

Inflammation and the Incidence of Type 2 Diabetes

The Multi-Ethnic Study of Atherosclerosis (MESA)

ALAIN G. BERTONI, MD, MPH¹
 GREGORY L. BURKE, MD, MSC^{1,2}
 JAMES A. OWUSU, MD¹
 MERCEDES R. CARNETHON, PHD³
 DHANANJAY VAIDYA, PHD⁴

R. GRAHAM BARR, MD, DRPH⁵
 NANCY S. JENNY, PHD⁶
 PAMELA OUYANG, MBBS⁴
 JEROME I. ROTTER, MD⁷

OBJECTIVE — Many studies have documented associations between inflammation and type 2 diabetes incidence. We assessed potential variability in this association in the major U.S. racial/ethnic groups.

RESEARCH DESIGN AND METHODS — Incident type 2 diabetes was assessed among men and women aged 45–84 years without prior clinical cardiovascular disease or diabetes in the prospective Multi-Ethnic Study of Atherosclerosis. Interleukin (IL)-6, fibrinogen, and C-reactive protein (CRP) were measured at baseline (2000–2002); fasting glucose and diabetes medication use was assessed at baseline and three subsequent in-person exams through 2007. Type 2 diabetes was defined as use of diabetes drugs or glucose ≥ 126 mg/dl. Covariates included baseline demographics, clinic, smoking, alcohol, exercise, hypertension medication, systolic blood pressure, insulin resistance, and BMI. Cox proportional hazards regression was used to calculate hazard ratios (HRs) by quartiles of CRP, IL-6, and fibrinogen.

RESULTS — Among 5,571 participants (mean age 61.6 years, 53% female, 42.1% white, 11.5% Chinese, 25.7% black, and 20.7% Hispanic), 410 developed incident diabetes during a median follow-up time of 4.7 years (incidence 16.8 per 1,000 person-years). CRP, IL-6, and fibrinogen levels were associated with incident diabetes in the entire sample. After adjustment, the associations were attenuated; however, quartile 4 (versus quartile 1) of IL-6 (HR 1.5 [95% CI 1.1–2.2]) and CRP (1.7 [1.3–2.4]) remained associated with incident diabetes. In stratified analyses, similar associations were observed among white, black, and Hispanic participants.

CONCLUSIONS — Higher levels of inflammation predict short-term incidence of type 2 diabetes in a multiethnic American sample.

Diabetes Care 33:804–810, 2010

A number of prospective studies (1–8) have demonstrated an association between high levels of inflammation and the development of type 2 diabetes. In this report, we assess three inflammatory markers: C-reactive protein (CRP), interleukin (IL)-6, and fibrinogen. CRP is an acute-phase reactant mainly

produced in the liver. Recent studies have shown that CRP can also be produced by fat cells (9), which raises the possibility that CRP may simply be a marker of obesity in people who go on to develop diabetes. Fibrinogen is involved in clotting but is also an acute-phase reactant and has been previously linked to incident diabe-

tes (8). IL-6 is made by leukocytes and other tissues that play a role in glucose homeostasis, including pancreatic islet cells, hepatocytes, adipocytes, and skeletal muscle cells, and is associated with incident diabetes (10). The graded positive association between most inflammatory markers and diabetes incidence remains significant following adjustment for established diabetes risk factors. However, a study (11) in the Pima population showed no association between inflammatory markers and the risk of diabetes after adjusting for established risk factors for diabetes. Although a sizable number of studies have documented the inflammatory marker–diabetes association, studies on ethnic/racial variations in this association are limited, despite the well-documented increased prevalence of diabetes in nonwhite populations in the U.S. (12). The aims of this analysis are 1) to explore the ability of CRP, IL-6, and fibrinogen to predict the incidence of diabetes in a prospective, multiethnic cohort and 2) to determine the extent to which observed associations are similar across racial/ethnic groups. We also consider whether observed associations are independent of the major known risk factors for diabetes (obesity, family history, insulin resistance, hypertension, age, and physical inactivity) (13).

RESEARCH DESIGN AND METHODS

The Multi-Ethnic Study of Atherosclerosis (MESA) is a population-based sample of 6,814 men and women from four ethnic groups (38% white, 28% African American, 22% Hispanic, and 12% Chinese) aged 45–84 years without clinical cardiovascular disease (CVD) prior to recruitment. Details regarding MESA's design and objectives have been published (14). Briefly, subjects who reported a medical history of heart attack, angina, coronary revascularization, pacemaker or defibrillator implantation, valve replacement, heart failure, or cerebrovascular disease were excluded. During the baseline exam (2000–2002), standardized questionnaires and calibrated devices were uti-

From the ¹Division of Public Health Sciences, Wake Forest University Health Sciences, Winston-Salem, North Carolina; the ²Department of Epidemiology and Prevention, Wake Forest University Health Sciences, Winston-Salem, North Carolina; the ³Department of Preventive Medicine, Northwestern University, Chicago, Illinois; the ⁴Department of Medicine, Johns Hopkins University, Baltimore, Maryland; the ⁵Departments of Medicine and Epidemiology, Columbia University Medical Center, New York, New York; the ⁶Department of Pathology, University of Vermont College of Medicine, Burlington, Vermont; and the ⁷Department of Medicine, Cedars Sinai Medical Center, Los Angeles, California.

Corresponding author: A.G. Bertoni, abertoni@wfulmc.edu.

Received 8 September 2009 and accepted 16 January 2010. Published ahead of print at <http://care.diabetesjournals.org> on 22 January 2010. DOI: 10.2337/dc09-1679.

© 2010 by the American Diabetes Association. Readers may use this article as long as the work is properly cited, the use is educational and not for profit, and the work is not altered. See <http://creativecommons.org/licenses/by-nc-nd/3.0/> for details.

The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked "advertisement" in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

lized to obtain demographic data, tobacco usage, medical conditions, current prescription medication usage, weight, waist circumference, and height. BMI was calculated as weight (kilograms)/height (meters)². Resting seated blood pressure was measured three times using a Dinamap automated oscillometric sphygmomanometer (model Pro 100; Critikon, Tampa, FL); the last two measurements were averaged for analysis. Hypertension was defined based on use of an antihypertensive medication or systolic blood pressure ≥ 140 mmHg or diastolic blood pressure ≥ 90 mmHg. Education was categorized as less than high school, completed high school, some college/technical school certificate or associate degree, bachelor's degree, or graduate or professional school. We utilized the MESA Typical Week Physical Activity Survey intentional exercise variable, defined as the sum of walking for exercise, sports/dancing, and conditioning metabolic equivalent (MET) hours per week (14). Alcohol use was queried and coded as current/former/never. Participants were asked about current or former cigarette smoking, age at which smoking started and ended, and number of daily cigarettes typically consumed. Pack-years was calculated, defined as number years smoking times packs per day (cigarettes per day divided by 20). Family history of diabetes was assessed at exam 2 and was unavailable for those who missed that exam.

Laboratory assessment

Fasting blood samples were drawn and processed using a standardized protocol and sent for measurement of glucose and inflammatory markers to central laboratories (15). Serum glucose was measured by the glucose oxidase method on the Vitros analyzer (Johnson & Johnson Clinical Diagnostics). Insulin was determined by a radioimmunoassay method using the Linco Human Insulin-Specific RIA Kit (Linco Research). The homeostasis model assessment of insulin resistance (HOMA-IR) was calculated as insulin mU/l \times (glucose mg/dl \times 0.055)/22.5 (16). CRP and fibrinogen were measured using the BNII nephelometer (N High-Sensitivity CRP and N Antiserum to Human Fibrinogen; Dade Behring). Intra-assay coefficients of variation (CVs) for CRP range from 2.3 to 4.4% and interassay CVs range from 2.1 to 5.7%. The intra-assay and interassay CVs for fibrinogen were 2.7 and 2.6%, respectively. IL-6 was measured by ultra-

sensitive enzyme-linked immunosorbent assay (Quantikine HS Human IL-6 Immunoassay; R&D Systems). The laboratory CV for the IL-6 assay was 6.3%.

Diabetes definition

Participants with diabetes at baseline, defined as using hypoglycemic drugs or if the fasting glucose was ≥ 7.0 mmol/l (126 mg/dl), were excluded from these analyses. Subjects were considered to have impaired fasting glucose if they did not have diabetes by the preceding criteria and their fasting glucose was ≥ 5.6 and < 7.0 mmol/l (> 100 and < 126 mg/dl). Subjects newly utilizing hypoglycemic medication or having fasting glucose ≥ 7 mmol/l at exams 2, 3, or 4 were considered to have incident type 2 diabetes.

Statistical analysis

Descriptive statistics were utilized to compare characteristics of participants who developed diabetes compared with those who did not. We primarily analyzed these data by dividing the population into quartiles of each inflammatory variable. We also noted that the distributions of IL-6 and CRP were skewed and differed by race (generally lower levels in Chinese and higher in black participants). Thus, we also assessed the relationship between log-transformed IL-6 and CRP and incident diabetes separately in each racial/ethnic group.

Unadjusted incidence rates were calculated using person-time analysis. Participants were censored at the last follow-up exam that they attended (exams 2, 3, or 4). Cox proportional hazards modeling was utilized to estimate hazard ratios (HRs) associated across quartiles. Model 1 included the inflammation marker of interest, age, sex, race/ethnicity site, exercise, education, systolic blood pressure, antihypertensive medication use, alcohol consumption, and smoking. Model 2 included model 1 variables plus HOMA-IR. Model 3 included model 2 variables plus BMI. All covariates were defined as their value at the baseline visit. Potential interactions between each inflammatory variable and race/ethnicity or sex were assessed using a multivariable interaction term. We performed additional analyses, first replacing BMI with waist circumference, and second adding family history of diabetes to model 3, which did not appreciably change our results; therefore, we do not present them. Finally, we constructed a composite inflammation score by standardizing the

distributions of IL-6 and CRP and averaging the two. The sample was then divided into quartiles according to the composite score. Incidence rates were calculated and hazard ratios modeled as detailed above. All analyses were performed using Stata 8.2 (Statacorp, College Station, TX).

RESULTS— After excluding 857 (12.5%) participants with diabetes at baseline, 349 (5.1%) without a follow-up visit, 25 (0.4%) missing exam 1 diabetes status, and 12 (0.2%) missing all inflammatory markers at baseline, our final study sample was 5,571 individuals. Of these, 410 were classified as having incident diabetes during a median follow-up time of 4.7 years (incidence rate 16.5 per 1,000 person-years). Participant characteristics, stratified by race/ethnicity, are presented in Table 1. Overall, diabetes risk factors differed significantly between racial/ethnic groups. Nonwhite participants had a greater prevalence of impaired fasting glucose at baseline and higher incident diabetes rates than white subjects. Chinese participants had, on average, a lower BMI and waist circumference and less frequently reported a family history of diabetes. Levels of all three inflammation markers differed significantly by race/ethnicity ($P < 0.001$). Mean IL-6, CRP, and fibrinogen were lower among Chinese and tended to be higher among Hispanic and black subjects when compared with white subjects.

There was a modest level of correlation between pairs of inflammatory markers and between each marker and BMI (online appendix Table [available at <http://care.diabetesjournals.org/cgi/content/full/dc09-1679/DC1>]). There was little to no correlation between CRP/IL-6/fibrinogen and age, blood pressure, glucose, exercise, and pack-years smoking. For each marker, rates of incident diabetes were higher for participants in higher quartiles compared with the lowest quartile (Table 2). Diabetes incidence was threefold higher among participants whose baseline levels of CRP or IL-6 were in the fourth versus the first quartile. The relationship between fibrinogen and incidence appeared different, with only the fourth quartile being significantly elevated compared with the lower three quartiles. The univariate and adjusted associations between inflammation and diabetes are shown in Table 2. In general, all the observed associations remained significant after adjusting for demographic factors, health behaviors, and blood pres-

Table 1—Characteristics of participants, by race/ethnicity, in MESA 2000–2007

	White	Chinese	African American	Hispanic
n	2,348	644	1,427	1,152
Age (years)	62.3 ± 10.2	61.3 ± 10.2	61.5 ± 10.2	60.5 ± 10.3
Female*	52.4%	51.7%	55.6%	52.6%
Current smoking	11.1%	5.4%	18.1%	13.7%
Pack-years (tobacco)	14.6 ± 27.3	4.3 ± 12.7	11.4 ± 18.9	7.4 ± 16.0
Former alcohol use	17.1%	15.5%	31.0%	24.3%
Current alcohol use	73.6%	32.2%	52.3%	51.1%
Education (more than a bachelor's degree)	51.4	40.9	36.3	11.0
Exercise (MET hours/week)	28.4 ± 37.3	19.7 ± 26.5	29.9 ± 48.4	22.8 ± 35.5
Weight (lb)	173.7 ± 36.4	138.3 ± 24.3	186.6 ± 37.4	168.7 ± 32.1
BMI (kg/m ²)	27.5 ± 4.9	23.9 ± 3.3	29.8 ± 5.8	29.1 ± 4.8
Waist (cm)	97.2 ± 14.1	86.6 ± 10.1	99.8 ± 14.2	99.5 ± 12.7
Hypertension medication	30.7%	24.5%	44.6%	28.0%
Hypertension	36.6%	34.2%	54.7%	37.3%
Systolic blood pressure (mmHg)	122.6 ± 19.9	122.8 ± 21.1	130.6 ± 21.2	125.2 ± 21.3
Diastolic blood pressure (mmHg)	70.2 ± 9.9	71.9 ± 10.3	74.6 ± 10.1	71.7 ± 10.1
Glucose (mg/dl)	87.8 ± 10.0	91.5 ± 10.1	90.2 ± 10.7	91.0 ± 10.8
Impaired fasting glucose	10.4%	17.2%	15.9%	17.3%
HOMA-IR	1.3 ± 1.0	1.4 ± 1.0	1.5 ± 1.1	1.7 ± 1.3
Family history†	29.1%	25.6%	42.3%	41.0%
CRP (mg/l)	3.3 ± 5.0	1.9 ± 5.2	4.5 ± 6.9	3.7 ± 4.7
CRP ≥3.0 mg/l	32.6%	12.3%	43.2%	40.9%
IL-6 (pg/ml)	1.4 ± 1.2	1.1 ± 1.0	1.6 ± 1.2	1.6 ± 1.1
Fibrinogen (mg/dl)	332.4 ± 68.7	327.6 ± 60.2	357.3 ± 77.4	354.0 ± 70.5
Incident diabetes‡	11.1/1,000	16.2/1,000	21.6/1,000	21.9/1,000

Data are means ± SD, unless otherwise indicated. All differences across race/ethnicity are significant at *P* < 0.001 except **P* = 0.2. †Family history data available for 5,404 subjects. ‡Rate per 1,000 person-years.

sure variables. For fibrinogen, only the fourth quartile was significantly associated with diabetes, and after adjusting for HOMA there was no longer a linear trend; with subsequent addition of BMI to the model, there was no evidence for any association between level of fibrinogen and diabetes. In contrast, while addition of HOMA and BMI both attenuated the HRs, there re-

Table 2—HRs for incident diabetes associated with inflammatory markers in MESA 2002–2007

	Quartile 1	Quartile 2	Quartile 3	Quartile 4	<i>P</i> trend
<i>n</i> = 5,554					
CRP (mg/l)	0.15–0.80	0.81–1.80	1.81–4.08	4.09–97.4	
Rate	9.0	13.4	16.7	27.4	<0.001
Univariate	1 (referent)	1.5 (1.1–2.1)	1.9 (1.3–2.6)	3.1 (2.3–4.1)	<0.001
Model 1	1 (referent)	1.5 (1.3–2.0)	1.8 (1.3–2.4)	2.8 (2.1–3.9)	<0.001
Model 2	1 (referent)	1.3 (0.9–1.8)	1.5 (1.0–2.0)	2.1 (1.5–2.9)	<0.001
Model 3	1 (referent)	1.2 (0.9–1.7)	1.3 (0.9–1.8)	1.7 (1.3–2.4)	<0.01
<i>n</i> = 5,445					
IL-6 (pg/ml)	0.12–0.73	0.74–1.14	1.15–1.792	1.793–12.9	
Rate	8.2	14.9	15.7	28.2	<0.001
Univariate	1 (referent)	1.8 (1.3–3.5)	1.9 (1.4–2.7)	3.4 (2.5–4.6)	<0.001
Model 1	1 (referent)	1.7 (1.2–2.4)	1.7 (1.2–2.4)	3.0 (2.2–4.2)	<0.001
Model 2	1 (referent)	1.5 (1.1–2.1)	1.3 (0.9–1.8)	2.0 (1.4–2.8)	<0.001
Model 3	1 (referent)	1.4 (1.0–2.0)	1.1 (0.8–1.6)	1.5 (1.1–2.2)	0.02
<i>n</i> = 5,557					
Fibrinogen (mg/l)	144–293	294–334	335–384	385–701	
Rate	13.9	13.3	16.4	22.5	<0.001
Univariate	1 (referent)	1.0 (0.7–1.3)	1.2 (0.9–1.6)	1.6 (1.2–2.1)	<0.001
Model 1	1 (referent)	1.0 (0.7–1.3)	1.1 (0.8–1.5)	1.5 (1.1–2.0)	0.01
Model 2	1 (referent)	0.9 (0.7–1.2)	1.1 (0.8–1.4)	1.2 (0.9–1.6)	NS
Model 3	1 (referent)	0.8 (0.6–1.1)	1.0 (0.7–1.3)	1.0 (0.8–1.3)	NS

Data are HRs (95% CI). Rate is defined as the rate of incident diabetes per 1,000 person-years. Model 1 is adjusted for age, sex, race, education, site, alcohol, smoking, exercise, systolic blood pressure, and antihypertensive medication use. Model 2 = model 1 plus HOMA-IR. Model 3 = model 2 plus BMI. *P* value for trend across quartiles is noted. NS, not significant.

Table 3—HRs for incident diabetes associated with inflammatory marker quartiles, stratified by race/ethnicity, in MESA 2002–2007

	Quartile 1	Quartile 2	Quartile 3	Quartile 4	P trend
CRP					
White					
Rate	5.5	10.4	10.5	18.8	<0.001
Model 1	1 (referent)	1.8 (0.9–3.3)	1.8 (1.0–3.5)	3.2 (1.8–5.8)	<0.01
Model 2	1 (referent)	1.4 (0.8–2.6)	1.3 (0.7–2.4)	2.0 (1.1–3.6)	NS
Model 3	1 (referent)	1.3 (0.7–2.4)	1.0 (0.5–2.0)	1.5 (0.8–2.9)	NS
Chinese					
Rate	12.9	14	24.3	28.6	NS
Model 1	1 (referent)	1.0 (0.5–2.0)	1.6 (0.8–3.3)	2.0 (0.7–5.8)	NS
Model 2	1 (referent)	0.8 (0.4–1.8)	1.4 (0.6–2.9)	1.0 (0.3–3.6)	NS
Model 3	1 (referent)	0.7 (0.3–1.5)	1.0 (0.5–2.2)	0.9 (0.3–3.0)	NS
African American					
Rate	12.8	14.7	20.1	33.2	<0.001
Model 1	1 (referent)	1.3 (0.6–2.5)	1.7 (0.9–3.1)	2.6 (1.5–4.7)	<0.01
Model 2	1 (referent)	1.2 (0.6–2.3)	1.6 (0.8–2.9)	2.2 (1.2–4.0)	<0.05
Model 3	1 (referent)	1.1 (0.6–2.2)	1.5 (0.8–2.7)	1.9 (1.0–3.5)	NS
Hispanic					
Rate	9.8	17.5	21.2	34.4	<0.001
Model 1	1 (referent)	1.8 (0.9–4.0)	2.1 (1.0–4.5)	3.5 (1.7–7.2)	<0.01
Model 2	1 (referent)	1.7 (0.8–3.7)	1.7 (0.8–3.6)	2.7 (1.3–5.5)	<0.05
Model 3	1 (referent)	1.7 (0.8–3.8)	1.7 (0.8–3.5)	2.5 (1.2–5.1)	NS
IL-6					
White					
Rate	5.1	9.1	12.2	20.5	<0.001
Model 1	1 (referent)	1.7 (0.9–3.4)	2.2 (1.2–4.2)	3.6 (1.9–6.7)	<0.001
Model 2	1 (referent)	1.2 (0.6–2.3)	1.4 (0.7–2.7)	1.8 (0.6–3.5)	NS
Model 3	1 (referent)	1.0 (0.5–2.0)	1.1 (0.6–2.2)	1.3 (0.7–2.6)	NS
Chinese					
Rate	9.7	23.4	18.2	22.3	NS
Model 1	1 (referent)	2.3 (1.1–4.7)	1.6 (0.7–4.1)	2.4 (0.9–5.8)	NS
Model 2	1 (referent)	2.1 (1.0–4.3)	1.0 (0.4–2.7)	1.9 (0.8–4.9)	NS
Model 3	1 (referent)	1.8 (0.9–3.7)	0.8 (0.3–2.1)	1.4 (0.5–3.7)	NS
African American					
Rate	11.0	18.0	18.6	34.8	<0.001
Model 1	1 (referent)	1.5 (0.8–3.2)	1.6 (0.8–3.1)	3.0 (1.6–5.6)	<0.001
Model 2	1 (referent)	1.4 (0.7–2.7)	1.4 (0.7–2.7)	2.3 (1.2–4.4)	0.01
Model 3	1 (referent)	1.3 (0.7–2.6)	1.2 (0.6–2.4)	1.9 (1.0–3.7)	NS
Hispanic					
Rate	12.8	18.8	17.9	34.1	<0.01
Model 1	1 (referent)	1.4 (0.7–2.9)	1.3 (0.6–2.6)	2.4 (1.2–4.6)	0.02
Model 2	1 (referent)	1.2 (0.6–2.5)	0.7 (0.4–1.5)	1.2 (0.6–2.4)	NS
Model 3	1 (referent)	1.2 (0.6–2.5)	0.7 (0.3–1.4)	1.1 (0.5–2.2)	NS

Data are HRs (95% CI). Rate is defined as the rate per 1,000 person-years. Model 1 adjusted for age, sex, race, education, site, alcohol, smoking, exercise, systolic blood pressure, and antihypertensive medication use. Model 2 = model 1 plus HOMA-IR. Model 3 = model 2 plus BMI. P value for trend across quartiles is noted. NS, not significant.

mained a significant association between both CRP and IL-6 and diabetes incidence.

There was no statistical evidence of a significant interaction between inflammation level and race/ethnicity (or sex). Nonetheless, given our primary study aim, we performed stratified analyses for CRP and IL-6 levels (Table 3). In each racial/ethnic group, participants with higher levels of IL-6 or CRP had a higher diabetes incidence than those with the lowest levels. Additionally, in a given

quartile of CRP, IL-6 incidence rates tended to be higher in each nonwhite racial/ethnic group compared with white subjects. CRP quartile was significantly associated with incident diabetes after adjustment for model 1 variables in white, African American, and Hispanic subjects. Adjustment for HOMA, and subsequently BMI, attenuated the associations; however, CRP quartile 4 remained associated with incident diabetes after full adjustment in African American and Hispanic

participants. IL-6 was associated with incident diabetes after adjustment for model 1 variables in all groups except Chinese. As with CRP, the magnitude of the associations were attenuated after addition of HOMA to the models; subsequent addition of BMI suggested that there was no longer an independent association between IL-6 and diabetes in any group.

Similar results were obtained when repeating analyses using log-transformed inflammation levels. Specifically, the risk

Table 4—Association between a composite score composed of IL-6 and CRP and incident diabetes in MESA 2002–2007

Sample	Quartile				P trend
	1	2	3	4	
Overall (n = 5,437)					
Rate	8.6	12.1	16.7	28.5	<0.001
Model 1	1 (referent)	1.5 (1.1–2.1)	1.8 (1.3–2.5)	3.0 (2.2–4.2)	<0.001
Model 2	1 (referent)	1.3 (0.9–1.8)	1.4 (1.0–1.9)	2.0 (1.4–2.8)	<0.001
Model 3	1 (referent)	1.2 (0.8–1.4)	1.2 (0.8–1.7)	1.6 (1.1–2.2)	0.03
White (n = 2,311)					
Rate	5.9	7.2	11.0	22.3	
Model 1	1 (referent)	1.2 (0.6–2.4)	1.8 (1.0–3.4)	3.7 (2.0–6.6)	<0.001
Model 2	1 (referent)	0.8 (0.4–1.6)	1.2 (0.6–2.2)	1.8 (1.0–3.3)	0.02
Model 3	1 (referent)	0.7 (0.4–1.4)	0.9 (0.5–1.8)	1.3 (0.7–2.5)	NS
Chinese (n = 633)					
Rate	11.1	23.8	14.2	25.6	NS
Model 1	1 (referent)	2.1 (1.0–4.1)	1.1 (0.4–3.0)	2.2 (0.9–3.0)	NS
Model 2	1 (referent)	1.6 (0.8–3.3)	0.9 (0.4–2.5)	1.4 (0.5–3.8)	NS
Model 3	1 (referent)	1.4 (0.7–2.8)	0.6 (0.2–1.7)	1.1 (0.4–3.0)	NS
African American (n = 1,376)					
Rate	9.7	13.6	23.3	33.5	<0.001
Model 1	1 (referent)	1.5 (0.7–3.1)	2.4 (1.2–4.8)	3.5 (1.8–6.8)	<0.001
Model 2	1 (referent)	1.4 (0.7–3.0)	2.1 (1.1–4.2)	2.7 (1.4–5.3)	0.01
Model 3	1 (referent)	1.3 (0.6–2.8)	1.9 (1.0–3.9)	2.2 (1.1–4.5)	NS
Hispanic (n = 1,117)					
Rate	12.0	18.6	19.8	32.7	<0.01
Model 1	1 (referent)	1.5 (0.7–3.0)	1.5 (0.7–3.1)	2.5 (1.3–4.9)	0.03
Model 2	1 (referent)	1.1 (0.5–2.2)	0.9 (0.4–1.9)	1.4 (0.7–2.9)	NS
Model 3	1 (referent)	1.0 (0.5–2.2)	0.9 (0.4–1.8)	1.3 (0.6–2.6)	NS

Data are HRs (95% CI). Rate is defined as incidence rate per 1,000 person-years. Model 1 adjusted for age, sex, race, education, site, alcohol, smoking, exercise, systolic blood pressure, and antihypertensive medication use. Model 2 = model 1 plus HOMA-IR. Model 3 = model 2 plus BMI. P value for trend across quartiles is noted. NS, not significant.

of diabetes per one-unit change in log-transformed CRP in the entire sample was 1.17 (95% CI 1.06–1.29) after full adjustment; for IL-6, it was 1.30 (1.09–1.55) and fibrinogen 1.10 (0.65–1.84). In stratified analyses, inflammation was similarly related to diabetes incidence in each racial/ethnic group, again, except in Chinese, in whom generally no statistically significant associations were observed (data not shown).

The composite score, constructed from the distribution of both IL-6 and CRP, demonstrated similar patterns as seen above (Table 4). In the entire sample, the rate of diabetes increased with each quartile (P for trend <0.001). The HRs were attenuated after adjusting for HOMA and BMI, but participants in quartile 4 remained with a 60% higher risk of incident diabetes compared with those with the least inflammation. In the race-ethnicity-stratified analyses, similar patterns were observed, although for Chinese participants, none of the trends were significant.

CONCLUSIONS— Our results suggest that in a multiethnic sample of Amer-

ican adults aged 45–84 years and free of clinical CVD at baseline, people with higher levels of CRP, IL-6, and fibrinogen have a higher incidence of diabetes over a 2- to 5-year interval. The observed patterns of association were largely consistent among the four racial/ethnic groups examined. The association between inflammation and incident diabetes was more consistent for CRP and IL-6; overall, and in each subgroup examined, rates of incident diabetes were two- to threefold higher for participants with levels in the highest quartile of this sample. The increased risk associated with higher levels of these two inflammatory markers was largely explained by established diabetes risk factors and in particular BMI. Consideration of both IL-6 and CRP distribution jointly showed similar patterns of association as with either marker alone. While fibrinogen level was also associated with incident diabetes, after adjustment for demographics, education, smoking, alcohol, exercise, hypertension, and HOMA-IR, there was no longer a significant association; further adjustment for BMI resulted in

no suggestion of an independent association.

Strengths of our analyses include utilizing a multiethnic, sex-balanced, and prospective cohort study to assess incident diabetes. MESA participants were carefully characterized with respect to standard diabetes risk factors, including fasting insulin. Rather than rely on self-reported diabetes at follow-up, MESA measured glucose and assessed medication use at the three follow-up examinations. Furthermore, all blood samples were analyzed centrally at one laboratory. There are several limitations to our analyses, however. By design, MESA excluded subjects with prevalent CVD, potentially limiting the generalizability of these results. We may have included some individuals with diet-controlled diabetes in the at-risk population, as we did not consider subjects with a self-report of diabetes who were not taking medicine and had normal fasting glucose to have diabetes. On the other hand, we used the same definition at follow-up, which should be a conservative bias (potentially underestimating the true incidence rate). The fail-

ure to find significant interactions by race/ethnicity may reflect inadequate power. We did not have adequate sample size to present analyses stratified by race/ethnicity and sex. Biomarkers were only measured once, and intraindividual variability cannot be accounted for. However, assay variability would be expected to bias findings toward the null so the observed associations are potentially underestimations.

Our findings among Chinese participants merit some discussion. The apparently different relationships between inflammation and incident diabetes may be solely due to insufficient power rather than any biological differences. Of note, we found few studies assessing inflammation level and diabetes in Chinese subjects, although CRP did appear associated with type 2 diabetes in a cross-sectional study (17). CRP was associated with remaining glucose intolerant or progressing to diabetes among 228 Hong Kong Chinese with impaired glucose tolerance; however, only 21 participants developed diabetes, precluding analyses about incident diabetes (18). The lower levels of inflammation present among Chinese in this sample may also explain our findings, if there is a threshold effect. At least one study (19) including U.S. resident Chinese women also reported lower levels of CRP compared with other race/ethnic participants, even after adjusting for BMI differences.

The current epidemic of diabetes is projected to worsen such that up to 7% of the U.S. population will be affected by 2050 (from 4% in 2000); up to 37% of the increased prevalence is estimated to be due to projected changes in the country's demographic composition with a decrease in whites and increase in minority groups (20). As such, investigating diabetes incidence in a multiethnic sample is important. One recent analysis (21) in a multiethnic cohort of postmenopausal women suggested both CRP and IL-6 predicted incident diabetes similarly in white, black, Hispanic, and Asian/Pacific Islander subjects. There are several important differences between the study by Liu et al. (21) and the present one herein, including a case-control design and reliance on self-report or hospital discharge diagnoses; nevertheless, they also observed significant attenuation of the association between CRP or IL-6 and incident diabetes when adjusting for BMI and baseline fasting glucose. CRP remained associated with incident diabetes in the

Insulin Resistance and Atherosclerosis Study (a white, African American, and Hispanic cohort) after adjustment for metabolic syndrome variables and insulin resistance; however, they did not present a stratified analysis (22). We are not aware of other studies reporting stratified results from a multiethnic sample including men. Our results suggest that the relationship between both CRP and IL-6 and incident diabetes is likely to be largely mediated by insulin resistance and adiposity in these racial/ethnic groups. Fat cells, in particular visceral adipocytes, are known to be important producers of inflammatory proteins. There is substantial debate whether inflammation is directly playing a pathophysiologic role or whether it is simply a marker for underlying processes that affect both insulin sensitivity and inflammation levels (10).

Our results suggest that CRP and IL-6, or a composite score encompassing both, but not fibrinogen level, may be considered as potentially independent predictors of diabetes in a racially diverse population such as the U.S. It is plausible that inflammatory markers may be utilized to refine diabetes risk prediction and thus better target (or motivate) individuals for lifestyle interventions. However, we note that CRP levels did not appreciably improve diabetes risk prediction in the Framingham Offspring cohort or the Insulin Resistance and Atherosclerosis Study (22,23). One potential implication of the many studies demonstrating a relationship between inflammation and diabetes is that trials of anti-inflammatory agents be undertaken to prevent diabetes. However, a recent randomized trial failed to demonstrate that chronic aspirin use prevented incident diabetes in women (24). In contrast, the Finish Diabetes Prevention Study lifestyle intervention reduced CRP and IL-6 (25). Future research aimed at understanding the potential causal relationship between inflammation and diabetes may yield novel approaches to preventing diabetes. However, it remains important to focus on ways to increase adherence to lifestyle interventions of exercise and diet to reduce diabetes risk.

Acknowledgments— This research was supported by contracts N01-HC-95159 through N01-HC-95165 and N01-HC-95169 from the National Heart, Lung, and Blood Institute.

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

A preliminary version of this study was presented in abstract form at the 67th Scientific Sessions of the American Diabetes Association, Chicago, Illinois, 22–26 June 2007.

The authors thank the other investigators, the staff, and the participants of the MESA study for their valuable contributions. A full list of participating MESA investigators and institutions can be found at <http://www.mesa-nhlbi.org>.

References

1. Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. *JAMA* 2001;286:327–334
2. Spranger J, Kroke A, Mohlig M, Hoffmann K, Bergmann MM, Ristow M, Boeing H, Pfeiffer AF. Inflammatory cytokines and the risk to develop type 2 diabetes: results of the prospective population-based European Prospective Investigation into Cancer and Nutrition (EPIC)-Potsdam Study. *Diabetes* 2003;52:812–817
3. Thorand B, Lowel H, Schneider A, Kolb H, Meisinger C, Frohlich M, Koenig W. C-reactive protein as a predictor for incident diabetes mellitus among middle-aged men: results from the MONICA Augsburg cohort study, 1984–1998. *Arch Intern Med* 2003;163:93–99
4. Hu FB, Meigs JB, Li TY, Rifai N, Manson JE. Inflammatory markers and risk of developing type 2 diabetes in women. *Diabetes* 2004;53:693–700
5. Thorand B, Kolb H, Baumert J, Koenig W, Chambless L, Meisinger C, Illig T, Martin S, Herder C. Elevated levels of interleukin-18 predict the development of type 2 diabetes: results from the MONICA/KORA Augsburg Study, 1984–2002. *Diabetes* 2005;54:2932–2938
6. Doi Y, Kiyohara Y, Kubo M, Ninomiya T, Wakugawa Y, Yonemoto K, Iwase M, Iida M. Elevated C-reactive protein is a predictor of the development of diabetes in a general Japanese population: the Hisayama Study. *Diabetes Care* 2005;28:2497–2500
7. Rutter MK, Meigs JB, Sullivan LM, D'Agostino RB Sr, Wilson PW. C-reactive protein, the metabolic syndrome, and prediction of cardiovascular events in the Framingham Offspring Study. *Circulation* 2004;110:380–385
8. Festa A, D'Agostino R Jr, Tracy RP, Haffner SM. Elevated levels of acute-phase proteins and plasminogen activator inhibitor-1 predict the development of type 2 diabetes: the Insulin Resistance Atherosclerosis Study. *Diabetes* 2002;51:1131–1137
9. Calabro P, Chang DW, Willerson JT, Yeh ET. Release of C-reactive protein in response to inflammatory cytokines by human adipocytes: linking obesity to

- vascular inflammation. *J Am Coll Cardiol* 2005;46:1112–1113
10. Kristiansen OP, Mandrup-Poulsen T. Interleukin-6 and diabetes: the good, the bad, or the indifferent? *Diabetes* 2005;54(Suppl. 2):S114–S124
 11. Krakoff J, Funahashi T, Stehouwer CD, Schalkwijk CG, Tanaka S, Matsuzawa Y, Kobes S, Tataranni PA, Hanson RL, Knowler WC, Lindsay RS. Inflammatory markers, adiponectin, and risk of type 2 diabetes in the Pima Indian. *Diabetes Care* 2003;26:1745–1751
 12. Cowie CC, Rust KF, Byrd-Holt DD, Eberhardt MS, Flegal KM, Engelgau MM, Saydah SH, Williams DE, Geiss LS, Gregg EW. Prevalence of diabetes and impaired fasting glucose in adults in the U.S. population: National Health and Nutrition Examination Survey 1999–2002. *Diabetes Care* 2006;29:1263–1268
 13. Rewers M, Hamman RF. Risk factors for non-insulin-dependent diabetes. In *Diabetes in America*. 2nd ed. Harris MI, Cowie CC, Stern MP, Boyko EJ, Reiber GE, Bennett PH, Eds. Washington D.C., U.S. Govt. Printing Office, 1995, p. 179–220
 14. Bild DE, Bluemke DA, Burke GL, Detrano R, Diez Roux AV, Folsom AR, Greenland P, Jacob DR Jr, Kronmal R, Liu K, Nelson JC, O'Leary D, Saad MF, Shea S, Szklo M, Tracy RP. Multi-ethnic study of atherosclerosis: objectives and design. *Am J Epidemiol* 2002;156:871–881
 15. Cushman M, Cornell ES, Howard PR, Bovill EG, Tracy RP. Laboratory methods and quality assurance in the Cardiovascular Health Study. *Clin Chem* 1995;41:264–270
 16. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* 1985;28:412–419
 17. Ye X, Franco OH, Yu Z, Li H, Hu FB, Liu H, Wang X, Tang H, Liu Y, Chen Y, Lin X. Associations of inflammatory factors with glycaemic status among middle-aged and older Chinese people. *Clin Endocrinol (Oxf)* 2009;70:854–862
 18. Tan KCB, Wat NMS, Tam SCF, Janus ED, Lam TH, Lam KSL. C-reactive protein predicts the deterioration of glycemia in Chinese subjects with impaired glucose tolerance. *Diabetes Care* 2003;26:2323–2328
 19. U.S. Preventive Services Task Force. Screening for high blood pressure: recommendations and rationale. *Am J Prev Med* 2003;25:159–164
 20. Boyle JP, Honeycutt AA, Narayan KM, Hoerger TJ, Geiss LS, Chen H, Thompson TJ. Projection of diabetes burden through 2050: impact of changing demography and disease prevalence in the U.S. *Diabetes Care* 2001;24:1936–1940
 21. Liu S, Tinker L, Song Y, Rifai N, Bonds DE, Cook NR, Heiss G, Howard BV, Hotamisligil GS, Hu FB, Kuller LH, Manson JE. A prospective study of inflammatory cytokines and diabetes mellitus in a multi-ethnic cohort of postmenopausal women. *Arch Intern Med* 2007;167:1676–1685
 22. Hanley AJG, Karter AJ, Williams K, Festa A, D'Agostino RB Jr, Wagenknecht LE, Haffner SM. Prediction of type 2 diabetes mellitus with alternative definitions of the metabolic syndrome: the Insulin Resistance Atherosclerosis Study. *Circulation* 2005;112:3713–3721
 23. Wilson PW, Meigs JB, Sullivan L, Fox CS, Nathan DM, D'Agostino RB Sr. Prediction of incident diabetes mellitus in middle-aged adults: the Framingham Offspring Study. *Arch Intern Med* 2007;167:1068–1074
 24. Pradhan AD, Cook NR, Manson JE, Ridker PM, Buring JE. A randomized trial of low-dose aspirin in the prevention of clinical type 2 diabetes in women. *Diabetes Care* 2009;32:3–8
 25. Herder C, Peltonen M, Koenig W, Sutfels K, Lindstrom J, Martin S, Ilanne-Parikka P, Eriksson JG, Aunola S, Keinanen-Kiukkaanniemi S, Valle TT, Uusitupa M, Kolb H, Tuomilehto J. Anti-inflammatory effect of lifestyle changes in the Finnish Diabetes Prevention Study. *Diabetologia* 2009;52:433–442