

Plasma Adiponectin Levels and Risk of Myocardial Infarction in Men

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ADIPONECTIN (ARCP30, ADIPOQ, apM1, or GBP28) is a recently discovered 247 amino acid peptide, predominantly secreted by adipocytes, that accounts for about 0.05% of total serum proteins.¹⁻⁴ It is induced early in adipocyte differentiation,¹ consists of an N-terminal collagenous and a C-terminal globular domain, and shares homology with subunits of complement factor C1q.^{1,3} Adiponectin expression is reduced in obesity, insulin resistance, and type 2 diabetes, and plasma concentrations are inversely related to body weight and insulin levels, and reflect peroxisome proliferator-activated receptor γ (PPAR- γ) activation.⁵⁻⁹ Treatment with adiponectin improves insulin sensitivity in animal models of insulin resistance^{10,11} and reverses diet-induced insulin resistance in adiponectin knockout mice.¹² Low plasma adiponectin levels have recently been shown to predict risk of developing type 2 diabetes in humans.^{8,13} Adiponectin is also inversely associated with other traditional cardiovascular risk factors, such as blood pressure, heart rate, and total and low-density lipoprotein (LDL) cholesterol and triglyceride levels,^{14,15} and is positively related to high-density lipoprotein (HDL) cholesterol levels.^{14,16} Furthermore, recent studies suggest that it may have antiatherogenic and anti-inflammatory properties.¹⁷⁻²² These data

Context Adiponectin, a recently discovered adipocyte-derived peptide, is involved in the regulation of insulin sensitivity and lipid oxidation and, purportedly, in the development of atherosclerosis and coronary heart disease in humans.

Objective To assess prospectively whether plasma adiponectin concentrations are associated with risk of myocardial infarction (MI).

Design, Setting, and Participants Nested case-control study among 18225 male participants of the Health Professionals Follow-up Study aged 40 to 75 years who were free of diagnosed cardiovascular disease at the time of blood draw (1993-1995). During 6 years of follow-up through January 31, 2000, 266 men subsequently developed nonfatal MI or fatal coronary heart disease. Using risk set sampling, controls were selected in a 2:1 ratio matched for age, date of blood draw, and smoking status (n=532).

Main Outcome Measure Incidence of nonfatal MI and fatal coronary heart disease by adiponectin level.

Results After adjustment for matched variables, participants in the highest compared with the lowest quintile of adiponectin levels had a significantly decreased risk of MI (relative risk [RR], 0.39; 95% confidence interval [CI], 0.23-0.64; *P* for trend <.001). Additional adjustment for family history of MI, body mass index, alcohol consumption, physical activity, and history of diabetes and hypertension did not substantially affect this relationship (RR, 0.41; 95% CI, 0.24-0.70; *P* for trend <.001). Further adjustment for hemoglobin A_{1c} or C-reactive protein levels also had little impact, but additional adjustment for low- and high-density lipoprotein cholesterol levels modestly attenuated this association (RR, 0.56; 95% CI, 0.32-0.99; *P* for trend = .02).

Conclusions High plasma adiponectin concentrations are associated with lower risk of MI in men. This relationship can be only partly explained by differences in blood lipids and is independent of inflammation and glycemic status.

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suggest that high plasma adiponectin levels may be related to a lower risk of coronary heart disease (CHD), but data in humans are lacking. Therefore, we conducted a case-control study nested within the Health Professionals Follow-up Study (HPFS) to assess the association between baseline plasma adiponectin levels and risk of myocardial infarction (MI) over a follow-up period of 6 years.

METHODS

Study Population

The HPFS is a prospective cohort investigation among 51 529 US male health care professionals aged 40 to 75 years at baseline in 1986, designed pri-

marily to evaluate associations between diet and chronic disease incidence.²³ Information about health and disease is assessed biennially by a self-

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administered questionnaire and diet every 4 years by a self-administered food frequency questionnaire.²⁴ Between 1993 and 1995, a blood sample was requested from all participants and returned by 18 225 participants. Men who provided samples were somewhat younger but were otherwise similar to those who did not provide samples. Based on this sample and after exclusion of participants with a history of cardiovascular disease prior to 1994, we identified 266 participants with incident nonfatal MI or fatal CHD between date of blood draw and return of the 2000 questionnaire (January 2000). Controls were randomly selected from participants with a blood sample and without history of cardiovascular disease at the time of case ascertainment in a 2:1 ratio and matched for age, date of blood draw, and smoking status (risk set sampling).²⁵ Our analysis includes 1 participant who was selected as a control and subsequently had an event during follow-up. No control was selected twice in our analysis during the random selection process.

All participants gave written informed consent, and the Harvard School of Public Health Human Subjects Committee Review Board approved the study protocol.

Assessment of Nonfatal MI and Fatal CHD

Study physicians blinded to participants' exposure status reviewed the medical records of all participants for whom nonfatal MI or fatal CHD was ever reported. Each questionnaire that is mailed biennially to participants of the HPFS contains a question on whether the participant has had "professionally diagnosed . . . myocardial infarction (heart attack)" in the preceding 2 years. Myocardial infarction was confirmed if it met the World Health Organization's criteria (symptoms plus either diagnostic electrocardiographic changes or elevated levels of cardiac enzymes).²⁶ In about 70% of participants with reported MI, the diagnosis was confirmed using these methods. Reasons for nonconfirmation were either

that no further information was available (because the participant or hospital did not consent to send the hospital records) or that a reported case was rejected. Nonconfirmed participants were excluded from the selection process. Deaths were identified from state vital statistics records and the National Death Index or reported by next of kin or the postal system. Fatal CHD was considered to have occurred if there was fatal MI confirmed by hospital records or on autopsy or if CHD was listed as the cause of death on the death certificate, if it was the underlying and most plausible cause, and if evidence of previous CHD was available. In our analysis, 196 participants had nonfatal MI and 70 had fatal CHD as the qualifying event.

Assessment of Medical History, Anthropometric Data, and Diet and Lifestyle Factors

Anthropometric data, lifestyle factors, and diet were derived from the 1994 questionnaire. Body mass index was calculated as weight in kilograms divided by the square of height in meters. Nutrient intake was computed based on a semiquantitative food frequency questionnaire (which inquires about average food intake during the past year) using composition values from the US Department of Agriculture sources,²⁷ supplemented with other data. Physical activity was expressed as metabolic equivalent task (MET)-hours based on self-reported types and durations of activities over the previous year.²⁸ One MET-hour is equivalent to energy expenditure while sitting quietly for 1 hour. Medical history was derived from the questionnaires between 1986 and 1994. The questionnaires and the validity and reproducibility of the collected data and measurements have been reported in detail elsewhere.^{24,29-33}

Measurement of Biochemical Variables

Blood samples were collected in three 10-mL liquid EDTA blood tubes, placed on ice packs, stored in Styrofoam containers, and returned to our labora-

tory via overnight courier, with more than 95% arriving within 24 hours. After arrival, blood samples were centrifuged and aliquoted for storage in the vapor phase of liquid nitrogen freezers (-130°C or colder). Fewer than 15% of the samples were slightly hemolyzed and very few were moderately hemolyzed ($<3\%$), lipemic ($<1\%$), or not cooled on arrival ($<0.5\%$).

Plasma adiponectin concentrations were measured by competitive radioimmunoassay (Linco Research Inc, St Charles, Mo) with a coefficient of variation of 3.4% ($n=39$). In a previous analysis, adiponectin levels had excellent intraclass correlation coefficients measured in participants over a period of 1 year and were not substantially affected by transport conditions.³⁴

Total cholesterol was measured enzymatically³⁵; LDL cholesterol by a homogeneous direct method from Genzyme Corp, Cambridge, Mass³⁶; HDL cholesterol using a direct enzymatic colorimetric assay³⁷; and triglycerides enzymatically with correction for endogenous glycerol.³⁸ The assays used for lipoprotein and lipid analysis are approved by the US Food and Drug Administration for clinical use, and coefficients of variation were less than 6%. Hemoglobin A_{1c} (HbA_{1c}) was measured by turbidimetric immunoinhibition, and C-reactive protein (CRP) concentrations were determined using an immunoturbidimetric high-sensitivity assay on a Hitachi 911 analyzer (Roche Diagnostics, Indianapolis, Ind) and reagents and calibrators from Denka Seiken (Niigata, Japan). The laboratory used (N.R.) is certified by the Centers for Disease Control and Prevention/National Heart, Lung, and Blood Institute Lipid Standardization Program.

Sixty-five percent of the participants in the present analysis provided fasting blood samples (≥ 8 hours since last meal; 67% among cases and 64% among controls) ($P=.36$). Data on LDL cholesterol and CRP levels were each missing in 1 participant; these values were replaced by the median concentrations in this cohort.

Statistical Analyses

Continuous variables are presented as means and standard deviations or medians and interquartile ranges and were compared between cases and controls using the unpaired *t* test or the Wilcoxon unpaired rank sum test. Proportions were compared using the χ^2 test. Associations between adiponectin levels and selected cardiovascular risk factors were examined in cases and controls using an age-adjusted Spearman partial correlation coefficient.

Adiponectin levels were categorized into quintiles based on control participants. Unconditional logistic regression adjusted for matched variables (age <50, 50-54, 55-59, 60-64, or ≥ 65 years; smoking status never, past, or current; and month of blood draw in 5 categories) was used to investigate the association between baseline adiponectin concentrations and incidence of nonfatal MI or fatal CHD (events). To test for linear trend across categories, we used log-transformed adiponectin levels. In our multivariable model, we further adjusted for family history of MI before age 60 years (yes/no), alcohol intake (nondrinker; 0.1-4.9, 5.0-14.9, 15.0-29.9, or ≥ 30.0 g/d; or missing), body mass index (<20, 20-24, 25-29, 30-34, or ≥ 35), physical activity (quintiles), and history of diabetes (yes/no) and hypertension (yes/no) at baseline. We repeated our main analysis using conditional logistic regression and found essentially the same results. Because of the design of our study, the odds ratio derived from logistic regression directly estimates the incidence rate (hazard) ratio and, therefore, the relative risk (RR).^{25,39}

We next examined the impact of potential intermediate biomarkers by sequentially adding LDL and HDL cholesterol and log-transformed triglyceride, HbA_{1c}, and CRP levels as continuous variables to our models, and estimated the multivariable-adjusted RR associated with an increase of (continuous) log-transformed adiponectin levels by 2 units on a log scale, which corresponds to a doubling in adiponectin levels on the original scale.

For stratified analysis, we also calculated the multivariable-adjusted RR associated with a doubling in adiponectin levels. We defined the metabolic syndrome similarly as recently proposed by the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults⁴⁰ as having 3 or more of the following 5 abnormalities: body mass index of at least 25; triglyceride levels of at least 150 mg/dL (0.20 mmol/L); HDL cholesterol levels of less than 40 mg/dL (1.04 mmol/L); history of hypertension; and history of diabetes, development of diabetes during follow-up, or HbA_{1c} levels of at least 7% at baseline. The distribution of metabolic abnormalities in controls in the present study was similar to that recently reported for the third National Health and Nutrition Examination Survey (≥ 1 metabolic abnormalities, 77.6% vs 71.5%; ≥ 2 , 48.9% vs 44.9%; ≥ 3 , 26.3% vs 24.0%; ≥ 4 , 9.0% vs 11.1%; and 5, 1.7% vs 2.4%).⁴¹ Results were similar when we restricted our analysis to fasting participants only. We tested interactions between adiponectin levels and subgroups with a cross-product term (subgroup \times log-transformed adiponectin levels) in the main effects model. Nondiscrete variables (body mass index, LDL and HDL cholesterol, triglycerides, log CRP, physical activity, and age) were used continuously for main effect and interaction terms. We assessed the goodness of fit of the models using the method described by Hosmer and Lemeshow⁴² and did not find any significant lack of fit. All *P* values presented are 2-tailed; *P* < .05 was considered statistically significant. All analyses were performed using SAS software, version 8.2 (SAS Institute Inc, Cary, NC).

RESULTS

Characteristics and biomarker levels of cases and controls are presented in TABLE 1. Cases had a nonsignificantly higher body mass index and were more likely to have a history of hypertension and diabetes and a family history

of MI before age 60 years, although the latter did not reach statistical significance. Cases consumed slightly less alcohol and were less physically active than controls, although these differences were not statistically significant. Cases had significantly lower mean adiponectin levels than controls (15.6 [SD, 8.5] mg/L vs 17.9 [SD, 8.8] mg/L; *P* < .001) and, as expected, significantly higher levels of total and LDL cholesterol, triglycerides, and HbA_{1c} and lower HDL cholesterol levels.

We next examined the association of adiponectin levels with selected cardiovascular risk factors among cases and controls (TABLE 2). After adjustment for age, adiponectin was significantly positively correlated with HDL cholesterol and physical activity and negatively correlated with triglyceride, CRP, and HbA_{1c} levels and body mass index. Results were similar when cases and controls were combined in this analysis.

TABLE 3 shows the estimated RRs of MI during 6 years of follow-up across quintiles of adiponectin levels at baseline. After adjustment for matched variables, participants in the highest compared with the lowest quintile of adiponectin levels had a significantly decreased risk of MI (RR, 0.39; 95% confidence interval [CI], 0.23-0.64; *P* for trend on a log scale < .001). Further adjustment for family history of MI, body mass index, alcohol consumption, physical activity, and history of diabetes and hypertension at baseline did not substantively affect this relationship (RR, 0.41; 95% CI, 0.24-0.70; *P* for log trend < .001), whereas additional adjustment for LDL and HDL cholesterol levels modestly attenuated the association (RR, 0.56; 95% CI, 0.32-0.99; *P* for log trend = .02).

We next examined the impact of potential intermediary biomarkers on the relation between adiponectin and risk of MI, by sequentially adding these markers as continuous variables to our model, and calculated the multivariable-adjusted RR associated with an increase of (continuous) log-transformed adiponectin levels by 2 units on

a log scale, which represents a doubling in adiponectin concentrations on the original scale; however, as can be seen from the FIGURE, after adjustment for LDL and HDL cholesterol, which modestly attenuated the association, addition of triglycerides, HbA_{1c}, and CRP levels did not substantively affect the results.

TABLE 4 shows the multivariable- and lipid-adjusted relative risk of MI associated with a doubling in adiponectin levels in subgroups of our population. In general, the results were similar among younger and older participants and in different strata of cardiovascular risk factors. Because our subset included only a limited number of participants with diabetes at baseline, we were not able to calculate effect estimates within this subgroup; however, exclusion of these participants did not substantively alter the results as presented in Table 4. Similarly, exclusion of participants with a parental history of MI before age 60 years did not affect the results. Results were also similar in different strata of alcohol consumption and physical activity and among those taking and not taking aspirin.

We repeated our main analysis excluding participants who developed the qualifying event during the first 2 years of follow-up and found essentially the same results (RR associated with a doubling in adiponectin levels, 0.77; 95% CI, 0.61-0.97; $P = .02$). Similarly, we found the same results when we excluded nonfasting participants from our main analysis (RR, 0.80; 95% CI, 0.61-1.06; $P = .11$) and when we excluded current smokers (RR, 0.83; 95% CI, 0.67-1.04; $P = .11$). When we stratified our analysis by type of first event, we found an inverse association between adiponectin and risk of nonfatal MI (RR, 0.73; 95% CI, 0.58-0.93; $P = .009$) but not risk of fatal CHD (RR, 0.97; 95% CI, 0.67-1.41; $P = .88$); however, the latter group included only 70 cases.

COMMENT

In this nested case-control study, we found high plasma adiponectin levels associated with lower risk of MI over a fol-

low-up period of 6 years among men without previous cardiovascular disease. This association was independent

of traditional cardiovascular risk factors that might be associated with adiponectin levels and, thus, are potential

Table 1. Baseline Characteristics of Men With Incident MI and Matched Controls During 6 Years of Follow-up*

Characteristics	Cases (n = 266)	Controls (n = 532)	P Value
Age, mean (SD), y	65.2 (8.3)	65.2 (8.3)	
Current smoker	32 (12.4)	64 (12.4)	
Body mass index, mean (SD)†	26.2 (3.5)	25.7 (3.5)	.06
Major ancestry‡			
White	251 (94.4)	503 (94.6)	.91
African American	0	2 (0.4)	.56
Asian	1 (0.4)	3 (0.6)	.99
Other	5 (1.9)	7 (1.3)	.55
Family history of MI before age 60 y	40 (15.0)	58 (10.9)	.09
Current aspirin use, ≥2/wk	103 (39.3)	185 (35.0)	.24
History of diabetes	25 (9.4)	24 (4.5)	.007
History of hypertension	112 (42.1)	164 (30.8)	.002
The metabolic syndrome§	107 (40.2)	140 (26.3)	<.001
Fat intake, mean (SD), % energy			
Total	30.9 (6.8)	30.3 (7.0)	.28
Saturated fat	10.4 (2.8)	10.1 (2.9)	.16
Alcohol consumption, median (IQR), g/d	5.6 (0.9-15.4)	7.0 (0.9-18.3)	.10
Physical activity, median (IQR), MET-h/wk	22.8 (8.5-46.3)	26.6 (11.8-49.0)	.07
Adiponectin level, mg/L			
Mean (SD)	15.6 (8.5)	17.9 (8.8)	<.001
Median (IQR)	14.3 (9.6-20.3)	16.5 (11.6-22.9)	<.001
Cholesterol level, mean (SD), mg/dL			
Total	214.7 (39.8)	205.0 (37.6)	<.001
LDL	135.6 (36.3)	126.8 (31.2)	<.001
HDL	42.1 (11.3)	45.9 (12.5)	<.001
Triglycerides level, mg/dL	181.7 (116.5)	153.9 (121.3)	.002
HbA _{1c} , median (IQR), %	5.74 (5.54-5.96)	5.64 (5.45-5.82)	<.001

Abbreviations: HbA_{1c}, hemoglobin A_{1c}; HDL, high-density lipoprotein; IQR, interquartile range; LDL, low-density lipoprotein; MET-h, metabolic equivalent task-hours; MI, myocardial infarction.

SI conversions: To convert total, HDL, and LDL cholesterol to mmol/L, multiply by 0.0259. To convert triglycerides to mmol/L, multiply by 0.0113.

*Data are expressed as No. (%) unless otherwise indicated. Age, smoking status, and month of blood draw were matched variables.

†Body mass index is expressed as weight in kilograms divided by the square of height in meters.

‡Major ancestry as reported by participants. Participants could select more than 1 major ancestry. Distribution does not sum to 100% because of missing values.

§The metabolic syndrome was defined as having 3 or more of the following 5 abnormalities: body mass index of at least 25; triglycerides level of at least 150 mg/dL (0.20 mmol/L); HDL cholesterol level of less than 40 mg/dL (1.04 mmol/L); history of hypertension; and history of diabetes, development of diabetes during follow-up, or HbA_{1c} level of at least 7% at baseline.

Table 2. Age-Adjusted Spearman Partial Correlation Coefficients Between Plasma Adiponectin Level and Selected Cardiovascular Risk Factors in the Case-Control Data Set

Cardiovascular Risk Factors	Cases (n = 266)		Controls (n = 532)	
	R	P Value	R	P Value
Total cholesterol	-0.06	.32	-0.02	.59
LDL cholesterol	-0.01	.91	-0.01	.80
HDL cholesterol	0.33	<.001	0.44	<.001
Triglycerides	-0.36	<.001	-0.39	<.001
HbA _{1c}	-0.12	.05	-0.10	.02
CRP	-0.17	.005	-0.18	<.001
Body mass index	-0.25	<.001	-.27	<.001
Physical activity	0.08	.22	0.10	.03

Abbreviations: CRP, C-reactive protein; HbA_{1c}, hemoglobin A_{1c}; HDL, high-density lipoprotein; LDL, low-density lipoprotein.

Table 3. Estimated RRs of Myocardial Infarction During 6 Years of Follow-up According to Quintile of Baseline Adiponectin Levels (n = 798)

	Quintile*					P Value for Trend†
	1	2	3	4	5	
Plasma adiponectin level, mg/L median (range)*	7.9 (2.4-10.5)	12.6 (10.6-14.5)	16.5 (14.6-18.5)	21.1 (18.6-24.8)	29.2 (24.9-56.1)	
Cases, No.	78	56	51	49	32	
Controls, No.	106	106	107	106	107	
Model, RR (95% CI)						
Adjusted for matched variables‡	1.00	0.70 (0.45-1.08)	0.63 (0.40-0.99)	0.61 (0.39-0.96)	0.39 (0.23-0.64)	<.001
Multivariable§	1.00	0.72 (0.46-1.14)	0.69 (0.43-1.09)	0.70 (0.44-1.13)	0.41 (0.24-0.70)	<.001
Multivariable, additionally adjusted for lipid levels¶	1.00	0.75 (0.47-1.20)	0.73 (0.46-1.18)	0.82 (0.50-1.33)	0.56 (0.32-0.99)	.02

Abbreviations: CI, confidence interval; RR, relative risk.

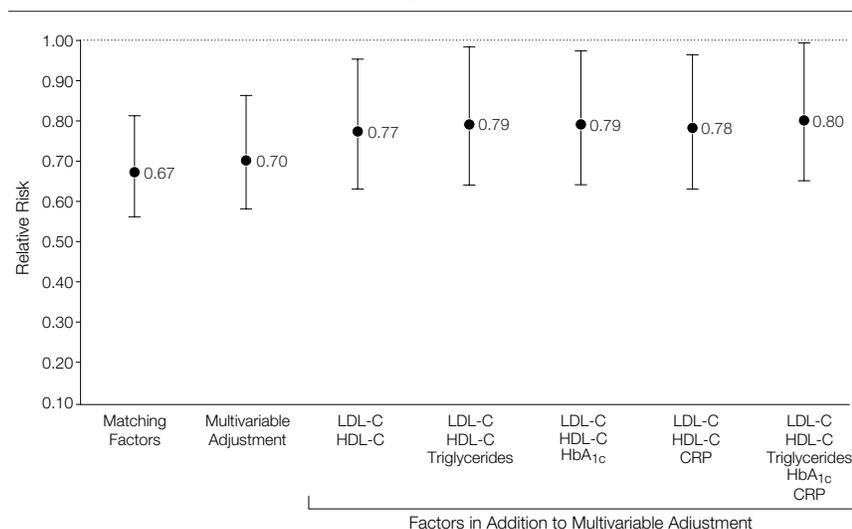
*Quintiles, medians, and ranges of adiponectin levels are based on controls only.

†P value for trend based on log-transformed adiponectin levels.

‡Adjusted for matched variables (age, smoking status, and month of blood draw).

§Adjusted for matched variables, body mass index, family history of myocardial infarction before age 60 years, history of diabetes, history of hypertension, alcohol intake, and physical activity.

¶Lipids include low- and high-density lipoprotein cholesterol.

Figure. Estimated Relative Risk of MI During 6 Years of Follow-up

Relative risk of myocardial infarction (MI) associated with a doubling of adiponectin levels after sequential adjustment for low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), triglyceride, hemoglobin A_{1c} (HbA_{1c}), and C-reactive protein (CRP) levels. Matching factors included age, smoking status, and month of blood draw. Multivariable adjustment included matching factors, body mass index, family history of MI, history of diabetes and hypertension, alcohol intake, and physical activity. Biomarkers were added to the model as continuous variables; adiponectin, triglycerides, HbA_{1c}, and CRP levels were long-transformed. Error bars indicate 95% confidence intervals.

confounders. Of note, the relationship was also independent of hypertension, diabetes, and HbA_{1c} levels, factors that were closely related to adiponectin levels in previous studies. Furthermore, the relationship was only partly explained by differences in blood lipid levels and was independent of CRP.

Our results are in line with previous cross-sectional studies of adiponectin levels and coronary artery disease.^{5,43,44} Ku-

mada et al⁴³ reported significantly lower adiponectin levels in 225 consecutive male patients aged 40 to 69 years with coronary artery disease compared with 225 voluntary blood donors and a 2.05-fold increased risk of coronary artery disease (95% CI, 1.29-4.95) comparing participants in the lowest and highest quartiles of adiponectin levels, after adjustment for diabetes, dyslipidemia, hypertension, smoking, and body mass

index. In a study by Hotta et al,⁵ adiponectin levels were lowest in participants with coronary artery disease and type 2 diabetes, intermediate in those without diabetes, and highest in non-diabetic individuals. These significant differences persisted after adjustment for other cardiovascular risk factors. Kojima et al⁴⁴ found significantly lower adiponectin levels in 34 participants with acute MI compared with 35 individuals without significant coronary artery stenosis who were matched for age, sex, and body mass index.

Among 227 hemodialysis patients, Zoccali et al⁴⁵ found in a prospective setting that after adjustment for cardiovascular risk factors, adiponectin was inversely related to cardiovascular events (hazard ratio, 0.97; 95% CI, 0.93-0.99 for an increase in adiponectin levels of 1 mg/L) over a mean follow-up of 2.5 years, although adiponectin levels did not predict overall mortality. A study by Lindsay et al⁴⁶ found no significant association between plasma adiponectin levels and risk of CHD in a nested case-control study among 372 American Indians after adjustment for other cardiovascular risk factors (odds ratio for 1-SD change in adiponectin, 0.90; *P* = .34) however, in stratified analyses, they found a significantly reduced risk among those with type 2 diabetes (comprising about 61% of their initial data set; odds ratio, 0.40; *P* = .02).

As an extension to these reports, our study is among the first to suggest that

plasma adiponectin levels may predict cardiovascular events years in advance in a population without diagnosed cardiovascular disease. In fact, several lines of evidence suggest that adiponectin may be not only a marker of cardiovascular risk but also a causal risk factor. First, adiponectin may lower the risk of cardiovascular disease by improving insulin sensitivity and blood lipid levels, as suggested by animal and human data.^{8,10-13} Adiponectin has been shown to result in activation of the adenosine monophosphate-activated protein kinase in skeletal muscle and liver, leading to phosphorylation of acetyl coenzyme A carboxylase, increased fatty acid oxidation and glucose uptake, reduced fatty acid synthesis, and reduction of molecules involved in gluconeogenesis.⁴⁷ Downstream effects may include reduced triglyceride content in liver and skeletal muscle and suppression of hepatic glucose production.^{10,11,48,49} This may also explain the finding in our study and in previous studies that adiponectin is inversely associated with triglyceride levels and positively associated with HDL cholesterol levels.¹⁴⁻¹⁶

Second, adiponectin may lower CRP and other inflammatory cardiovascular risk factors.^{12,50} Thus, injection of an adiponectin-producing adenovirus was shown to reverse the significantly increased adipose tissue tumor necrosis factor α (TNF- α) messenger RNA and plasma TNF- α levels in adiponectin knockout mice.¹² In line with these animal experiments, human studies found inverse associations between adiponectin and the inflammatory markers TNF- α , interleukin 6, and CRP.⁵¹⁻⁵⁵

Third, adiponectin may modulate the vascular response to lipid and inflammatory stimuli. Thus, adiponectin inhibits endothelial nuclear transcription factor NF- κ B signaling, which mediates the effects of TNF- α and other cytokines.²⁰

Furthermore, adiponectin suppresses lipid accumulation and class A scavenger receptor expression in macrophages and, consequently, the transformation of macrophages to foam cells,

which plays an important role in the atherogenic process.²¹ It was shown that adiponectin binds to subendothelial collagens and suppresses proliferation and migration of human aortic smooth muscle cells.^{22,56} In apolipoprotein E-deficient mice, adiponectin significantly reduced the development of atherosclerosis that usually occurs in these animals.^{57,58} Consistent with these animal studies, low adiponectin levels were

found in participants with coronary artery disease.^{5,43,45}

Our study has some limitations. The inverse relationship between plasma adiponectin levels and risk of MI was not linear but, rather, plateaued in the middle quintiles and was most pronounced in the highest quintile; however, we used rather conservative methods to estimate the RRs, which makes it unlikely that our results are driven by outliers.

Table 4. Multivariable-Adjusted Estimated RRs of Myocardial Infarction During 6 Years of Follow-up Associated With a Doubling in Adiponectin Level by Subgroup*

Subgroup	Cases	Controls	RR (95% CI)	P Value	P Value for Interaction†
Age, y‡					
<65	123	245	0.72 (0.52-0.99)	.04	.14
≥65	143	286	0.77 (0.57-1.05)	.09	
Hypertension§					
No	154	368	0.66 (0.50-0.88)	.005	.09
Yes	112	163	0.95 (0.67-1.34)	.77	
Overweight					
No (BMI <25)	98	240	0.68 (0.46-1.00)	.05	.24
Yes (BMI ≥25)	168	291	0.77 (0.60-1.00)	.05	
The metabolic syndrome					
No	159	392	0.76 (0.58-1.01)	.05	.84
Yes	107	139	0.80 (0.56-1.14)	.21	
Aspirin use					
No	163	347	0.77 (0.59-1.02)	.07	.51
Yes	103	185	0.75 (0.52-1.09)	.14	
LDL cholesterol level, mg/dL					
<130	115	305	0.74 (0.54-1.03)	.07	.50
≥130	151	226	0.76 (0.56-1.03)	.07	
HDL cholesterol level, mg/dL					
<40	125	185	0.68 (0.49-0.94)	.02	.10
≥40	141	346	0.84 (0.62-1.14)	.26	
Triglyceride level, mg/dL					
<150	129	342	0.79 (0.59-1.07)	.13	.80
≥150	137	189	0.75 (0.55-1.04)	.08	
CRP level, mg/L‡					
<1.08	94	264	0.86 (0.60-1.25)	.44	.73
≥1.08	172	268	0.70 (0.54-0.93)	.01	
Physical activity, MET-h/wk‡					
<26.6	148	265	0.71 (0.53-0.96)	.02	.62
≥26.6	118	266	0.88 (0.63-1.24)	.47	
Alcohol intake§					
Nondrinkers	65	122	0.89 (0.57-1.39)	.60	.69
Moderate drinkers	169	327	0.81 (0.62-1.07)	.14	

Abbreviations: BMI, body mass index (expressed as weight in kilograms divided by the square of height in meters); CI, confidence interval; CRP, C-reactive protein; HDL, high-density lipoprotein; MET-h, metabolic equivalent task-hours; LDL, low-density lipoprotein; RR, relative risk.

SI conversions: To convert HDL and LDL cholesterol to mmol/L, multiply by 0.0259. To convert triglycerides to mmol/L, multiply by 0.0113.

*Effect estimates and P values were calculated using log-transformed adiponectin level as a continuous variable, adjusted for matched variables (age, smoking status, and month of blood draw), body mass index, family history of myocardial infarction, history of diabetes, history of hypertension, alcohol intake, physical activity, and LDL and HDL cholesterol levels.

†P values for interaction were calculated using continuous or dichotomous variables, where applicable, and log-transformed adiponectin level.

‡Indicates the median of the corresponding value for controls.

§Not adjusted for the stratification variable.

||Moderate drinkers includes participants who drank 0.1-29.9 g/d of alcohol. Heavy drinkers (≥30 g/d) were excluded from this subgroup analysis.

Furthermore, this type of nonlinear relationship is consistent with other reports on adiponectin and risk of CHD and type 2 diabetes.^{13,43} A single assessment of adiponectin may be susceptible to short-term variation, which would bias the results toward the null. However, in a previous analysis, we found intraindividual adiponectin levels to be reasonably stable over time, with an intraclass correlation coefficient of 0.85 for adiponectin levels measured within participants 1 year apart.³⁴ It is currently unknown whether long-term storage of blood samples affects plasma adiponectin levels; however, if anything, this would tend to increase measurement error and, thus, bias the results toward the null. Because the ranges of anthropometric parameters in the present study were quite broad, the biological relationships found should be generalizable. Future studies should address whether our findings also apply to women and individuals with different racial/ethnic origins and socioeconomic status and whether adiponectin levels predict cardiovascular events beyond a follow-up period of 6 years. It is possible that the observed association is confounded by a yet-to-be-determined factor; however, we adjusted our analysis for established and novel cardiovascular risk factors. Furthermore, although the results failed to reach formal statistical significance in certain subgroups, possibly because of limited sample size, we found comparable and robust risk reductions in various low and high cardiovascular risk groups. While we excluded participants with diagnosed cardiovascular disease at baseline, we cannot exclude the possibility that participants had undiagnosed atherosclerosis. However, we found similar results when we excluded participants who developed MI during the first 2 years of follow-up. Not all participants in our data set provided fasting blood samples, which could affect the measurement of triglyceride levels; however, any potential misclassification should be nondifferential. Furthermore, results were similar when we restricted our analysis to fasting participants. Finally, LDL cholesterol levels in

our study were measured directly,³⁶ without using the Friedewald formula, and, thus, do not depend on fasting status. We used body mass index instead of waist-hip ratio to assess abdominal obesity, which may lead to misclassification of the metabolic syndrome; however, the distribution of metabolic abnormalities was similar to that recently reported for the general population,⁴¹ and the metabolic syndrome was used for secondary analyses only. Since plasma insulin levels were not available in our data set, we were unable to examine the impact of this potentially intermediary variable; however, adjustment for HbA_{1c} as a marker of glycemic control did not appreciably change the results. It should be noted that some of the variables that we adjusted for may be causal mediators instead of confounders, which would underestimate the true relationship between adiponectin levels and risk of MI.

In conclusion, we found that high plasma adiponectin levels are associated with a lower risk of MI over a follow-up period of 6 years among men without previous cardiovascular disease, independent of traditional cardiovascular disease risk factors. Particularly, this relationship can only partly be explained by differences in blood lipids and is independent of inflammation and glycemic status at baseline. The effect of adiponectin on risk of CHD merits further study.

Author Contributions: Dr Pischon had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Study concept and design: Pischon, Girman, Hotamisligil, Rimm.

Acquisition of data: Rimm.

Analysis and interpretation of data: Pischon, Girman, Hotamisligil, Rifai, Hu, Rimm.

Drafting of the manuscript: Pischon, Girman.

Critical revision of the manuscript for important intellectual content: Pischon, Girman, Hotamisligil, Rifai, Hu, Rimm.

Statistical expertise: Pischon, Girman, Hu.

Obtained funding: Girman, Rimm.

Administrative, technical, or material support: Girman, Rimm.

Supervision: Hotamisligil, Rimm.

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