

# RACIAL DIFFERENCES OF LIPOPROTEIN SUBCLASS DISTRIBUTIONS IN POSTMENOPAUSAL WOMEN

**Background:** We assessed racial differences in lipoprotein particle size, a marker of atherosclerosis risk, among women with coronary disease.

**Methods:** We studied 378 women (33% non-White, predominantly African American) at the baseline visit of the Women's Angiographic Vitamin and Estrogen Trial (WAVE), a multicenter trial of hormone replacement and antioxidant vitamin therapy in postmenopausal women with established coronary artery disease. Average particle sizes for high-density lipoprotein (HDL), low-density lipoprotein (LDL), and very low-density lipoprotein were measured by nuclear magnetic resonance in these women, and angiography was performed at baseline and followup.

**Results:** Adjusted for age, race, diabetes, smoking, blood pressure, and use of lipid-lowering and antihypertensive medications, non-White women had larger LDL particle size (difference .2 nm, 95% CI .1–.3 nm) and HDL particle size (difference .2 nm, 95% CI .1–.2 nm). Neither angiographic disease progression nor survival without myocardial infarction (median follow-up time of 2.8 years) was associated with lipoprotein particle size or race.

**Conclusions:** Non-White women have a less atherogenic profile of lipoprotein particle sizes than do White women. However, this difference did not affect event-free survival or angiographic progression of coronary atherosclerosis. (*Ethn Dis.* 2008;18:176–180)

**Key Words:** Lipoprotein Subclass Distributions, Race Differences, Postmenopausal Period

From the Department of Medicine, Johns Hopkins University School of Medicine, Baltimore, Maryland (AV, PO, DV); Department of Medicine, Division of Cardiovascular Disease, University of Alabama at Birmingham, Birmingham, Alabama (VB); San Francisco General Hospital, San Francisco, California (DW), USA; Department of Medicine, Montreal Heart Institute, University of Montreal, Montreal, Quebec, Canada (JCT).

Address correspondence and reprint requests to: Dhananjay Vaidya, Assistant Professor of Medicine; 1830 East Monument St, Rm 8028-A; Johns Hopkins Medical Institutions; Baltimore, MD 21287; 410-955-7781; 410-955-0321 (fax); dvaidya1@jhmi.edu

Amit N. Vora, BS; Pamela Ouyang, MB, BS; Vera Bittner, MD; Jean-Claude Tardif, MD; David D. Waters, MD; Dhananjay Vaidya, PhD

## INTRODUCTION

During metabolism of lipoproteins, lipid components are added and removed by the liver, in peripheral organs and by enzymes in circulation, which results in particles of different density and size.<sup>1</sup> Smaller, denser low-density lipoprotein (LDL) particles, also known as “phenotype B,” have been shown to confer more atherogenic risk than do their larger, more buoyant counterparts in some but not all studies.<sup>2–5</sup> This risk is thought to result from a greater susceptibility to oxidative modification.<sup>6</sup> Small, dense LDL particles are also usually associated with higher triglyceride and lower high-density lipoprotein (HDL) cholesterol levels, and lipoprotein particle size distribution is affected by insulin resistance.<sup>7,8</sup> Lipoprotein subclass differences may partially account for the widely varying coronary artery disease (CAD) incidence in patients with similar classical lipid profiles.<sup>9,10</sup>

In addition to the phenotype B pattern and its increased risk of CAD, other lipoprotein subclass measures can also help assess disease risk. Large, buoyant HDL particles protect against coronary atherosclerosis,<sup>11</sup> while increasing concentrations of small HDL or large very low-density lipoprotein (VLDL) particles correlate with disease.<sup>10</sup> Racial and sex differences have been noted with respect to lipoprotein patterns; Blacks tend to have higher HDL cholesterol,<sup>12,13</sup> lower LDL cholesterol,<sup>14,15</sup> and lower triglyceride levels,<sup>14–16</sup> than do Whites, and women tend to have higher HDL cholesterol and lower triglyceride levels than do men.<sup>17</sup> The Studies of a Targeted Risk Reduction Intervention through Defined Exercise (STRRIDE) study

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showed that, compared to Blacks, Whites had significantly more intermediate-density lipoprotein (IDL), small LDL, medium VLDL, and large VLDL with less large LDL.<sup>18</sup>

Whether these racial differences persist in older women with CAD is not known. We examined the differences in lipoprotein subclasses by race in postmenopausal women with known CAD enrolled in the Women's Angiographic Vitamin and Estrogen (WAVE) trial and whether these differences underlay any racial differences in angiographic progression or the risk of incident myocardial infarction or death.

## Methods

### Subjects and Study Design

The WAVE trial (1997–1999) was a randomized, double-blind, placebo-controlled study designed to test whether hormone therapy or antioxidant vitamins (vitamins C and E) could prevent angiographic CAD progression.<sup>19</sup> The study design, 2×2 factorial randomization to hormone therapy or placebo and active vitamins C and E or placebo, has been previously de-

scribed.<sup>20</sup> Women were eligible if they were postmenopausal as defined by either bilateral oophorectomy, age 45–55 years with follicle-stimulating hormone levels >40 mIU/mL, or age >55 years and if a protocol angiogram performed within 4 months of the start date showed a 15%–75% stenosis in at least one coronary artery. Exclusion criteria included hormone therapy within the past 3 months, concurrent use of vitamins C and E, history of breast cancer, endometrial cancer, uncontrolled diabetes or hypertension, prior recent myocardial infarction, planned or prior coronary artery bypass graft, fasting triglyceride level >500 mg/dL, creatinine >2 mg/dL, symptomatic gallstones, New York Heart Association class IV heart failure or known ejection fraction <25%, history of pulmonary embolism, deep venous thrombosis, or history of osteoporosis.

The principal results for this trial that showed no association of either treatment with angiographic progression have been published.<sup>19</sup> For our analysis, we used a subset of 378 women, for whom lipoprotein analysis by nuclear magnetic resonance and risk factor assessment at baseline were available, using a partially deidentified dataset obtained from the WAVE data coordinating center. This dataset is now available through the National Heart, Lung, and Blood Institute (<http://www.nhlbi.nih.gov/resources/deca/descriptions/wave.htm>).

### Lipoprotein Analyses

Lipoprotein particle sizes and concentrations were analyzed using nuclear magnetic resonance spectroscopy, which calculates lipoprotein subclass concentrations by deconvoluting the plasma nuclear magnetic resonance spectrum.<sup>21–24</sup> Nuclear magnetic resonance lipoprotein measurement shows high correlation with gradient gel electrophoresis.<sup>9,10,25</sup> For this analysis, we analyzed concentrations of three subclasses of HDL (small 7.3–8.2 nm,

medium 8.2–8.8 nm, large 8.8–13.0 nm), three subclasses of LDL (small 18.3–19.7 nm, medium 19.8–21.2 nm, large 21.3–23.0 nm), IDL (23–27 nm), and three classes of VLDL (small 27–35 nm, medium 35–60 nm, large 60–200 nm). Variables included in confirmatory analysis included mean lipoprotein particle size.

### Statistical Analysis

We tabulated the age, cardiovascular risk factors, and medication use in Whites and non-Whites. Non-Whites were predominantly African American in WAVE (84%);<sup>19</sup> however, the non-White racial group was not further described in the public-use dataset. Data were reported as the mean plus or minus standard deviation or absolute number and percentage, and differences by race were tested by using *t* tests or  $\chi^2$  tests.

Concentrations of lipoprotein subclasses were not normally or log-normally distributed. We plotted the median and interquartile ranges for the subfraction concentrations and tested differences by race by using nonparametric rank sum tests. For adjusted regression analysis, we determined bias-corrected, empirically derived 95% confidence intervals of differences of lipoprotein concentrations using bootstrapped regression models with 1000 iterations.

We assessed racial differences in the hazard of the first event of death or myocardial infarction by using Cox proportional hazard regression models adjusting for age, diabetes status, and ever smoking (smoked >100 cigarettes during lifetime). We tested for racial differences in angiographic disease (minimum lumen diameter of coronary arteries) at study followup, adjusting for baseline diameter, age, diabetes status, follow-up time, and ever smoking by using generalized estimating equations to account for intrasubject correlations because multiple arterial segments were measured during angiography. Further models for survival and angiographic

progression were estimated by adding mean particle size for HDL, LDL, and VLDL in separate regression analyses to assess if doing so changed the association of outcomes with race. All statistical analyses were performed with Stata 8 (StataCorp LP, College Station Texas).

### Results

White and non-White women had similar age distributions and medication use (Table 1). Non-White women were more obese and had more diabetes than did White women, but they were less likely to have smoked in their lifetimes. In the baseline traditional lipid profile, total and LDL cholesterol levels did not differ by race; however non-Whites had higher HDL cholesterol and lower triglyceride levels than did Whites.

The non-White women had a significantly higher concentration of large HDL and large LDL, while White women had a greater concentration of medium HDL, small LDL, large VLDL, and medium VLDL (Figure 1)

All findings were significant in regression analyses adjusting for age, body mass index, systolic and diastolic blood pressure, smoking, diabetes, and the use of lipid-lowering and antihypertensive medications (Table 2). Consistent with these subclass distributions, in confirmatory analyses (data not shown) non-Whites had larger mean HDL and LDL particle sizes and smaller VLDL particle sizes.

We found no racial difference in the hazard of myocardial infarction or death (relative hazard of non-Whites vs Whites 1.48, *P*=.35) or angiographic progression (.01 mm greater minimal lumen diameter at followup among non-Whites vs Whites, *P*=.65). These associations remained nonsignificant in models that included the mean lipoprotein particle sizes for HDL, LDL, and VLDL. Furthermore, the variables for mean particle size were not significantly

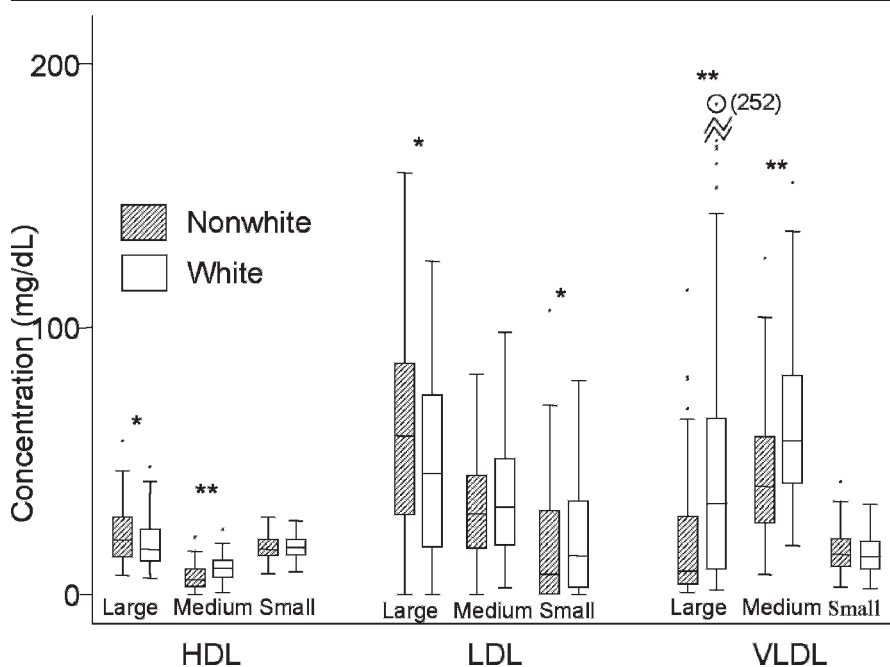
**Table 1. Characteristics of the study sample**

	Mean $\pm$ Standard Deviation or <i>n</i> (%)		
	Non-White ( <i>n</i> =122)	White ( <i>n</i> =256)	<i>P</i> value
<b>Baseline Characteristics</b>			
Age (years)	65.0 $\pm$ 8.6	65.4 $\pm$ 8.5	.69
BMI (kg/m <sup>2</sup> )	31.9 $\pm$ 6.3	30.1 $\pm$ 5.9	.01
SBP (mm Hg)	139.9 $\pm$ 19.6	138.3 $\pm$ 19.3	.45
DBP (mm Hg)	77.9 $\pm$ 10.1	74.4 $\pm$ 10.0	<.001
Diabetic	61 (50%)	74 (29%)	<.001
Lipid-lowering drugs	65 (53%)	159 (62%)	.10
Antihypertensive drugs	112 (92%)	232 (91%)	.71
Smoked >100 cigarettes in lifetime	70 (57%)	177 (69%)	.03
Baseline MLD (mm)†	2.3 $\pm$ .85	2.14 $\pm$ .73	.08
<b>Traditional Lipid Profile Measures</b>			
Total cholesterol (mg/dL)	199.71 $\pm$ 47.83	199.99 $\pm$ 39.93	.95
HDL cholesterol (mg/dL)	52.64 $\pm$ 14.2	49.06 $\pm$ 11.65	.01
LDL cholesterol (mg/dL)	121.45 $\pm$ 41.06	116.49 $\pm$ 36.55	.24
Triglyceride (mg/dL)	125.60 $\pm$ 56.28	176.60 $\pm$ 93.49	<.001
<b>Selected Follow-up Measures</b>			
Events (death or MI)*	11 (9%)	14 (5%)	.19
Follow-up MLD (mm)†	2.3 $\pm$ .76	2.11 $\pm$ .69	.04

BMI = body mass index, SBP = systolic blood pressure, DBP = diastolic blood pressure, MLD = minimal luminal diameter, MI = myocardial infarction.

\* Follow-up for events was available in 256 White and 122 non-White women.

† Angiographic data were available in 194 White and 88 non-White women.



**Fig 1. Distributions of lipoprotein subclass concentrations by race. Size designations: HDL (small, 7.3–8.2 nm; medium 8.2–8.8 nm; large 8.8–13 nm), LDL (small, 18.3–19.7 nm; medium 19.8–21.2 nm; large 21.3–23 nm), VLDL (small, 27–35 nm; medium 35–60 nm; large 60–200 nm) \**P*<.05; \*\**P*<.001**

related to survival or angiographic progression in these models.

## DISCUSSION

This study is the first to analyze racial differences in lipoprotein subclass distribution in a sample of postmenopausal women with angiographically proven CAD. In this sample, non-White women had a higher concentration of large HDL and large LDL, while the White women had a greater concentration of medium HDL, small LDL, large VLDL, and medium VLDL after adjusting for age, body mass index, blood pressure (systolic and diastolic), smoking history, diabetes status, use of lipid lowering agents, and use of antihypertensive medications. However, our analyses showed no difference in death or myocardial infarction or angiographic disease progression between Whites and non-Whites attributable to these lipoprotein subfraction differences in WAVE.

In the US population, White women have a more traditionally atherogenic lipid profile, including higher total cholesterol and LDL and lower HDL, than do African American women.<sup>26</sup> Despite a worse lipid profile, White women have a lower prevalence of cardiovascular disease (35.0% vs 49.2%), CAD (6.0% vs 7.8%), and myocardial infarction (2.5% vs 3.3%).<sup>26</sup> We speculated that this difference may be due to a more atherogenic lipid subfraction profile in African Americans. Because non-Whites in WAVE were predominantly African American, our analyses suggest that the difference in CAD prevalence does not lie in the subclasses. In WAVE, compared to Whites, non-Whites had a worse risk factor profile and similar severity of angiographic disease but less atherogenic lipoprotein subclass distribution.

This study shows that lipoprotein subclass distributions may highlight

**Table 2. Adjusted difference in lipoprotein subclass concentrations in non-Whites compared to Whites**

Lipoprotein Measure	Difference	95% Confidence Interval
High-density lipoprotein		
Large (mg/dL)	4.4	1.9 to 6.8*
Medium (mg/dL)	-3.0	-4.1 to -1.7*
Small (mg/dL)	-.4	-1.5 to 0.7
Particle size (nm)	.2	.1 to .2*
Low-density lipoprotein		
Large (mg/dL)	13.9	5.1 to 21.7*
Medium (mg/dL)	-7.6	-12.6 to -2.3*
Small (mg/dL)	-1.7	-7.1 to 4.2
Particle size (nm)	.2	.1 to .3*
Very low-density lipoprotein		
Large (mg/dL)	-30.6	-39.5 to -21.4*
Medium (mg/dL)	-21.1	-27.1 to -13.9*
Small (mg/dL)	.8	-1.2 to 2.8
Particle size (nm)	-4.5	-5.8 to -3.0*

\*  $P < .05$ , adjusted for age, body mass index, systolic and diastolic blood pressure, smoking, diabetes, and the use of lipid-lowering and antihypertensive medications.

different facets of lipoprotein metabolism than does the traditional lipid profile. In our sample, while the classical lipid profiles differed between the races in the same manner as the broader US population, further racial differences were observed in subclass distributions. Some reports have shown that lipoprotein subfractions may confer a differential risk pattern.<sup>5,10,16</sup> Large, buoyant HDL particles have been shown to be cardioprotective and anti-atherosclerotic in some studies.<sup>9,11,27,28</sup> Some studies have also noted that increasing concentrations of small HDL and large VLDL are positively associated with worsening CAD.<sup>10</sup> Our data showed that the non-White women had a greater concentration of large HDL. Our study showed no significant difference between the races in small HDL and a very significant difference in large VLDL concentration, with a much higher VLDL in White women. This finding confirms those of the STRIDE study, which noted no significant difference in small HDL but a very significant difference in large VLDL.<sup>18</sup>

The racial breakdown for non-Whites was not available in the dataset. However, based on the original publi-

cation,<sup>19</sup> non-Whites were predominantly (84%) African American, and we are confident that the racial differences we have reported pertain predominantly to differences between African Americans and Whites. The small sample size of WAVE with respect to events and angiographic progression reduces our ability to detect racial differences in longitudinal outcomes attributable to their differing lipoprotein subfraction profiles at baseline. The study sample is limited to postmenopausal women with angiographically proven CAD; thus, our results may not be applicable to younger women with CAD or women of any age without CAD. Furthermore, women with severe hypertriglyceridemia were excluded from the WAVE trial, so our analyses cannot determine whether ethnic differences in lipoprotein subfractions and disease progression exist in such women.

In summary, after adjusting for major cardiovascular risk factors, non-Whites have a significantly less atherogenic lipoprotein particle and subclass profile than do Whites. In our sample with established coronary disease, however, this difference did not confer any advantage in event-free survival or

*...after adjusting for major cardiovascular risk factors, non-Whites have a significantly less atherogenic lipoprotein particle and subclass profile than do Whites. In our sample with established coronary disease...*

angiographic progression of coronary atherosclerosis. The underlying racial/ethnic differences in lipoprotein particle and subclass profile should be taken into account if lipoprotein subclass analyses are performed for risk assessment.

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## AUTHOR CONTRIBUTIONS

*Design concept of study:* Vora, Ouyang, Vaidya

*Acquisition of data:* Ouyang, Bittner, Tardif, Waters

*Data analysis and interpretation:* Vora, Ouyang, Bittner, Tardif, Waters, Vaidya

*Manuscript draft:* Vora, Ouyang, Bittner, Vaidya

*Statistical expertise:* Vora, Vaidya

*Acquisition of funding:* Ouyang, Bittner, Tardif, Waters

*Administrative, technical, or material assistance:* Ouyang, Bittner, Tardif, Waters

*Supervision:* Ouyang, Vaidya