables in multivariate adjusted model, WHR in quintiles is also included in this model.

and $apoB_{100}$ levels ($P = 0.002$). Both triglycerides and $apoB_{100}$ levels were significantly and inversely associated with adiponectin in analysis stratifying for insulin use, but the inverse association was much more pronounced in the group of insulin users (n = 180).

Discussion

Accumulating evidence from animal and human studies demonstrates that adiponectin plays an important role in the pathophysiology of insulin resistance, diabetes (9, 19–22), lipid metabolism (7, 19), and inflammation (23) and thus affects risk for cardiovascular disease (23). In this study we found that, similar to diabetic men (11), adiponectin levels were strongly and positively associated with HDL, whereas they were strongly, but inversely, associated with triglycerides, non-HDL, and $apoB_{100}$ among women with type 2 diabetes. We also confirm previously shown inverse associations with fibrinogen (11), CRP, and sICAM-1 (11, 12) and demonstrate for the first time a weak, positive association with Lp(a), which does not persist after multivariate adjustment, an inverse association with sE-selectin, and a null association with sTNF-αRII. Furthermore, we demonstrate that the reported associations were independent from the potential confounding effect of lifestyle factors such as smoking, alcohol consumption, and physical activity as well as body fat mass and body fat distribution, aspirin or hormone use, history of hypertension or hypercholesterolemia, or family history of myocardial infarction in diabetic women. Importantly, the reported associations with inflammatory markers were also independent from glycemia and lipidemia, as reflected by adjusting for $HgbA_{1c}$, total cholesterol, and HDL, suggesting that adiponectin may have direct antiinflammatory effects not mediated by body composition, glycemia, or lipidemia. Thus, in addition to mediating the effect of overall and central adiposity, adiponectin may have direct effects on cardiovascular risk factors.

Adiponectin is an adipocyte-derived protein that has been closely and inversely associated with obesity, body fat distribution, insulin resistance, and atherosclerosis (24). Adiponectin knockout mice develop insulin resistance (19, 25). Low adiponectin levels are associated with insulin resistance and predict risk for developing diabetes in animal models (20, 26, 27) and in humans (22), and administration of adiponectin to rodents or of medications that increase adiponectin levels in humans improves insulin sensitivity.

In vitro mechanistic studies have demonstrated that recombinant human adiponectin suppresses in a dose-dependent manner endothelial expression of adhesion molecules, the proliferation of vascular smooth muscle cells, and the transformation of macrophages to foam cells (28). Moreover, adiponectin both decreases the attachment of monocytic THP-1 cells to human aortic endothelial cells and suppresses the secretion of TNF-α from human monocyte/macrophages (29) as well as the action of TNF-α (23, 28, 30). In mice, adiponectin suppresses the expression of adhesion molecules, scavenger receptors, and TNF-α levels (31). Adiponectin may also exert antiinflammatory effects indirectly through its effects on glycemia, which may affect circulating cytokine concentrations (32, 33) and lipidemia (11), altering the expression of adhesion molecules on vascular endothelial cells (34) and inhibiting platelet aggregation (35). The independent association between adiponectin and inflammatory markers shown herein demonstrates that adiponectin's effects on inflammatory markers are independent from the potential confounding effect of other known cardiovascular risk factors. The weak and borderline significant (11), or nonsignificant after multivariate adjustment, association between adiponectin and sICAM-1 or soluble vascular cell adhesion molecule may indicate that adiponectin plays a less important role in monocyte adhesion, although its role in subsequent stages of arteriosclerosis, *i.e.* macrophage cytokine production and macrophage to foam cell transformation, is probably more important (4, 11).

Our data also indicate that a decrease in adiponectin levels by 10 μg/ml corresponds to a decrease in HDL by approximately 25%, an increase in triglycerides by approximately 25% and in non-HDL by about 7%, as well as substantial increases in $apoB_{100}$. Similar associations with HDL and triglycerides have been observed by our group and others in nondiabetic and diabetic (11, 12, 36, 37) subjects. Inverse associations with $apoB_{100}$ have been reported in two, but not a third, study (11, 38). Adiponectin mediates only in part the effects of body fat distribution on lipids and lipoproteins (6, 36–42), because the associations reported herein weaken, but remain largely independent from, several other potential confounders, including overall obesity and central body fat distribution.

Adiponectin levels have also been associated with glucose tolerance and plasminogen activator inhibitor-1 independently of waist circumference or WHR in women with prior gestational diabetes mellitus (43). We report for the first time that adiponectin levels are inversely and significantly associated with glycemic control in the diabetic women we studied, but this association becomes nonsignificant after adjusting for potential confounding variables, including body fat distribution. Adiponectin increases insulin-induced tyrosine phosphorylation of the insulin receptor in skeletal muscle to improve glucose tolerance and also acts directly at the level of the muscle and the liver to increase free fatty acid oxidation and clearance; it may also affect lipid metabolism through additional mechanisms, which remain to be fully elucidated (20, 22, 27).

The strengths of this study include the use of a well-established cohort, the NHS, and the high power provided by the large study sample. The limitations of this study include its cross-sectional nature, which precludes inference of causality, and the lack of adjustment for a potential effect of lipid lowering and oral hypoglycemic medications. Nondifferential misclassification due to the lack of collection of detailed data on medications at the time of blood drawing could result in depression of effect estimates toward the null and could thus have potentially only underestimated the corresponding *P* values reported. Finally, the associations reported in this study are consistent with a large and accumulating body of evidence (20, 32, 33) on the role of adiponectin in metabolism and inflammation, but some weak associations, albeit statistically significant due to the high power of the study, may not be as clinically important.

In conclusion, circulating adiponectin levels in diabetic

women are associated with improved glycemia and lipidemia as well as a lower inflammatory state. Our findings of an inverse and weak association between adiponectin levels and levels of sICAM-1 (11, 12) as well as in inverse and strong association with CRP and fibrinogen confirm previous reports (11, 12, 36, 39, 42–46) and extend them by indicating the independence of associations after multivariate adjustments for many potential confounders, whereas the reported inverse association with sE-selectin, the weak association with Lp(a), which becomes nonsignificant by multivariate adjustment, and the null association with sTNF-α RII are novel. Importantly, this is also the first time several of these associations have been studied in diabetic women and have been shown to be independent of other known risk factors for cardiovascular disease. The strong independent associations with lipoprotein levels and inflammatory markers support the idea that adiponectin may have a beneficial effect on the development of atherogenic lesions by exerting direct anti-inflammatory and antiatherogenic actions. Lifestyle changes and medications that increase adiponectin levels warrant additional study, because they may improve the cardiovascular risk profile and decrease cardiovascular morbidity and mortality in type 2 diabetes.

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Address all correspondence and requests for reprints to: Dr. Christos S. Mantzoros, Division of Endocrinology, Diabetes, and Metabolism, Beth Israel Deaconess Medical Center, Harvard Medical School, 330 Brookline Avenue, ST 816, Boston, Massachusetts 02215. E-mail: cmantzor@bidmc.harvard.edu.

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