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Association of Endothelial and Oxidative Stress with Metabolic Syndrome and Subclinical Atherosclerosis: Multi-Ethnic Study of Atherosclerosis

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Abstract

Objectives—A cluster of metabolic abnormalities termed metabolic syndrome (MetS) is associated with vascular endothelial dysfunction and oxidative internal milieu. We examined whether the association of MetS with subclinical atherosclerosis is explained by biomarkers of endothelial damage and oxidative stress.

Methods—MESA is a population based study of 45-84 year old individuals of four US ethnicities without clinical cardiovascular disease. A random sample of 997 MESA participants had data on the following biomarkers: von Willebrand Factor, soluble intercellular adhesion molecule-1 (sICAM1), CD40 ligand, soluble thrombomodulin, E-selectin, and oxidized LDL (oxLDL). We examined whether the associations of MetS with B-mode ultrasound-defined common and internal carotid intimal medial thickness (IMT) and coronary artery calcium (CAC) measured using computerized tomography were explained by the biomarkers using multiple regression methods.

Results—MetS was associated with higher levels of each of the biomarkers ($p<0.001$, CD40L suggestive association $p=0.004$), with greater IMT ($p<0.001$), and with greater extent of CAC in those in whom CAC was detectable ($p=0.01$). The association of MetS with measures of subclinical atherosclerosis remained unchanged after adjustment for the biomarkers. After

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adjusting for MetS, oxLDL was suggestively associated with greater prevalence of detectable CAC (p=0.005) and thicker internal carotid IMT (p=0.002), while sICAM-1was significantly associated with greater prevalence of detectable CAC (p=0.001).

Conclusions—The association of MetS with subclinical atherosclerosis was independent of its association with biomarkers of endothelial damage and oxidative stress, suggesting that metabolic abnormalities and oxidative endothelial damage may lead to atherosclerotic disease through distinct mechanisms.

Keywords

Metabolic syndrome; biomarkers; coronary artery atherosclerosis; carotid arteries

Introduction

The metabolic syndrome (MetS) is a combination of cardiovascular risk factors (abnormal elevations in glucose, lipid, or blood pressure levels) and of a anthropometric marker (waist circumference) that is associated with atherosclerotic disease (Grundy et al., 2005).

MetS is associated with impaired flow-mediated dilatation (Hamburg et al., 2008), which is a measure of endothelial dysfunction and an early step in the pathogenesis of subclinical vascular disease progression (Halcox et al., 2009). In addition, MetS is also a proinflammatory state (Sakkinen et al., 2000) associated with oxidative stress (Holvoet, 2008; Holvoet et al., 2004; Holvoet et al., 2008). Endothelial dysfunction as evinced by biomarkers of endothelial damage (Blann et al., 2002) and oxidative stress as revealed by oxidized LDL (Holvoet et al., 2003) are themselves proximal antecedents of atherosclerosis. Thus it would seem likely that the association between MetS and early atherosclerosis is solely due to a coexistent state of oxidative stress and endothelial damage. We thus hypothesize that the biomarkers of endothelial function that are associated with MetS and subclinical atherosclerosis partly explain the association between the MetS and subclinical atherosclerosis.

We investigate this hypothesis in two steps, investigating whether : 1) MetS is associated with circulating markers of endothelial function and oxidative stress and; 2) the association of MetS with subclinical atherosclerosis is attenuated by circulating biomarkers of endothelial damage and oxidative stress.

Methods

We used baseline data from the Multi-Ethnic Study of Atherosclerosis, a multi-center study of four US race/ethnicities (White, Chinese, Black and Hispanic). Of the 6814 men and women free of clinical cardiovascular disease at baseline, a random sample of 1000 individuals had measurements of circulating biomarkers. This analysis included 997 individuals who had measurements available for oxidized low density lipoprotein (ox-LDL, a marker for oxidative stress) or for the following markers of endothelial damage: soluble intercellular adhesion molecule-1 (sICAM-1), CD40 ligand (CD40L), soluble thrombomodulin (sTM), soluble E-selectin (sESEL) and von Willebrand factor (vWF).

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MetS was defined according to the modified NCEP-ATP-III criteria (Grundy et al., 2005) using medical history, anthropometric measurements, seated blood pressure, and fasting glucose and lipid panels. Persons having ≥3 of the following abnormalities met the criteria for MetS: (1) waist circumference ≥0.88 m in women, ≥1.02 m in men, (2) serum triglycerides ≥1.695 mmol/L, (3) HDL-C ≤1.295 mmol/L in women, ≤1.036 mmol/L in men, (4) systolic blood pressure ≥135 and/or diastolic blood pressure ≥85 mmHg or the use of antihypertensive medications, (5) fasting glucose ≥ 5.55 mmol/L, or taking anti-diabetic medications.

Assessment of demographic and clinical variables

Personal history including current smoking during the past 30 days, education, exercise, demographic data, and medical history were collected using interviewer-administered forms. Medication use was confirmed by examination of medication containers, if available. Height and weight were measured using a stadiometer and platform balance, respectively. Waist girth was measured horizontally at the level of the umbilicus in the standing position using an anthopometric tape. Serum lipid profile and plasma glucose were measured from fasting blood draws and analyzed at the Core Laboratory at University of Vermont, Burlington, VT.

Biomarker measurements

Levels of oxLDL were measured by Dr. Holvoet's laboratory, with an mAb-4E6-based competition ELISA (analytical CV 7.4-8.3%).(Holvoet et al., 1998) All other biomarkers were assayed at the Laboratory for Clinical Biochemistry Research (University of Vermont, Burlington, VT) as follows: sICAM-1 by ELISA (Parameter Human sICAM-1 Immunoassay; R&D Systems, Minneapolis, MN, Laboratory; Analytical CV 5.0%), CD40L by ultra-sensitive quantitative sandwich enzyme immunoassay (Quantikine Human soluble CD40 Ligand Immunoassay; R&D Systems, Minneapolis, MN, CV 4.5-6.4%), sTM by ELISA (Asserachrom Thrombomodulin, Diagnostica Stago; Asnières-sur-Seine, France, CV 12%), soluble E-selectin by high sensitivity quantitative sandwich enzyme immunoassay (Parameter Human sE-Selectin Immunoassay; R&D Systems, Minneapolis, MN, CV 4.7-8.8%), vWf by immunoturbidimetric assay on the STAR analyzer (Liatest vWF; Diagnostica Stago, Parsippany, NJ, intra- and inter-assay CV 3.7-4.5%).

Assessment of subclinical atherosclerosis

Coronary artery calcium was measured using computerized tomography using either electron beam tomography or helical tomography (Carr et al., 2005). The Agatston score (Agatston et al., 1990) was used to quantify coronary artery calcification (CAC). The intimal-medial thickness (IMT) of the common and internal carotid arteries was assessed using B-mode ultrasound (O'Leary et al., 1991). Multiple views were obtained of the left and right sides, and the maximal IMT in each view was averaged for the common (C-IMT) and internal (I-IMT) carotid arteries.

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Statistical methods

The difference in demographic variables by MetS status was tabulated. Differences were evaluated using t-tests for continuous variables and chi-squared tests for categorical variables.

The medians and interquartile ranges of biomarker variables were tabulated by MetS status, and age, sex, and race-adjusted differences in log-transformed biomarker levels were tested using linear regression. For multivariable regression analyses, the biomarker levels were log-transformed because they had a right-skewed distribution. A large number of individuals had no detectable CAC, and those with detectable CAC had a right-skewed variable distribution. Thus CAC was analyzed in two stages: in the first stage, the relative prevalence of detectable CAC was modeled as a dichotomous variable; in the second stage, log-transformed CAC was used in regression analysis only among those with detectable CAC. The standard error of association statistics for IMT may be incorrectly estimated because of non-normality of the IMT distributions. We have thus estimated bootstrapped standard errors for these regression analyses using 1000 resampled datasets.

Adjusted models assessing the association of subclinical atherosclerosis variables with MetS and the biomarkers were performed using general linear model methods. Presence of dichotomous detectable CAC was modeled with a logarithmic link function, and Gaussian error, to obtain prevalence ratios associated with the independent variables. All other dependent variables were continuous, and modeled using ordinary least squared linear regression. Choice of covariates for adjustment included the demographic covariates age, sex and race, as well as those cardiovascular risk factors not represented among the metabolic abnormalities included in the metabolic syndrome definition. BMI was not included as a covariate because it is strongly correlated with waist girth (Spearman correlation = 0.85 within the sample).

In analysis of MetS (as defined by NCEP) dichotomizing each MetS component for classification and making a simple count of abnormalities may lose information regarding independent associations. Thus, we repeated all models above including the individual components of MetS, in place of MetS presence or absence.

The sICAM-1 assay immunoreactivity depends on the K56M polymorphism (rs5491) (Register et al., 2004), which is frequent in African Americans but not in Caucasian, East Asian or Hispanic populations (NCBI, 2009). Thus sICAM-1 analyses were also performed excluding the African-American sample, as done by others (Tang et al., 2007).

Because many biomarkers are tested, there are issues regarding inflation of false positive hypothesis tests due to multiple testing. Thus age, sex and race adjusted associations of 6 biomarkers is considered primary analysis, with the significance level for p-values being set at 0.05/6 = 0.0083. All other covariate models are exploratory and explanatory.

Sensitivity analyses: In a multi-site multiethnic study, there is a possibility that study site and socioeconomic status may confound associations. Furthermore habitual physical activity may confound the associations. We performed all primary association analyses including

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study site, educational status (as a proxy for socioeconomic status), and habitual intentional weekly exercise (METS/week) as covariates to assess whether the associations remain after these adjustments. Analyses using fasting glucose and insulin measures instead of the metabolic syndrome to estimate insulin resistance (HOMA-IR, Matthews et al., 1985) or its inverse transformation (QUICKI, Katz et al., 2000) are presented in the online appendix.

Results

The demographic and cardiovascular risk factor characteristics are shown by MetS status in Table 1. MetS was present in a third of the sample. The individuals with MetS were somewhat older, had a lower percentage of men, and were more likely to be Black or Hispanic than those without MetS. There was no difference in prevalence of current smoking or high cholesterol levels. The predominant components of the MetS were a large waist and hypertension.

All biomarkers were higher in those with than in those without MetS, including after adjustment for age, sex and race (Table 2), except for vWF. The association of Table 3 shows that sICAM and E-selectin were associated with greater prevalence of CAC, when adjusted for age, sex, race, total cholesterol and current smoking. The correlation of oxLDL with total cholesterol was of a magnitude that neither total cholesterol nor oxLDL were separately associated with CAC prevalence, though they were jointly associated using likelihood ratio test. Thus we tabulated the association in a model not adjusted for total cholesterol. These associations remained significant after adjustment for either the metabolic syndrome or the five individual metabolic abnormalities. None of the biomarkers were associated with the magnitude of calcification among those with detectable calcium.

E-selectin was associated with thicker IMT in the common carotid artery after age, sex, race, total cholesterol and smoking-adjustment, but this association was marginal on adjustment for MetS (Table 4) or the five individual metabolic abnormalities. Ox-LDL was associated with thicker internal carotid IMT, when adjusted for age, sex, race and current smoking, and also after further adjustment for MetS or the five individual metabolic abnormalities.

After adjusting for age, sex, total cholesterol, current smoking and race, MetS was not associated with prevalence of CAC, but it was associated with a higher level of CAC if detectable (ratio of geometric means of CAC levels 1.55) (Table 5). This association was unchanged when biomarker variables were added as covariates. None of the individual dichotomized components of the metabolic syndrome were significantly associated with CAC prevalence or extent in any model in this subsample of MESA. Those with MetS had significantly greater IMT in both the common and internal carotid arteries. These associations remained after adjustment for the markers of oxidative stress and endothelial dysfunction. The individual components large waist circumference ($\beta = 0.04$ mm, $p<0.001$), low HDL-C ($\beta = 0.02$ mm, $p=0.025$), high blood pressure ($\beta = 0.03$ mm, $p=0.001$) and glucose abnormality ($\beta = 0.02$ mm, $p = 0.065$) were associated with common carotid IMT, significantly or at the borderline and these associations were not much changed by addition of biomarkers (large waist circumference: $\beta = 0.04$ mm, $p<0.001$, low HDL-C: $\beta = 0.02$ mm, $p=0.030$, high blood pressure: $\beta = 0.04$ mm, $p=0.001$, and glucose abnormality: $\beta = 0.02$ mm, $p = 0.049$).

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For internal IMT there were significant of borderline associations with the individual components of low HDL (β = 0.07 mm, p=0.057) and high blood pressure (β = 0.09 mm, p=0.002), and on addition of biomarkers in the model only the association of high blood pressure (β = 0.10 mm, p=0.002) remained significant.

Sensitivity analyses: All associations for ox-LDL and sICAM-1 remained unchanged in models adjusting for site, education level (categorized by completion of school and various kinds of higher education) and exercise (total intentional weekly exercise in METS). The associations of E-selectin with coronary calcium prevalence became significant at the borderline (p=0.1), and with common carotid intimal medial thickness became non-significant (p=0.15) in models adjusting for education level.

Discussion

The main findings in this study were that persons with MetS have greater extent of CAC if any was detectable, and thicker carotid IMT, however, these associations were not explained by higher levels of the biomarkers of oxidative stress and endothelial damage. Indeed, others have reported that inflammatory and vascular biomarkers only partially explain the association of metabolic syndrome with coronary artery disease (Jacobs et al., 2009). Though we and others have previously shown in the whole study population that there was significant association of MetS with CAC prevalence (Bertoni et al., 2007; Vaidya et al., 2007), and others have shown that there was some association of the homeostatic model insulin resistance measure (HOMA-IR) with CAC prevalence, in this smaller sample of individuals with biomarker measures, the association of MetS with CAC prevalence within MESA was not significant. OxLDL was previously shown to be associated with carotid plaque and CAC in MESA (Holvoet et al., 2007). We have shown that ox-LDL was associated with greater CAC prevalence and greater internal carotid IMT, while sICAM-1 and E-selectin were associated with greater CAC prevalence independent of MetS. Thus the presence of multiple metabolic abnormalities may be associated with structural subclinical disease in the coronary and carotid arteries through mechanisms that are not fully captured in these biomarkers. Such mechanisms might include the proinflammatory (Sakkinen et al., 2000) and prothrombotic (Sakkinen et al., 2000) milieu associated with MetS. Although the co-occurrence of abnormalities in the form of MetS is associated with CAC prevalence in our sample, none of the individual metabolic abnormalities has an independent association in this subsample of MESA. All but one of the abnormalities (high triglycerides being the exception) were associated with intimal-medial thickening in at least one of the carotid arteries. This suggests that the association of the individual abnormalities with subclinical atherosclerosis may underlie some but not all of the association of MetS with vascular effects.

We also observed that some of the biomarkers were independently associated with subclinical atherosclerosis. OxLDL represents the accumulation of LDL modified by a milieu under oxidative stress (Holvoet and Collen, 1994). Our analyses suggest that oxidative stress is associated with the presence of detectable calcified coronary plaque and internal carotid IMT independently of the presence of MetS. Presumably this is due to the direct impact of oxidative stress on the atherosclerotic process (Holvoet, 2008).

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E-selectin is a protein expressed on the surface of endothelial cells when they are activated by chemokines such as TNF-alpha or interleukin-1 (Constans and Conri, 2006). Our results suggest that these processes are associated with atherosclerosis independent of metabolic syndrome. There is a suggestion in our analysis that this association may be confounded by socioeconomic status (using educational level as the proxy). However, it is not clear if the E-selectin associations reported in the main results were due to confounding, or that they could not be detected as significant in the sensitivity analyses due to inadequate power after adding additional covariates.

The other biomarkers we evaluated are all markers of endothelial damage that represent distinct biochemical or structural processes. Thrombomodulin is a vasoprotective antithrombotic molecule expressed in the endothelial cell membrane (Wu, 2003). The presence of the soluble fragment of this molecule may represent either an adaptive excess of the protective molecule on an essentially healthy endothelium, or be a marker of excessive endothelial damage if also accompanied by high levels of other markers such sICAM (Wu et al., 2003). vWF is produced by endothelial cells and secreted into the subendothelium and the plasma. vWF is directly involved with platelet aggregation and indirectly involved in thrombosis as a chaperone for coagulation factor VIII (Spiel et al., 2008). Thus, vWF is both normally secreted, and abnormally released into the circulation by endothelial damage. Previous studies have suggested that vWF levels are only weakly associated with coronary disease among initially healthy individuals, but are more strongly associated with coronary disease among those with established coronary disease (reviewed by Spiel et al.(Spiel et al., 2008)) CD40 ligand is present in a bound form on $CD4^+$ T cells, platelets and endothelial cells, and in the soluble form in the plasma (Schonbeck and Libby, 2001). CD40L was associated with unstable, rather than stable angina in one study (Aukrust et al., 1999). ICAM-1 is induced on the surface of endothelial cells in response to inflammatory signaling and endothelial cell shedding, and is presumably a major source of circulating sICAM-1 (Lawson and Wolf, 2009). Levels of sICAM, which may represent endothelial damage, were found to be significantly associated with atherosclerosis risk and unrelated to sTM in the Atherosclerosis Risk in Communities study (Wu et al., 2003). Of the endothelial biomarkers assessed here, only sICAM-1 was independently associated with the risk of a subclinical atherosclerosis measure. This is consistent with the possibility that the levels of the other markers are partially adaptive and partially maladaptive in the otherwise healthy individual.

A significant strength of our study is its large, well-characterized population sample in terms of the metabolic syndrome, biomarkers and subclinical atherosclerosis imaging. Among our study's limitations was its cross sectional design, which, given the lack of information on temporality, prevented us from making strong inferences on the mediating role of biomarkers. This study was also not designed to assess the association of mediation of individual metabolic components in the association of the biomarkers with subclinical atherosclerosis, but rather joint prevalence of multiple metabolic factors as a syndrome. Additionally, because biomarkers may be imprecise measures of endothelial dysfunction, we cannot categorically assert that the association of MetS with subclinical atherosclerosis is independent of endothelial dysfunction.

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In summary, MetS is associated with biomarkers of oxidative stress and endothelial dysfunction, but this does not seem to explain the association of MetS with subclinical atherosclerosis. OxLDL, a marker of oxidative stress, and sICAM-1, a marker of proinflammatory endothelial injury, were independently associated with some measures of subclinical atherosclerosis. Our study suggests that metabolic abnormalities associated with MetS are likely to cause vascular dysfunction through mechanisms independently of the biomarkers we have investigated. Alternatively, the co-occurrence of multiple metabolic abnormalities may be a more precise marker of endothelial dysfunction than any of the biomarker levels included in this study. Additional research using a longitudinal design is needed to distinguish between the two possibilities.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Table 1

Characteristics of the Multi-Ethnic Study of Atherosclerosis random sample at baseline (2000-2002)

	Metabolic Syndrome		
	Present	**Absent**	**p**
N	328	669	
Age	61.0 (9.4)	58.6 (9.8)	0.003
Male sex	123 (38%)	304 (45%)	0.017
Race			
White	136 (41%)	322 (48%)	0.017
Chinese	25 (8%)	73 (11%)	
Black	75 (23%)	134 (20%)	
Hispanic	92 (28%)	140 (20%)	
Current Smoking	53 (16%)	101 (15%)	0.663
Total cholesterol < 5.18 mmol/L	179 (55%)	395 (59%)	0.393
Total cholesterol 5.18-6.20 mmol/L	117 (36%)	218 (33%)	
Total cholesterol > 6.21 mmol/L	32 (10%)	56 (8%)	
Metabolic syndrome components*			
High fasting glucose (>5.55 mmol/L) or diabetes (or anti-diabetic medication)	163 (50%)	55 (8%)	
Large Waist (≥0.88 m in women, ≥1.02 m in men)	279 (85%)	264 (39%)	
High blood pressure (systolic ≥135 and/or diastolic ≥85 mmHg) or using antihypertensive medications	251 (77%)	238 (36%)	
Low High Density Lipoprotein - Cholesterol (≤1.295 mmol/L in women, ≤1.036 mmol/L in men)	223 (68%)	121 (18%)	
High Triglycerides (serum triglycerides ≥1.695 mmol/L)	221 (67%)	75 (11%)	

*Distribution of variables included in the metabolic syndrome definition automatically differ by metabolic syndrome presence. Thus no statistical test of difference was performed.

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Table 2

Median and interquartile ranges of circulating biomarkers, and differences according to metabolic syndrome (MetS), Multi-Ethnic Study of Atherosclerosis, 2000-2002.

	Non-MetS	MetS	unadjusted (rank sum)	*adjusted p
von Willebrand Factor (%)	126.5 [97 to 165.5]	138 [103 to 182]	0.003	0.22
E-selectin (ng/mL)	48.4 [34.6 to 62.3]	56.8 [45.2 to 77.1]	<0.001	<0.001
Soluble Thrombomodulin (ng/mL)	34 [25 to 43]	37 [27 to 48]	<0.001	<0.001
CD40 ligand (pg/dL)	3.4 [2.1 to 5.2]	3.9 [2.5 to 5.7]	0.004	0.021
Soluble Intercellular Adhesion Molecule 1 (ng/mL)	266 [230.7 to 306]	293 [250 to 338]	<0.001	<0.001
Oxidized LDL (mg/dL)	0.70 [0.51 to 1.0]	0.90 [0.62 to 1.31]	<0.001	<0.001

* adjusting for age sex and race

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Table 3

Measures of association of biomarker levels [95% confidence intervals] with the presence of detectable coronary artery calcium (CAC) and magnitude of calcification (if detectable), Multi-Ethnic Study of Atherosclerosis, 2000-2002.

	Prevalence of detectable CAC	**Relative CAC proportion (among those with detectable CAC)**
	Model 1: Individual biomarker associations adjusted for race, sex, age, total cholesterol, and current smoking	
	Prevalence ratio/1 log-unit greater biomarker	% greater CAC per % higher level of biomarker
oxLDL	1.10 [0.97 to 1.24]	-0.02 [-0.38 to 0.34]
oxLDL*	1.15 [1.05,1.25]	-0.08 [-0.38, 0.21]
sICAM1	1.47 [1.18,1.82]	0.34 [-0.30, 0.98]
sICAM1 (excluding African-Americans)	1.65 [1.27 to 2.15]	0.48 [-0.32, 1.25]
CD40L	1.03 [0.95,1.13]	0.03 [-0.23,0.29]
sTM	0.97 [0.87,1.08]	0.21 [-0.15, 0.57]
ESEL	1.14 [1.01,1.30]	-0.06 [-0.32,0.43]
vWF	1.08 [0.93,1.25]	0.08 [-0.30,0.45]
	Model 2: Model 1 further adjusted for MetS	
	relative prevalence/1 log-unit greater biomarker	% greater CAC per % higher level of biomarker
oxLDL	1.08 [0.95 to 1.23]	-0.11 [-0.48 to 0.25]
oxLDL*	1.14 [1.04,1.25]	-0.13 [-0.43, 0.16]
sICAM1	1.46 [1.17,1.81]	0.25 [-0.40, 0.89]
sICAM1 (excluding African-Americans)	1.64 [1.26, 2.14]	0.32 [-0.46, 1.11]
CD40L	1.03 [0.94,1.12]	0.00 [-0.26,0.26]
sTM	0.95 [0.85,1.07]	0.16 [-0.20, 0.52]
ESEL	1.14 [1.00,1.29]	-.02 [-0.40,0.36]
vWF	1.07 [0.92,1.24]	0.06 [-0.32,0.43]

OxLDL – Oxidized low-density lipoprotein, sICAM1 – soluble intercellular adhesion molecule-1, CD40L – CD40 ligand, ESEL – E-selectin, vWF – von Willebrand factor;

*model not adjusting for total cholesterol. In models that include both oxLDL and total cholesterol, the individual associations of the two variables with CAC prevalence are non-significant, though the two variables are jointly significant in the model using the likelihood ratio test.

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Table 4

Measures of association of biomarker levels [95% confidence intervals] with common and internal carotid intimal-medial thickness (IMT), Multi-Ethnic Study of Atherosclerosis, 2000-2002

	Common Carotid IMT	Internal Carotid IMT
	mm/1 log-unit difference in biomarker	
	Model 1: adjusted for race, sex, age, total cholesterol, and current smoking	
oxLDL	0.01 [0.00, 0.03]	0.09 [0.03 to 0.16]
oxLDL*	0.01 [0.00, 0.03]	0.12 [0.06, 0.19]
sICAM1	0.01 [-0.02, 0.05]	0.13 [0.02, 0.23]
sICAM1 (excluding African-Americans)	0.00 [-0.04, 0.04]	0.13 [-0.01,0.27]
cd40L	0.01 [-0.05, 0.03]	0.01 [-0.04,0.06]
sTM	0.01 [-0.01, 0.03]	0.02 [-0.05,0.08]
ESEL	0.02 [0.00, 0.04]	0.06 [0.00,0.13]
vWF	-.01 [-0.03, 0.02]	0.00 [-0.08, 0.07]
	Model 2: Model 1 further adjusted for MetS	
oxLDL	0.00 [-0.02 to 0.02]	0.07 [0.00 to 0.14]
oxLDL*	0.01 [-0.01, 0.02]	0.11 [0.04,0.17]
sICAM1	0.00 [-0.03,0.04]	0.10 [-0.01,0.21]
sICAM1 (excluding African-Americans)	-0.01 [-0.06,0.03]	0.10 [-0.03, 0.23]
cd40L	0.01 [-0.01,0.02]	0.01 [-0.04,0.05]
sTM	0.00 [-0.02,0.02]	0.00 [-0.06,0.06]
ESEL	0.01 [-0.01,0.03]	0.04 [-0.03,0.11]
vWF	-0.01 [-0.04,0.01]	-0.01 [-0.09, 0.07]

OxLDL – Oxidized low-density lipoprotein, sICAM1 – soluble intercellular adhesion molecule-1, CD40L – CD40 ligand, ESEL – E-selectin, vWF – von Willebrand factor;

*model not adjusting for total cholesterol. In models that include both oxLDL and total cholesterol, the individual associations of the two variables with CAC prevalence are non-significant, though the two variables are jointly significant in the model using the likelihood ratio test.

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Table 5

Measures of association [95% confidence intervals] of subclinical atherosclerosis according to the presence of metabolic syndrome (MetS), Multi-Ethnic Study of Atherosclerosis, 2000-2002

	Model 1: Association adjusted for age, sex, race, total cholesterol and current smoking	**Model 2: Model 1 + all biomarkers (allowing race by sICAM interaction)**
Prevalence ratio of detectable CAC (MetS:non-MetS)	1.07 [0.95, 1.20]	1.02 [0.90,1.16]
Ratio of geometric mean CAC (MetS:non-MetS)	1.54 [1.10, 2.16]	1.48 [1.03, 2.12]
Difference in mean common carotid IMT (MetS vs. non-MetS, mm)	0.05 [0.03,0.07]	0.05 [0.02,0.07]
Difference im mean internal carotid IMT (MetS vs. non-MetS, mm)	0.13 [0.06,0.20]	0.12 [0.04,0.20]