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Relationship of Autoantibodies to MDA-LDL and ApoB-Immune Complexes to Sex, Ethnicity, Subclinical Atherosclerosis and Cardiovascular Events

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Abstract

Objective—Modifications of lipid constituents within atherosclerotic lesions generates neoepitopes that activate innate and adaptive immune responses. We aimed to define the prevalence, distribution, and relationship of autoantibody titers of oxidized lipoproteins to subclinical atherosclerosis and major adverse cardiovascular events (MACE) in different ethnic groups.

Approach and Results—IgG and IgM autoantibodies to malondialdehyde(MDA)-LDL and apolipoprotein B-100-immune complexes (ApoB-IC) were measured in 3509 individuals (1814 Blacks, 1031 Whites, 589 Hispanics, 85 no race identifier) from the Dallas Heart Study with median 10.5-year follow-up. Coronary artery calcium score (CAC), abdominal aortic plaque by MRI and MACE were quantified. IgG MDA-LDL and IgG and IgM apoB-IC were significantly different between groups, with Blacks having the highest levels of IgG MDA-LDL and IgG ApoB-IC and Hispanics the highest levels of IgM ApoB-IC ($p<0.001$ for all). IgGs tended to be higher and IgMs lower with age for all markers. In multivariable-adjusted binary logistic regression analysis, a doubling of IgG MDA-LDL levels was associated with prevalent CAC > 10 Agatson Units (OR (95%CI) 1.21 (1.07–1.36, p=0.002). Multivariable-adjusted Cox regression analysis revealed that IgG MDA-LDL was independently associated with time to incident MACE in the entire group (HR (95% CI) 1.76 (1.16–2.72, p=0.009) for 4th vs. 1st quartile). This effect was particularly prominent in Black subjects (HR 2.52 (1.39–4.57), p=0.002).

Conclusion—Autoantibodies to oxidized lipoproteins and immune complexes with apoB-100 lipoproteins vary significantly by sex, age and ethnicity. Higher baseline IgG MDA-LDL titers

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independently associate with new MACE. These findings may contribute to the understanding of differences in ethnic-specific MACE events.

Keywords

Oxidation; biomarkers; lipids; ethnicity; cardiovascular disease; autoantibodies; innate immunity

INTRODUCTION

Cardiovascular disease (CVD) continues to be the major cause of morbidity and mortality in the developed world[1] and is rapidly becoming a key determinant of premature death in developing nations. Known risk factors, such as lipid and inflammatory genes and biomarkers, predict a relatively small fraction of risk and additional insights are needed to identify all determinants of CVD risk.[2] This is particularly true among sexes as well as certain ethnic groups, where similarly potent risk factors do not seem to have the same impact.[1,3–6]

It has been established that atherosclerosis, the main etiological driver of CVD, is primarily a disease of lipid abnormalities with superimposed chronic inflammation. Modification of the lipid constituents within atherosclerotic lesions generates neoepitopes, called danger-associated molecular patterns (DAMPs),[7] that activate innate and adaptive immune responses. These responses initiate a pro-inflammatory response to inactivate and clear such antigens.[8,9] The generation of pro-inflammatory oxidation-specific epitopes (OSE) in vivo is well described.[10]

At the clinical level, one can measure plasma IgG and IgM autoantibodies that target chemically and pathophysiologically well-defined OSE such as malondialdehyde(MDA)-lysine adducts on proteins. IgG and IgM present on apolipoprotein B-100 lipoproteins as part of immune complexes can also be measured.[11,12] In general, elevated plasma levels of IgG autoantibodies to OSE represent responses to antigen exposure and tend to correlate with worse cardiovascular risk. In contrast, IgM autoantibodies to OSE often represent “natural” antibodies present at birth that are likely evolutionarily conserved to protect against such DAMPs present in sites of inflammation, such as apoptotic cells and cell walls of infectious pathogens. These so-called natural antibodies also bind to OSE on modified lipoproteins as the target DAMPs are either identical or act as molecular mimics of those present on apoptotic cells and infectious agents.[9,13] In contrast to IgGs to OSE, IgMs tend to be associated with lower CVD events.[11,12,14,15]

The prevalence and impact on sub-clinical and clinical CVD of such biomarkers of oxidized lipoproteins in different sexes and ethnicities has not been well studied. Because the titers of OSE-autoantibodies are strongly influenced by hereditary,[16] it might be anticipated that sex/ethnic differences might have an important impact on their titers. Therefore, the purpose of the present study is to examine the relationship between IgG and IgM autoantibodies to MDA-LDL and apoB-IC and subclinical atherosclerosis and major adverse cardiovascular events (MACE) over a 10.5-year prospective follow-up in a large, multi-ethnic, population-based epidemiological cohort from the Dallas Heart Study.

Materials and Methods

Materials and Methods are shown in the Supplementary Appendix.

RESULTS

Subject characteristics

The participant characteristics in the entire group combined and by ethnicity are summarized in Table 1. The study consisted of 3509 participants, with a mean age of 43.7±10.1 years and 44.1% of the subjects were male, 1814 (51.7%) were African American, 1021 (29.1%) Caucasian, and 589 (16.8%) Hispanic. Eighty-five subjects did not self-report ethnicity. Significant differences were observed in baseline characteristics between ethnic groups. There was a high prevalence of traditional cardiovascular risk factors in the population; 34.4% hypertension, 11.6% diabetes, and 29.3% current smokers. Notably, Blacks had the lowest total cholesterol, triglycerides, VLDL-C levels, and highest HDL-C levels. LDL-C levels were comparable among the three ethnic groups. Hispanics had a lower prevalence of current smoking, while Blacks had the highest prevalence of hypertension.

Significant differences were also noted among autoantibodies and ApoB-IC, with Blacks having highest levels of IgG MDA-LDL and IgG ApoB-IC and Hispanics having the highest levels of IgM ApoB-IC, measured as relative light units (RLU) (Table 1).

Relationship between sex, age and autoantibody titers to oxidized lipoproteins

In the entire group, median (IQR) IgM MDA-LDL (13525(9546–18954) vs. 16678(12344–22724), p<0.001) and IgM ApoB-IC (3065(2044–4494) vs. 3653(2410–5330), p<0.001) were lower and IgG ApoB-IC (4240(3302–5353) vs. 4113(3208–5232), p=0.018) were higher in males versus females. However, IgG MDA-LDL levels were not different between males and females 5055(3401–7771) vs. 5485(3593–8150), p=0.98). Within ethnic groups, in Blacks, IgM MDA-LDL (p<0.001) were lower and IgG ApoB-IC (p<0.001) were higher in males. In Whites and Hispanics, IgM MDA-LDL (p<0.001 for both) and IgM ApoB-IC (p<0.001 for both) were lower in males.

In the entire group, IgG MDA-LDL and IgG apoB-IC levels tended to be higher with advancing age and IgM MDA-LDL and IgM ApoB-IC tended to be lower with age (Figure 1). Comparing biomarkers within ethnic groups, in Blacks IgG MDA-LDL (p=0.086) and IgG ApoB-IC (p= 0.002) tended to be higher with age, while in contrast IgM MDA-LDL (p = 0.03) and IgM ApoB-IC (p<0.001) were significantly lower with age. In Whites, only IgM ApoB-IC (p<0.001) was lower with age, but in Hispanics both IgG MDA-LDL (p = 0.007) and IgM ApoB-IC (p<0.001) were significantly lower with age (Table 2).

Supplemental Table I shows the median (IQR) levels of the autoantibody titers and apoB-immune complexes in patients according to sex. Women of all ethnicities tended to have higher IgM MDA-LDL and apoB-IC levels. Supplemental Table II shows the median (IQR) levels of autoantibody titers and apoB-immune complexes in patients according to demographic variables. Diabetic subjects had lower levels of IgM MDA-LDL and IgM

ApoB-IC, and subjects with hypertension had higher levels of IgG MDA-LDL but lower levels of IgG ApoB-IC.

Supplemental Table III shows the Spearman correlations between continuous variables. IgM MDA-LDL and IgM apoB-IC were modestly correlated (r=0.61, p<0.001), while weak to modest correlations were present for other variables.

Relationship between biomarkers of oxidized lipoproteins to coronary calcium score and abdominal aortic plaque area

Of the 2740 subjects with a CAC scan, 582 (21.2%) had a CAC score >10. When evaluated as a continuous variable, CAC showed weak but significant positive Spearman correlations with IgG MDA-LDL (r=0.073, p<0.001) and IgG ApoB-IC (r=0.040, p=0.036). In contrast, inverse correlations were noted with CAC score and IgM MDA-LDL (r= −0.107, p<0.001) and IgM ApoB-IC (r= −0.167, p<0.001).

When using CAC >10 as a positive score and CAC <10 as a negative score to remove artifact bias of very low CAC scores, and evaluating the data with binary logistic analysis with multivariable adjustment with the variables in Table 1, (age per decile, male sex, ethnicity, diabetes, current smoking, BMI, LDL-C per SD, HDL per SD, log triglycerides per SD, HTN, and log of the 4 oxidative biomarker variables per SD), the log IgG MDA-LDL per 1 SD OR (5% CI) was 1.21 (1.07–1.36), p=0.002) and log IgG ApoB-IC per 1 SD was 0.88 (0.79–0.99, p=0.026) were independently associated with CAC >10 (Table 3). The p-value for the interaction test of ethnicity (included all 3 groups)*log IgG MDA-LDL per 1 SD was p=0.27. Evaluating these data by ethnicity, similar trends were noted (Supplemental Table IV),

For abdominal aortic atherosclerosis measured as aortic plaque burden by magnetic resonance imaging, 977 (39.5%) out of 2475 had a positive scan. Weak but significant positive correlations were noted with IgG MDA-LDL (r=0.053, p=0.009) and inverse correlations were noted with IgG ApoB-IC (r=−0.048, p=0.017) and IgM ApoB-IC (r= −0.094, p<0.001).

When using plaque area =0 or >0 mm^2 as a binary variable in a multivariable, logistic regression analysis including variables in Table 1, age per decile, diabetes, current smoking, BMI, HDL-C per 10 mg/dL, LDL-C per 25 mg/dL, log_2 triglycerides (1.21 (1.08–1.36, p = 0.001) and log_2 IgG ApoB-IC (0.81 (0.68–0.97, p=0.022), were independent predictors of plaque area >0 mm^2 (Table 3). The p-value for the interaction test of ethnicity (included all 3 groups)*log IgG ApoB-IC per 1 SD was p=0.43. Evaluating these data by ethnicity, generally similar findings were noted (Supplemental Table V).

Relationship of biomarkers of oxidized lipoproteins to cardiovascular outcomes

There were a total of 190 MACE events out of total of 2914 subjects with adjudicated MACE until Dec 31 2011, with 132 (out of 1498 subjects or 8.81%) in Blacks, 44 (out of 911 subjects, or 4.83%) in White and 12 (out of 447 subjects, or 2.68%) in Hispanics. There were 2 events in subjects whose ethnicity was not identified and who were not included in the analyses. Cox regression analysis with time to MACE was performed in the entire group

in 3 models; Model 1: oxidation biomarkers alone; Model 2: plus age in deciles, sex, hypertension, diabetes, current smoking, LDL-C per 1 SD, HDL-C per 1 SD and log triglycerides per 1 SD; and Model 3: plus ethnicity. IgG MDA-LDL were independently associated with time to MACE with an HR (95% CI) of 2.10 (1.40–3.15, p=0.001) for 4th quartile vs. first (Table 4). Further adjustment in model 2 and model 3 attenuated the results slightly but significance was maintained. The ethnicity*quartile IgG MDA-LDL interaction was significant for Q3 (p=0.006) and Q4 (p<0.001) vs Q1. Removing all revascularization events did not appreciably change the HR (95% CI) 1.75 (1.12–2.75, p=0.014) for Q4 vs.Q1 in the fully adjusted model. Figure 2A demonstrates the temporal relationship of time to MACE in the entire group for IgG MDA-LDL autoantibodies. The IgM MDA-LDL, IgG ApoB-IC and IgM ApoB-IC were not significantly different after full adjustment (Table 4). In the model of IgG MDA-LDL including all covariates including ethnicity, addition of IgM MDA-LDL per 1 SD, IgG ApoB-IC per 1 SD, IgM ApoB-IC per 1 SD did not appreciably affect the findings: Q4 HR 1.88 (1.19–2.95), p=0.006.

Similar analyses were performed in individual ethnic groups. In fully adjusted analyses, the results in the entire group were primarily driven by the Black group, which had an HR (95% CI) of 2.52 (1.39–4.57, p=0.002) comparing 4th vs 1st quartile. No significant differences were noted in Whites HR (95% CI) of 1.00 (0.41–2.46, p=1.00) or Hispanics HR (95% CI) of 0.71 (0.20–1.98, p=0.17) (Figure 2B–D).

DISCUSSION

This study demonstrates several novel observations regarding circulating autoantibodies and immune complexes reflecting OSE over a 10.5 year prospective follow-up in the Dallas Heart Study: First, significant differences were present between ethnic groups in several measures, with Blacks having highest levels of IgG MDA-LDL and ApoB-IC, while whites had the lowest levels and Hispanics had the highest levels of IgM ApoB-IC; Second, a significant age effect was noted, with IgGs trending higher and IgMs lower with advancing age; Third, IgG MDA-LDL levels were associated with prevalent CAC; Fourth, elevated IgG MDA-LDL levels were predictive of time to MACE in the entire group, an effect particularly prominent in Black subjects. Remarkably, these findings were present even though individuals were relatively young (mean age ~44 years old) upon entry into the study.

Since the initial description of autoantibodies to oxidized LDL in atherosclerotic lesions and plasma of both animals and humans,[17–19] a large database has been generated on experimental[13,14,20,21] and clinical aspects of such autoantibodies.[11,12,22,23] Furthermore, the research realm has expanded beyond CVD to other inflammatory conditions, such as lupus, rheumatoid arthritis,[24] infections,[25] renal failure[26] and Alzheimer's disease.[27] However, most of these studies have been performed in subjects of European descent and little data is present in other ethnic groups. This is the first dedicated study evaluating autoantibody titers to oxidized lipoproteins and immune complexes with apoB-100 lipoproteins in the context of race and ethnicity. Significant differences in racial/ethnic groups were present, and in turn, higher IgG MDA-LDL titers were associated with higher CAC score and likelihood for MACE. Although the underlying etiology of these differences

cannot be determined from this study, the data suggest that appropriate interpretation of these measurements in epidemiological studies should include race/ethnicity as a variable.

The association of age with oxidative biomarkers is also of interest, particularly since age is universally one of the strongest risk factors for CVD. The DHS has the advantage of evaluating the relationship of age to these biomarkers since it includes a broad range of 18–65 year old subjects. Although age was analyzed in a cross-sectional rather than a prospective analysis in DHS, one can speculate that continued exposure to the various OSE led to increasing IgG titers as subjects age. Conversely, as IgMs are thought to be derived from B-1 cells present at birth or shortly thereafter, a decline in titers to OSE over time may suggest loss of certain B-1 cell subsets, resulting in decreased innate immune responses to chronic diseases caused by OSE and certain infections.[13,21,28–30] Consistent with a genetic influence of circulating autoantibodies to OSE, a recent genome-wide linkage study and genetic association in twins, the heritability (h^2) of IgM MDA-LDL and IgM apoB-IC were 0.69 and 0.80, respectively, and for IgG MDA-LDL and apoB-IC was 0.62 and 0.53, respectively, which was similar or higher than physiological, inflammatory, or lipid traits.[16]

In the current study, IgG MDA-LDL titers were predictive of subclinical atherosclerosis as measured by CAC, which to our knowledge is the first study to evaluate this phenotype with autoantibodies to OSE. This is consistent with some,[12,18,31–33] but not all studies,[34–36] that show similar associations with carotid or peripheral arterial disease. Analyses by ethnicity showed that this association was only significant in Blacks, although the interaction test for ethnicity was not significant but trends across ethnicity were consistent. It is also interesting that Blacks in the United States, as reflected in the Dallas area in this study, have up to 25% European or Native American genes, which further complicates analysis based on race/ethnicity.[37]

This study also showed that elevated levels of IgG MDA-LDL were independently predictive of time to MACE over a median of 10.5 years, which seemed to be primarily driven by the Black cohort. These observations are consistent with the Bruneck study in Whites, where IgG titers to Cu-OxLDL were associated with higher risk of MACE (HR: 1.18; 95% CI: 1.02 to 1.37, p=0.028 for 1-SD unit increase),[12] as well as in other studies.[11,12,38–40] However, these data are not entirely consistent, as other studies have shown no association.[11,36,39,41,42] Additionally, a recent study has shown that low levels of IgG to selected apoB-100 peptides and MDA-modified apoB-100 peptides (which may not reflect IgG titers to MDA epitopes) are associated with higher risk of CVD.[43] In a recent report from the Dallas Heart Study, we reported that lipoprotein(a) [Lp(a)], oxidized phospholipids on apolipoprotein B-100 (OxPL-apoB), apolipoprotein(a) isoforms and LPA snps varied significantly by ethnicity. In turn, elevated Lp(a) and/or OxPL-apoB were independent predictors of MACE, even after adjusting for apo(a) isoforms and LPA snps with consistent results in the 3 ethnic groups.[44]

IgM biomarkers in this study were not predictive of CVD events. In other studies, low levels of IgM autoantibodies to a variety of OSE predict worse outcomes,[12,45] consistent with a potentially atheroprotective effect, which has been consistently observed in experimental studies (reviewed in [9,22]) In DHS, with a decline in IgM levels in more advanced ages,

power may not have been optimal to assess these relationships, compared to prior studies that recruited more elderly individuals.

The underlying reasons for the divergent results in these studies is not clear, but our current observations suggest that the failure to consider ethnic/race of the populations studied may be one such factor. Aside from other biases in the populations studied, study design, methodological assessments of subclinical atherosclerosis endpoints and different endpoints confound these studies. In addition, there are likely multiple technical issues in the way these antibody titers were assessed including differences in assay antigen preparation. It is well known that variations in the extent of oxidation result in different preparations of MDA-LDL, or other MDA modified proteins, which can result in differences in measured autoantibody titers[46,47] The generation of standardized antigens has been suggested to overcome this limitation and standardize assays across laboratories. In that regard, we have described the development of small amino acid mimotopes apparently reflecting the 3-dimensional structure of MDA and validated that they have similar immunological properties of MDA and may be useful in future studies.[48] Ultimately, whether such autoantibody titers to OSE are causally related to CVD or are epiphenomena await genome wide association and other studies evaluating causality. For example, a recent genome-wide association study using the PROMIS database showed that novel variants on chromosome 6 at the HLA-B locus and on chromosome 12 at the CIT locus were associated with circulating IgM titers to MDA-LDL, IgM to phosphocholine-modified bovine serum albumin and ApoB-IC, and these variants were subsequently linked to CVD events in PROMIS, as well as in the CARDIOGRAmplusC4D consortium database.[49] These clinical observations in conjunction with experimental data strongly suggest genetic influences on circulating levels of autoantibody titers to OSE, which may then be reflected as differences in circulating levels among different ethnic groups, and finally to cardiovascular events.

Limitations of this study included that the Hispanic group was small with a relatively small number of events and therefore the study may have been underpowered to evaluate relationships to CVD.

In conclusion, significant age and ethnic-specific difference are present in IgG and IgM autoantibody titers to OSE and ApoB-IC. Elevated IgG MDA-LDL is associated with time to MACE, particularly in Black individuals and may provide some explanations for the disparity in outcomes among different ethnic groups.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Pharmaceuticals. JLW is a consultant for Ionis Pharmaceuticals, Intercept, CymaBay and Prometheus. The other co-authors have no conflicts of interest.

Abbreviations

CAD	Coronary Calcium Score
CVD	Cardiovascular Disease
DAMPs	Danger associated molecular patterns
HR	Hazard Ratio
IgM	Immunoglobulin M
IgG	Immunoglobulin G
MDA-LDL	Malondialdehyde-modified low density lipoprotein
OSE	Oxidation-specific epitopes
ApoB-IC	Apolipoprotein B-100-immune complexes
MACE	Major adverse cardiovascular events

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Highlights

- **I**gG and IgM autoantibodies to malondialdehyde(MDA)-LDL and apolipoprotein B-100-immune complexes (ApoB-IC) were measured in 3509 individuals from the Dallas Heart Study with median 10.5-year follow-up.
- IgG MDA-LDL and IgG and IgM apoB-IC were significantly different between groups, with Blacks having the highest levels of IgG MDA-LDL and IgG ApoB-IC and Hispanics the highest levels of IgM ApoB-IC ($p<0.001$ for all).
- A doubling of IgG MDA-LDL levels was associated with prevalent CAC > 10 Agatson Units (OR (95%CI) 1.21 (1.07–1.36, p=0.002).
- IgG MDA-LDL was independently associated with time to incident MACE in the entire group (HR (95% CI) 1.76 (1.16–2.72, p=0.009) for 4^{th} vs. 1^{st} quartile). This effect was particularly prominent in Black subjects (HR 2.52 (1.39–4.57), p=0.002).
- These findings may contribute to the understanding of differences in ethnic-specific MACE events.

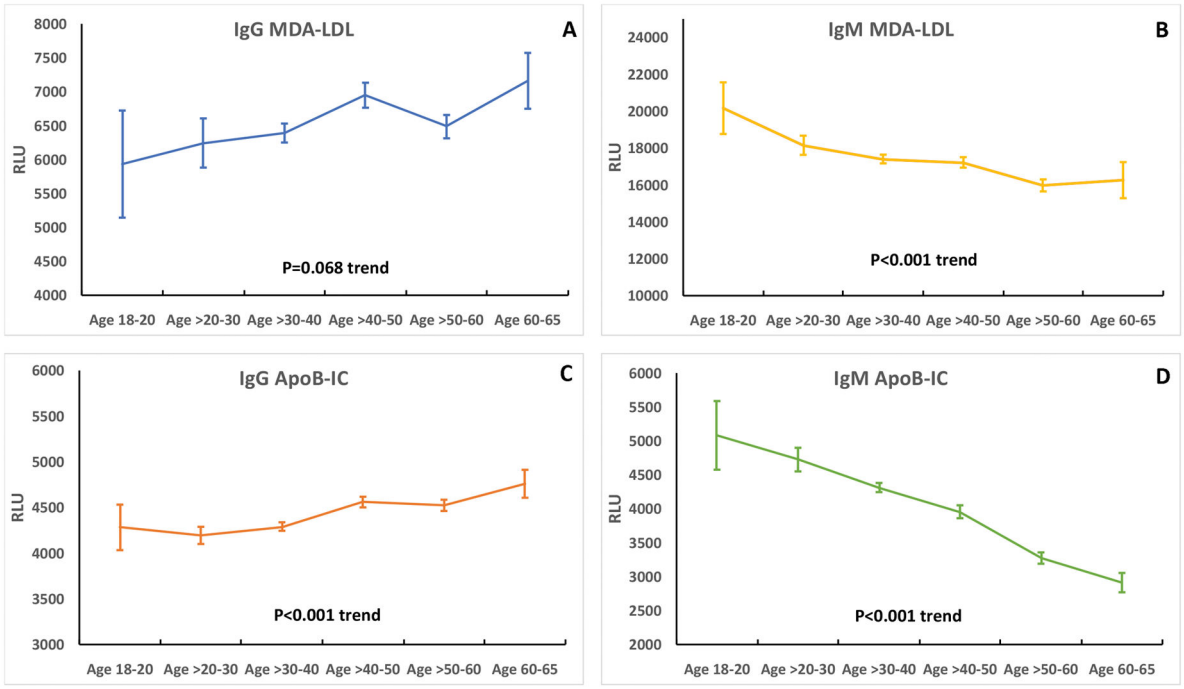

Figure 1.

Relationship of age per decade to autoantibodies to MDA-LDL and apoB-immune complexes in the entire Dallas Heart Study group. A= IgG MDA-LDL, B= IgM MDA-LDL, C= IgG apoB-IC, D= IgM apoB-IC.

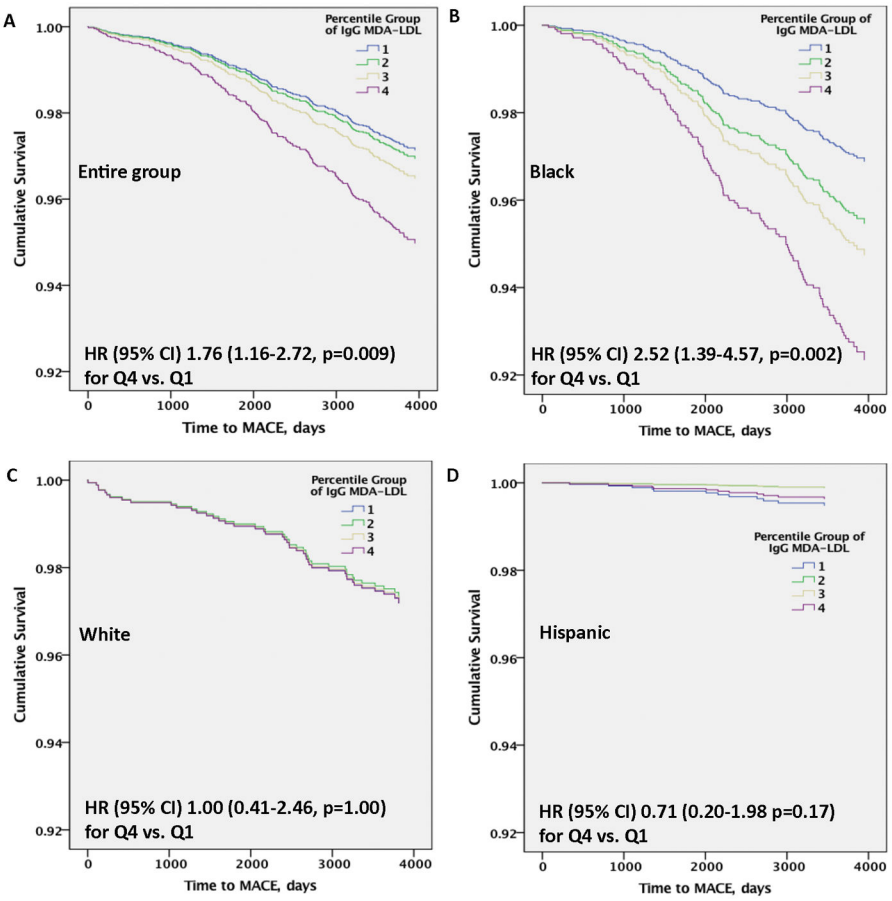

Figure 2.

Multivariable adjusted Cox regression analysis of IgG MDA-LDL with time to MACE over a 10.5-year follow-up in the entire group and by ethnic cohort. A= entire group, B= Black, C= White, D= Hispanic.

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Table 1

Baseline characteristics of study subjects in the entire group and by ethnicity

	Entire Group (N=3509)	Black (N=1814)	White (N=1031)	Hispanic (N=589)	P-Value for ethnicity
Age, years (SD)	43.7 (10.1)	44.5 (10.2)	44.7 (10.0)	40.1 (9.1)	<0.001
Male, N (%)	1546 (44.1)	765 (49.5)	493 (47.8)	244 (41.4)	0.001
BMI (SD)	30.5 (7.6)	31.6 (8.2)	29.0 (6.7)	30.4 (6.6)	<0.001
HTN, N (%)	1177 (34.4) *	679 (38.2)	287 (29.0)	180 (32.6)	<0.001
Diabetes, N (%)	408 (11.6)	258 (14.2)	70 (6.8)	72 (12.2)	<0.001
Current smoking, N (%)	1026 (29.3)	605 (33.4)	288 (28.0)	120 (20.4)	<0.001
Laboratory variables					
Total Cholesterol, mg/dl	180.3 (39.6)	177.7 (40.1)	183.6 (38.4)	181.9 (40.1)	0.001
LDL-C, mg/dl	106.3 (35.4)	104.8 (36.6)	108.3 (34.4)	107.0 (33.4)	0.071
HDL-C, mg/dl	59.8 (14.8)	52.2 (15.3)	48.2 (15.0)	45.7 (11.2)	<0.001
VLDL-C, mg/dl	24.2 (18.8)	20.8 (17.7)	27.1 (18.0)	29.3 (21.5)	<0.001
Triglycerides, mg/dl	96 (67–147)	85 (62–123)	110 (75–167)	118 (81–175)	<0.001
IgG MDA-LDL, RLU	5280 (3522–7969)	6010 (4204–8857)	4179 (2952–6435)	4689 (3094–7247)	<0.001
IgM MDA-LDL, RLU	15355 (10943–21243)	15427 (10738–21119)	15349 (11051–21418)	15357 (11422–21328)	0.64
IgG ApoB-IC, RLU	4179 (3243–5304)	4272 (3281–5382)	4023 3158–5073)	4064 (3246–5252)	<0.001
IgM ApoB-IC, RLU	3391 (2233–4944)	3285 (2172–4971)	3227 (2108–4534(	4056 (2768–5700)	<0.001

75 Individuals either did not have ethnicity identified or identified as “other” and were not included in the ethnicity analysis.

* In the database, of the 3509 individuals, 3419 had an entry for prevalent hypertension.

Table 2

Relationship of Age to Autoantibodies to MDA-LDL and ApoB-Immune Complexes in Different Ethnic Groups

Variable	Age 18–20 (N=33)	Age >20–30 (N=246)	Age >30–40 (N=1157)	Age >40–50 (N=1091)	Age >50–60 (N=816)	Age 60–65 (N=166)	P-Value for age
Black							
IgG MDA-LDL, RLU *	5.2 (3.1–7.8)	6.1 (4.6–8.5)	5.8 (4.0–8.4)	6.5 (4.4–9.5)	5.8 (4.1–8.9)	7.0 (4.6–9.1)	0.086
IgM MDA-LDL, RLU *	19.2 (13.4–26.4)	16.7 (13.1–22.9)	16.3 (11.4–21.3)	15.2 (10.8–21.5)	14.4 (9.5–19.8)	13.9 (8.7–19.8)	0.03
IgG ApoB-IC, RLU *	4.4 (2.9–5.2)	4.1 (3.2–5.3)	4.0 (3.2–5.1)	4.4 (3.3–5.5)	4.4 (3.2–5.6)	4.5 (3.4–5.8)	0.002
IgM ApoB-IC, RLU *	4.4 (2.7–7.2)	3.7 (2.9–5.7)	3.8 (2.5–5.6)	3.2 (2.2–5.1)	2.8 (1.6–3.4)	2.2 (1.6–3.4)	<0.001
White							
IgG MDA-LDL, RLU *	3.7 (3.3–6.4)	3.8 (2.8–5.8)	4.3 (3.0–6.3)	4.0 (2.9–7.1)	4.1 (2.9–5.8)	5.5 (3.2–8.2)	0.11
IgM MDA-LDL, RLU *	16.9 (14.4–25.1)	16.9 (10.7–23.3)	16.4 (11.5–22.1)	15.6 (11.1–22.0)	14.4 (10.4–20.2)	12.3 (9.2–18.3)	0.16
IgG ApoB-IC, RLU *	3.9 (2.8–4.5)	3.8 (3.1–4.5)	3.9 (3.0–4.8)	4.1 (3.2–5.3)	4.2 (3.3–5.1)	4.5 (3.7–5.4)	0.068
IgM ApoB-IC, RLU *	4.5 (3.6–5.0)	3.8 (2.9–5.7)	3.7 (2.6–4.9)	3.2 (2.2–4.6)	2.7 (1.6–4.0)	2.3 (1.6–3.6)	<0.001
Hispanic							
IgG MDA-LDL, RLU *	2.9 (−)	4.6 (3.3–6.8)	4.8 (3.2–7.4)	4.5 (2.8–6.6)	5.2 (3.3–8.4)	5.8 (3.4–7.8)	0.28
IgM MDA-LDL, RLU *	17.2 (−)	16.3 (12.0–22.0)	16.3 (12.8–22.4)	15.5 (10.2–20.8)	12.7 (9.7–17.3)	10.7 (7.2–16.8)	0.007
IgG ApoB-IC, RLU *	6.1 (−)	3.9 (3.2–4.9)	4.0 (3.2–5.2)	4.2 (43.2–5.3)	4.1 (3.3–5.7)	3.9 (3.0–6.0)	0.17
IgM ApoB-IC, RLU *	4.5 (−)	4.4 (2.9–6.5)	4.5 (3.1–6.2)	3.9 (2.4–5.4)	3.0 (2.2–4.2)	3.9 (1.7–4.3)	<0.001

* Values represent RLU X 1000. The Hispanic age group 18–20 years old had only 2 subjects.

Table 3

Multiple logistic regression analysis for the presence or absence of coronary artery calcium and the presence or absence of abdominal aortic plaque in the entire group

	Odds ratio	**95% CI**		
CAC		**Lower**	**Upper**	**P-Value**
Age per decile	4.29	3.68	5.01	<0.001
Male sex	3.33	2.58	4.30	<0.001
Black	1.08	0.83	1.40	0.56
White	0.79	0.55	1.13	0.20
Hispanic	0.58	0.96	1.52	0.25
Hypertension	1.21	0.96	1.52	0.11
Diabetes	2.05	1.49	2.81	<0.001
Current smoking	2.48	1.94	3.18	<0.001
BMI	1.03	1.01	1.05	0.006
LDL-C per SD	1.08	0.97	1.21	0.14
HDL-C per SD	1.02	0.90	1.15	0.81
Triglycerides per SD	1.10	0.97	1.24	0.14
IgG MDA-LDL per SD	1.21	1.07	1.36	0.002
IgM MDA-LDL per SD	1.00	0.87	1.15	0.99
IgG ApoB-IC per SD	0.88	0.79	0.99	0.026
IgM ApoB-IC per SD	1.05	0.91	1.21	0.48
	Odds ratio	95% CI		
Abdominal Plaque		Lower	Upper	P-Value
Age per decile	2.27	2.04	2.52	<0.001
Male sex	1.07	0.88	1.30	0.52
Black	0.84	0.67	1.05	0.12
White	0.91	0.69	1.20	0.52
Hispanic	1.36	0.72	1.56	0.34
Hypertension	0.94	0.79	1.16	0.55
Diabetes	1.41	1.08	1.95	0.025
Current smoking	2.12	1.73	2.61	<0.001
BMI	0.97	0.96	0.99	<0.001
LDL-C per SD	1.09	0.99	1.19	0.076
HDL-C per SD	0.86	0.78	0.96	0.006
Triglycerides per SD	1.19	1.08	1.32	0.001
IgG MDA-LDL per SD	1.08	0.98	1.19	0.13
IgM MDA-LDL per SD	1.10	0.97	1.24	0.13
IgG ApoB-IC per SD	0.89	0.81	0.98	0.019
IgM ApoB-IC per SD	0.93	0.80	1.05	0.23

Table 4

Cox Regression Hazard Ratios for MACE According to Quartiles of IgG and IgM Autoantibodies to MDA-LDL and ApoB-IC.

	Quartile 1	Quartile 2	Quartile 3	Quartile 4	P-value Q1 vsQ4
Model 1: IgG MDA-LDL	1	1.25 (0.82–1.95)	1.38 (0.89–2.12)	2.10 (1.40–3.15)	<0.001
Model 2: Plus age in deciles, sex, HTN diabetes, smoking, BMI, LDL-C per SD, HDL-C per SD, log TG per SD	1	1.13 (0.72–1.78)	1.29 (0.83–2.01)	1.97 (1.30–2.99)	0.001
Model 3: Plus Ethnicity (Black vs others)	1	1.07 (0.67–1.70)	1.23 (0.79–1.94)	1.76 (1.16–2.72)	0.009
Model 1: IgM MDA-LDL	1	0.70 (0.48–1.01)	0.62 (0.42–0.92)	0.58 (0.39–0.86)	0.007
Model 2: Plus age in deciles, sex, HTN diabetes, smoking, BMI, LDL-C per SD, HDL-C per SD, log TG per SD	1	0.91 (0.62–1.33)	0.92 (0.63–1.36)	0.96 (0.63–1.45)	0.84
Model 3: Plus Ethnicity (Black vs others)	1	0.92 (0.62–1.36)	0.90 (0.61–1.34)	0.95 (0.63–1.44)	0.81
Model 1: IgG ApoB-IC	1	0.74 (0.49–1.12)	1.02 (0.69–1.50)	1.04 (0.71–1.53)	0.83
Model 2: Plus age in deciles, sex, HTN diabetes, smoking, BMI, LDL-C per SD, HDL-C per SD, log TG per SD	1	0.92 (0.60–1.41)	1.22 (0.82–1.80)	1.08 (0.73–1.60)	0.71
Model 3: Plus Ethnicity (Black vs others)	1	0.97 (0.63–1.49)	1.20 (0.81–1.78)	1.07 (0.72–1.61)	0.730
Model 1: IgM ApoB-IC	1	0.66 (0.46–0.95)	0.60 (0.41–0.88)	0.47 (0.31–0.71)	<0.001
Model 2: Plus age in deciles, sex, HTN diabetes, smoking, BMI, LDL-C per SD, HDL-C per SD, log TG per SD	1	0.87 (0.60–1.27)	0.96 (0.65–1.42)	1.04 (0.67–1.60)	0.88
Model 3: Plus Ethnicity (Black vs others)	1	0.83 (0.561.21)	0.95 (0.64–1.41)	1.00 (0.64–1.55)	1.00