Elevated Lp-PLA2 Levels Add Prognostic Information to the Metabolic Syndrome on Incidence of Cardiovascular Events Among Middle-Aged Nondiabetic Subjects

Margaretha Persson, Bo Hedblad, Jeanenne J. Nelson, Göran Berglund

Background—To explore potential interrelationships between lipoprotein-associated phospholipase A2 (Lp-PLA2), the metabolic syndrome (MetS), and incident cardiovascular disease (CVD).

Methods and Results—MetS was defined by the National Cholesterol Education Program Adult treatment Panel III criteria in 4480 nondiabetic Malmö Diet and Cancer Study subjects without history of CVD. Incidence of first CVD event (stroke [130 cases] or myocardial infarction [131]) was monitored over 10 years of follow-up. Lp-PLA2 activity and mass were significantly higher in subjects with MetS. Lp-PLA2 activity compared with Lp-PLA2 mass was more strongly correlated to individual components and increased more linearly with number of MetS components. Elevated Lp-PLA2 activity (top compared with bottom tertile), but not elevated Lp-PLA2 mass, increased risk for incident CVD (relative risk, RR: 1.54, 95% CI 1.07 to 2.24), as did MetS (1.42, 1.06 to 1.90) after taking possible confounders into account. Relative to those without either elevated Lp-PLA2 activity or MetS, combination of MetS and elevated Lp-PLA2 activity increased risk for CVD (1.97, 1.34 to 2.90). Elevated Lp-PLA2 activity without MetS increased risk for CVD (1.40, 1.03 to 1.92) but not MetS without elevated Lp-PLA2 activity (1.46, 0.94 to 2.27).

Conclusion—Lp-PLA2 is associated to the MetS. Higher plasma levels of Lp-PLA2 increased risk for incident CVD regardless of MetS. The simultaneous presence of elevated Lp-PLA2 activity and MetS may identify an especially high risk individual. (Arterioscler Thromb Vasc Biol. 2007;27:1411-1416.)

Key Words: metabolic syndrome cardiovascular risk cohort study Lp-PLA2

Several studies have demonstrated an increased cardiovascular risk associated with presence of the metabolic syndrome (MetS).1,2 Whether this risk is confounded by other factors associated with the occurrence of cardiovascular disease (CVD) remains controversial. Lipoprotein-associated phospholipase A2 (Lp-PLA2) is a novel inflammatory marker associated with several components constituting the MetS, low-density lipoprotein (LDL) cholesterol, atherosclerotic disease, and incident CVD.3-10 To the best of our knowledge, no studies have explored whether the risk associated with MetS is confounded by an association with elevated levels of Lp-PLA2.

The influence of Lp-PLA2 on CVD risk can be assessed in terms of either the mass or activity of the enzyme.7-10 Which of these parameters is the best marker for incident CVD remains controversial.11

The study objectives in this population-based nondiabetic cohort have been to study the relationship between MetS and Lp-PLA2, defined in terms of mass and activity, and secondly to assess the independent contribution of MetS and Lp-PLA2 on incident CVD.

Methods

Subjects

The Malmö Diet and Cancer Study (MDCS) is a prospective cohort study examining the association between diet and cancer.12 Subjects aged 45 to 69 years living in Malmö city were eligible for the study. Between October 1991 and February 1994, every other participant was invited to also take part in a substudy of the epidemiology of carotid artery disease.13 Of those 6103, 5540 accepted an invitation for blood sampling under standardized fasting circumstances. Subjects with history of MI or stroke (n=143) and subjects with diabetes mellitus (defined as history of diabetes, use of antidiabetic medication, or with fasting whole blood glucose ≥6.0 mmol/L [n=358]) were excluded from this analysis. Subjects with incomplete baseline data (n=559) for variables constituting MetS (eg, high-density [HDL]-cholesterol, triglycerides, blood pressure, waist circumference, and blood glucose), Lp-PLA2 activity or mass, smoking, LDL-cholesterol, use of lipid-lowering medication, leisure time physical activity, and alcohol consumption were also excluded from the analysis. The study population included 4480 participants. The MDSC was approved by the Ethics Committee of Lund University, Sweden. All participants provided informed consent.
Baseline Examinations

The baseline examination (including anthropometry, blood pressure measurement, blood sampling and a self-administered questionnaire ascertaining previous and current diseases, medication, dietary habits and lifestyle factors including smoking habits and physical activity) has been described in detail previously.\(^{13-14}\) In short, subjects were categorized into current, former- and never smokers. High alcohol consumption was characterized as consumption >30 g alcohol/d for women and >40 g alcohol/d for men. Low level of physical activity was defined as the lowest tertile of a score revealed through 18 questions covering a range of activities in the 4 seasons.\(^{13}\) All subjects were seen by a nurse for standardized anthropometrics and supine blood pressure measurement. Supine blood pressure (mm Hg) was measured once after 10 minutes rest. Waist circumference (in centimeters) was measured in the standing position midway between the lower rib margin and the iliac crest. All participants were instructed to abstain from smoking, alcohol, and food intake, eg, overnight fasting or at least 10 hours before sample drawing. Blood samples were drawn for analysis of blood lipids (total- and HDL-cholesterol and triglycerides) and blood glucose according to standard procedures at the Department of Clinical Chemistry, Malmö University Hospital. LDL-cholesterol concentration was calculated according to Friedewald formula. The assessment of C-reactive protein (CRP) was performed using the Tina-quant CRP latex high-sensitive assay (Roche Diagnostics) on an ADVIA 1650 Chemistry System (Bayer Healthcare). The average coefficient of variation (CV) was 4.59%.

Definition of the Metabolic Syndrome

Presence of MetS was defined in accordance to the current National Cholesterol Education Program Adult Treatment Panel III (NCEP/ATPIII) criteria.\(^{15-16}\) Participants who had 3 or more of the following criteria were considered to have MetS: abdominal obesity (≥102 cm for men and ≥88 cm for women), hypertension (diastolic blood pressure ≥90 mm Hg or systolic blood pressure ≥130/85 mm Hg or on drug treatment for reduced blood pressure), low HDL-cholesterol (men <1.03 mmol/L or <1.30 mmol/L for women or on drug treatment for reduced LDL), high blood pressure (≥130/85 mm Hg or current use of blood pressure lowering medication), and elevated fasting blood glucose (≥5.6 to 6.0 mmol/L).

Measurement of Lp-PLA\(_2\) Activity and Mass

Measurement of Lp-PLA\(_2\) activity in the MDCS has been described in detail previously.\(^{3}\) In short, Lp-PLA\(_2\) activity was measured using [\(^{3}H\)-]platelet activating factor as substrate. The range of detection was 8 to 150 nmol per min per mL. All study samples were tested in duplicate. Samples were retested if the replicate CV was >20%. The average CV was 5.78%. Lp-PLA\(_2\) mass measurements were performed using a commercially available enzyme-linked immunosorbant assay (ELISA) kit (second generation PLAC test diaDexus Inc). All samples were analyzed in duplicate, and samples were retested if the replicate CV was >20%. The average CV was 4.62% on random of 50 first subjects in the MDCS.

Classification of Cardiovascular Events

The procedure for case retrieval has been described previously.\(^{13,14}\) Briefly, the Swedish Hospital Discharge Registry,\(^{17}\) the Stroke register of Malmö,\(^{18}\) and Cause of Death Registry were used. The ascertainment of cases and validity of these registries has been shown to be high.\(^{17,18}\) A CVD event was defined as fatal or nonfatal ascertainment of cases and validity of these registries has been shown to be high.\(^{17,18}\) A CVD event was defined as fatal or nonfatal myocardial infarction (ICD-9: 410), fatal or nonfatal stroke (ICD-9: 430, 431 and 434), or death attributable to CHD (ICD-9: 412 to 414), whichever came first. All subjects were followed from baseline examination until first-occurring CVD event, emigration from Sweden, or death until December 31\(^{*}\) 2003.

Statistical Methods

SPSS was used for the statistical analysis. The distributions of glucose, triglycerides, and hsCRP were markedly skewed and were therefore log-transformed. \(t\) test for continuous variables and \(\chi^2\) test for dichotomous variables were used to examine differences between participants with or without MetS.

Pearson correlation coefficients were calculated between Lp-PLA\(_2\) activity and mass, respectively, and blood pressure, waist circumference, HDL-cholesterol, triglycerides, and blood glucose. Kappa statistics \((\kappa)\) was used to assess the level of agreement between Lp-PLA\(_2\) activity and mass (in tertiles). The incidence (per 1000 person-years) was standardized for sex and age (5-year groups) using direct standardization, and weighted for age-distribution of the present cohort. A general linear model was used to adjust the relations for age, sex, and LDL-cholesterol and to test the linear effects of Lp-PLA\(_2\) across the number of components involved in the MetS. Age- and sex-adjusted \(c\) statistics, analogous to the area under the receiver operator characteristic (ROC) curve, were used to assess the discrimination of CVD prediction models based on high Lp-PLA\(_2\) alone versus those having the MetS alone. The relation of Lp-PLA\(_2\) activity and mass, respectively, and presence of MetS with CVD events during follow-up was assessed by Kaplan–Meier and life table method and quantified by means of the log-rank test. Cox regression model was used to assess the relative risk (RR) of CVD events in relation to Lp-PLA\(_2\) activity or mass. Four steps of adjustment were performed. A basic model included age and sex; the second step included age, sex, LDL-cholesterol, and current use of lipid-lowering medication; a third step included additional adjustment for smoking, the log transformed hsCRP, low physical activity, and high alcohol consumption. In the last step, presence of MetS was added. Possible interactions between elevated Lp-PLA\(_2\) and age and sex, respectively, and between elevated Lp-PLA\(_2\) and presence of MetS on incident CVD were evaluated by including interaction terms in the final multivariate model. Probability values less than 0.05 were considered statistically significant for noninteraction terms and \(P<0.10\) was considered significant for interaction terms.

Results

Baseline characteristics among subjects with and without MetS are shown in Table 1. The proportion of subjects with MetS was 16.4% (14.0% in women and 20.5% in men). Subjects with than without MetS had statistically significant higher levels of Lp-PLA\(_2\) activity (51.3±14.0 versus 43.8±12.2 nmol/min/mL, \(P<0.001\)) and Lp-PLA\(_2\) mass (280.9±84.3 versus 266.5±78.5 ng/mL, \(P=0.005\)).

The Interrelationship Between Lp-PLA\(_2\) Activity and Mass

The correlation coefficient between Lp-PLA\(_2\) activity and mass in this study was 0.57. The concordance between Lp-PLA\(_2\) activity and mass in tertiles is depicted in Table 2. There was a moderate agreement between levels of Lp-PLA\(_2\) activity and mass (\(\kappa\) statistic 0.29). Forty percent (n = 618) of subjects with elevated Lp-PLA\(_2\) activity (top tertile) had levels of Lp-PLA\(_2\) mass in the lowest or middle tertile. Similarly, 40% (n = 616) of subjects with mass in the top tertile had levels of Lp-PLA\(_2\) activity in the lowest or middle tertile. The 5 MetS factors were compared between these 2 discordant groups. After adjustment for age and sex, subjects with high Lp-PLA\(_2\) activity/low to middle Lp-PLA\(_2\) mass had significantly lower HDL-cholesterol levels, higher triglycerides levels, and a higher percentage of subjects with elevated blood pressure (according to NCEP/ATPIII definition) compared with subjects with high Lp-PLA\(_2\) mass/low to middle Lp-PLA\(_2\) activity (for HDL, 1.27±0.34 versus 1.41±0.34 mmol/L, \(P<0.001\); for triglycerides, 1.5±0.7 versus 1.2±0.5, \(P<0.001\); for percentage with high blood pressure, 85% versus 77%, \(P<0.001\), respectively). Other metabolic variables involved in the MetS (eg, glucose, waist circumference) did not differ between the 2 groups (data not shown).

Correlation Between Lp-PLA\(_2\) and Components of the Metabolic Syndrome

Lp-PLA\(_2\) activity correlated with all 5 MetS components, though the association was weaker for fasting glucose and
systolic blood pressure (HDL-cholesterol: $r = -0.30$; waist circumference: $r = 0.22$, triglycerides: $r = 0.31$; fasting glucose: $r = 0.13$ and systolic blood pressure: $r = 0.07$). The corresponding association for Lp-PLA2 mass with MetS components were weaker than that of activity ($r = -0.16$; $r = 0.16$; $r = 0.12$; $r = 0.10$ and $r = 0.07$, respectively). Both mean level of Lp-PLA2 activity and mass increased by increasing number components involved in the MetS ($P$ for trend $<0.001$ and $P<0.001$, respectively; Table 3). This linear relationship remained statistically significant for Lp-PLA2 activity, but not for Lp-PLA2 mass, after taking age, sex, and LDL-cholesterol into account ($P$ for trend $<0.001$ and 0.472, respectively).

Baseline Lp-PLA2 Activity and Mass, MetS, and Incidence of CVD Events
During a mean follow-up time of 10.6 years (47 453 person-years), 261 first CVD events (28 fatal coronary events, 103 non-fatal myocardial infarctions, 101 ischemic, and 29 hemorrhagic strokes) occurred, corresponding to an age- and sex-standardized annual event rate of 6.3 per 1000 person-years (Table 2). Cumulative event probabilities for incident CVD demonstrated similar divergence of the CVD event-free survival curves in groups defined by tertiles of Lp-PLA2 activity and mass levels; event-free survival curves started to deviate early after the baseline examination and continued throughout follow-up.
up, with \( P < 0.001 \) from log-rank tests of significance across tertiles of Lp-PLA2 activity and mass (data not shown). Lp-PLA2 activity were associated with incident CVD (age- and sex-adjusted RR, 1.60; 95% CI, 1.29 to 2.00, \( P = 0.001 \), for top tertile compared with the bottom tertile). The age- and sex-adjusted RR per SD increase was 1.25; 1.11 to 1.40, \( P = 0.002 \), for Lp-PLA2 activity. The corresponding figure for Lp-PLA2 mass was 1.23; 1.06 to 1.44, \( P = 0.009 \), and 1.14; 1.01 to 1.28, \( P = 0.032 \), respectively (Table 4). The area under the ROC curve for Lp-PLA2 activity and mass as continuous variables was 0.62 and 0.58, respectively. After further adjustment for LDL-cholesterol and current lipid-lowering treatment, higher tertiles of Lp-PLA2 activity, but not Lp-PLA2 mass, remained associated with CVD (\( P \) for linear trend 0.007 and 0.075, respectively). The RR for the top tertile of Lp-PLA2 activity was 1.59, \( P = 0.013 \) and for mass it was 1.35, \( P = 0.075 \). Additional adjustment for smoking, hsCRP, physical activity, high alcohol consumption, and presence of MetS attenuated this relationship for activity but it remained statistically significant (RR: 1.46; \( P = 0.047 \); Table 4). For MetS, the risk increase for incident CVD in the model adjusted for all the above factors, including Lp-PLA2 was 1.42; 1.06 to 1.90, \( P = 0.017 \). No evidence of a statistically significant multiplicative interaction was observed between Lp-PLA2 activity and age, sex, or MetS (\( P = 0.71 \); \( P = 0.96 \) and \( P = 0.22 \), respectively). In addition, excluding subjects (n=723) with pharmacological treatment for hypertension and hyperlipidemia did not change the risk increase for incident CVD associated with elevated Lp-PLA2 activity (RR: 1.52), though it did affect the statistical significance attributable to smaller number of subjects (95% CI, 0.98 to 2.36, \( P = 0.063 \)).

Table 2 shows the age- and sex standardized incidence of CVD in relation to groups defined by tertiles of Lp-PLA2 activity and mass levels. For moderate and elevated Lp-PLA2, mass incident CVD increased by increasing level of Lp-PLA2 activity. However, for the same Lp-PLA2 activity increasing levels of Lp-PLA2 mass did not show a similar pattern.

### Combined Analyses of Lp-PLA2 and MetS on First CVD

In age- and sex-adjusted analyses, the area under the ROC curve associated with elevated Lp-PLA2 activity alone (0.71) was similar to MetS alone (0.71). As both elevated (upper tertile) Lp-PLA2 activity and presence of MetS assessed simultaneously in the same model were independently of traditional risk factors associated with incident CVD, we evaluated the potential additive effect of both markers for risk prediction. The cohort was divided into 4 groups based on low-to-mid (tertile 1 plus 2) versus elevated Lp-PLA2 activity levels in combination with and without MetS. The referent group was subjects with low-to-mid Lp-PLA2 activity and no MetS. The combination of both elevated Lp-PLA2 and presence of MetS was, after adjustment for age- and sex, associated with a statistically significantly increased risk for future CVD events (RR: 2.38; 1.67 to 3.41, \( P < 0.001 \)) and was stronger to either elevated marker alone in predicting risk. The RRs for elevated Lp-PLA2 alone and MetS alone were 1.56; 1.16 to 2.08, \( P = 0.003 \), and 1.62; 1.05 to 2.49, \( P = 0.029 \), respectively. After further adjusting for LDL-cholesterol, lipid-lowering treatment, smoking, hsCRP, physical activity, and high alcohol consumption, the association for the combination of high Lp-PLA2 activity and MetS was 1.97 (1.34 to 2.90, \( P = 0.001 \)) compared with the relative risk of 1.40 (1.03 to 1.92, \( P = 0.034 \)) for high Lp-PLA2 activity alone and 1.46 (0.94 to 2.27, \( P = 0.095 \)) for presence of MetS alone.

### Table 3. Mean Levels of Lp-PLA2 Activity and Lp-PLA2 Mass in Relation to No. of Components Involved in the Metabolic Syndrome (MetS)

<table>
<thead>
<tr>
<th>No. of MetS Components</th>
<th>No. of Subjects</th>
<th>Lp-PLA2 Activity (nmol/min/mL)</th>
<th>Lp-PLA2 Mass (ng/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>657</td>
<td>40.8 ± 11.3</td>
<td>252.0 ± 73.5</td>
</tr>
<tr>
<td>1</td>
<td>2082</td>
<td>43.5 ± 11.8</td>
<td>266.3 ± 77.6</td>
</tr>
<tr>
<td>2</td>
<td>1007</td>
<td>46.1 ± 12.9</td>
<td>274.5 ± 82.2</td>
</tr>
<tr>
<td>3</td>
<td>531</td>
<td>50.0 ± 13.6</td>
<td>281.3 ± 84.5</td>
</tr>
<tr>
<td>4 or 5</td>
<td>203</td>
<td>52.5 ± 14.8†‡</td>
<td>276.9 ± 85.5†‡</td>
</tr>
</tbody>
</table>

†\( P \) for trend < 0.001, unadjusted. †\( P \) for trend < 0.001 for activity and 0.472 for mass, respectively, after adjustment for age, sex, and LDL-cholesterol.

### Table 4. Incidence and Estimated Covariate-Adjusted Relative Risk (RR) of First CVD Events (Coronary Events [CHD] or Stroke) by Baseline Lp-PLA2 Activity Level in Tertiles (T1 to T3) During 10 Years of Follow-Up

<table>
<thead>
<tr>
<th>CVD Events</th>
<th>Tertile 1 (n=1493)</th>
<th>Tertile 2 (n=1494)</th>
<th>Tertile 3 (n=1493)</th>
<th>( P ) for Linear Trend</th>
<th>Per SD (12.79)</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of events (CHD/stroke)</td>
<td>53 (23/30)</td>
<td>76 (40/36)</td>
<td>132 (68/64)</td>
<td>\langle 0.001</td>
<td>1.25 (1.11–1.40)</td>
</tr>
<tr>
<td>Incidence/1000 person-years</td>
<td>3.3</td>
<td>4.8</td>
<td>8.5</td>
<td>\langle 0.001</td>
<td>1.25 (1.11–1.40)</td>
</tr>
<tr>
<td>Age and sex-adjusted RR</td>
<td>Reference</td>
<td>1.19 (0.84–1.70)</td>
<td>1.80 (1.29–2.50)</td>
<td>\langle 0.001</td>
<td>1.25 (1.11–1.40)</td>
</tr>
<tr>
<td>Model 1*</td>
<td>Reference</td>
<td>1.12 (0.78–1.61)</td>
<td>1.59 (1.10–2.29)</td>
<td>0.007</td>
<td>1.19 (1.04–1.36)</td>
</tr>
<tr>
<td>Model 2†</td>
<td>Reference</td>
<td>1.11 (0.77–1.60)</td>
<td>1.54 (1.07–2.24)</td>
<td>0.012</td>
<td>1.16 (1.01–1.33)</td>
</tr>
<tr>
<td>Model 3‡</td>
<td>Reference</td>
<td>1.08 (0.75–1.56)</td>
<td>1.46 (1.01–2.13)</td>
<td>0.028</td>
<td>1.12 (0.98–1.29)</td>
</tr>
</tbody>
</table>

*Model 1 adjusted for age, sex, LDL-cholesterol, and lipid-lowering treatment.
†Model 2 adjusted for age, sex, LDL-cholesterol, lipid-lowering treatment, smoking, hsCRP, physical activity, and high alcohol consumption.
‡Model 3 adjusted for age, sex, LDL-cholesterol, lipid-lowering treatment, smoking, hsCRP, physical activity, high alcohol consumption, and the metabolic syndrome.
Discussion

This study provides insights that Lp-PLA2 levels are higher among subjects with than without the MetS. Only 6 of 10 individuals with elevated Lp-PLA2 activity levels defined by the top tertile of the Lp-PLA2 distribution are concordant for elevated Lp-PLA2 mass level, and vice versa. Furthermore, Lp-PLA2 activity is in comparison to Lp-PLA2 mass more strongly correlated to each individual component of MetS, and increased more linearly in the cumulative increase of number of components involved in the syndrome. Presence of elevated Lp-PLA2 activity, but not mass, and MetS were independently associated with an increased risk and a similar discriminatory capacity for incident CVD. Individuals with elevated Lp-PLA2 activity but without MetS and those with MetS but without elevated Lp-PLA2 activity are at increased risk for CVD. However, the combined presence of elevated Lp-PLA2 activity and MetS in this nondiabetic cohort indicated the highest increased risk for CVD.

The addition of both factors, however, did not result in a synergistic effect, as demonstrated by the lack of statistical significant interaction (P=0.22) on a multiplicative scale and the approximate equality of the RR comparing presence of both factors to neither factor (eg, 1.97) with the product of the RRs comparing each factor to neither (eg, 1.46×1.40 resulting in a derived RR of 2.04). However, given the wide 95% CI (1.34 to 2.90) associated for the joint presence of elevated Lp-PLA2 activity and MetS, it is possible that true interaction between these factors cannot be ruled out. Although both Lp-PLA2 activity and MetS are associated with small, dense, highly atherogenic LDL,5,6 they may provide complementary information in risk of CVD events by nature of disparate pathways in the development of atherosclerosis.19

The relationship of Lp-PLA2 activity to MetS in our study extends findings from a small study from Rizos et al20 who found Lp-PLA2 activity levels higher in 60 patients with the MetS compared with 110 matched subjects without MetS.

No other studies have, to our best knowledge, reported on the Lp-PLA2-CVD risk association in subjects with and without MetS. Ridker et al21 studied another inflammatory marker, hsCRP, in relation to CVD risk in women with MetS. However, the risk increase for CVD events associated with Lp-PLA2 activity in our study was not affected by hsCRP. Furthermore, we have previously reported on no relationship (r=0.01) between Lp-PLA2 activity and hsCRP in our cohort.3 Thus, this suggests that these 2 markers may reflect distinctively different mechanisms on the atherosclerotic process.

One possible explanation of the different association between Lp-PLA2 activity and mass levels and MetS in the present study is that the activity of the enzyme, but not mass, has been shown preferentially associated with LDL-cholesterol3,7,8,9 and with the small, dense LDL versus larger particles.5,6 Atherogenic small dense LDL is shown to be a common feature in subjects having MetS.1 Furthermore, subjects in our study who were discordant for elevated levels of the activity of the enzyme in comparison to elevated levels of mass were more likely to have higher levels of triglycerides and lower levels of HDL cholesterol.

Strengths and Limitations

There are several strengths of our study. It is a large, population-based cohort comprising 4480 apparently nondiabetic healthy men and women with a homogenous ethnic composition. Regional and national registers7,18 were used to ascertain cases of CVD. A validation study from the national Hospital Discharge Registry17 demonstrated that the diagnosis “myocardial infarction” was false in only five percent of cases, and another study has showed the validity of the Stroke Registry of Malmo.18

Several limitations should be stated. In this study the 16% prevalence of MetS was somewhat less compared with other similar population-based studies,1,22 which might be explained by a lower attendance rate (42%) in our cohort. Characteristics of participants and nonparticipants in MDCS have been reported separately.23 It has been shown that the cohort is fairly representative with respect to prevalence of overweight and smoking. Although the mortality rate has been demonstrated higher in MDCS nonparticipants,23 the age- and sex-adjusted RR (1.68, 95% CI: 1.28 to 2.21) for CVD events associated with MetS in our nondiabetic cohort was similar to what has been reported in the ARIC study with similar age and sex-distribution.22

The validity of MetS merits discussion. It is well-known that use of a self-administered questionnaire and a single measurement of blood glucose, blood lipid, and blood pressure may overestimate the prevalence of diabetes, hyperlipidemia, and hypertension. In this case, the direction of the bias would be expected toward a dilution of the associations. Change of exposure is an inherent problem in long-term studies. Without reexaminations, it is impossible to know what happens between the baseline examination and outcome. Subjects with newly detected cardiovascular risk factors, eg, hypertension, lipid disorders, etc, at baseline were referred for evaluation and treatment in other clinics. One might assume that these subjects may have a reduced risk; however, excluding patients with pharmacological treatment for hypertension and hyperlipidemia from the analysis revealed a risk increase associated with elevated Lp-PLA2 activity of 1.52, eg, a point estimate almost identical to the model including all these patients (eg, 1.46). In the present study we did adjust for leisure time physical activity and high alcohol consumption. However, it has been proposed that this adjustment would lead to an overadjustment, as physical activity may constitute a link in the chain of event between MetS, its major components, and CVD.24 Additionally, we were unable to examine sex-specific associations in our study attributable to limited number of CVD end points, but this is a relevant question for future studies.

The discrepancy of association between elevated Lp-PLA2, in terms of activity and mass, and incident CVD could be attributable to differences in the methodology for measuring these parameters.

Although similar methods in our study compared with the PROVE-IT TIMI-22 Trial11 were used to measure Lp-PLA2 activity, defined in terms of mass and activity, the correlation between these parameters is much higher in the present study (r=0.57 versus 0.36). Furthermore, the coefficient of variation for intra- and interassay precision of Lp-PLA2 activity and mass, respectively, in the present study was rather similar and
almost identical to what have been reported by others. The fact that better agreement between mass (monoclonal antibody-based) and activity (substrate-based) measurements can be achieved with more extensive solubilization of the enzyme in plasma (ie, to ensure all available enzyme is released from the lipoprotein complex) suggests that the monoclonal antibody-based test and the substrate-based test may not quantify identical populations of the Lp-PLA2 enzyme when it is in association with lipoprotein particles (B. Wolpert, unpublished observations, 2006). We found only a moderate agreement (r=0.29) between Lp-PLA2 activity and mass in tertiles. The highest age- and sex-standardized incident CVD was observed in subjects exposed to presence of both moderate to elevated levels of Lp-PLA2 activity and mass. Future studies are needed to identify factors (eg, genetic, biomolecular, or assay) that could explain the discrepancies between the mass and activity measurements of the enzyme, especially at higher values, and further explore whether information of both measurements increases the possibility to identify high risk individuals for future CVD.

Conclusions
In conclusion, in this population-based nondiabetic cohort Lp-PLA2 is associated with the MetS. Higher levels of plasma Lp-PLA2 activity were related to increased risk for incident CVD regardless of MetS. The simultaneous presence of elevated Lp-PLA2 activity and MetS may identify an especially high risk individual.

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Disclosure
Jeanenne J. Nelson is employed by GlaxoSmithKline (GSK) as a senior epidemiologist.

References
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