High-Density Lipoprotein Cholesterol and Particle Concentrations, Carotid Atherosclerosis and Coronary Events: The Multi-Ethnic Study of Atherosclerosis

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Abstract

Objectives—To evaluate independent associations of high density lipoprotein cholesterol (HDL-C) and particle (HDL-P) concentrations with carotid intima-media thickness (cIMT) and incident coronary heart disease (CHD).

Background—HDL-C is inversely related to CHD, but also to triglycerides, LDL particles (LDL-P), and related metabolic risk. HDL-P associations with CHD may be partially independent of these factors.

Methods—In a multi-ethnic study of 5598 men and women ages 45-84, without baseline CHD, excluding subjects on lipid-lowering medications, triglycerides >400 mg/dl or missing values, we evaluated associations of HDL-C and NMR-spectroscopy-measured HDL-P with cIMT and incident CHD (myocardial infarction, CHD death, angina, n=227 events, 6.0 years mean follow-up). All models were adjusted for age, sex, ethnicity, hypertension and smoking.

Results—HDL-C and HDL-P correlated with each other (π=0.69) and LDL-P (π = −0.38, −0.25, respectively), p<0.05 for all. For (1-SD) higher HDL-C (15 mg/dl) or HDL-P (6.64 μmol/l), cIMT differences (95%CI) were −26.1(−34.7,−17.4) and −30.1 (−38.8,−21.4) μm, and CHD hazard ratios (HR (95%CI)) were 0.74 (0.63, 0.88) and 0.70 (0.59, 0.82), respectively. Adjusted for each other and LDL-P, HDL-C was no longer associated with cIMT (2.3 (−9.5, 14.2) μm) or CHD (0.97(0.77, 1.22)), but HDL-P remained independently associated with cIMT (−22.2(−33.8,−10.6) μm) and CHD (0.75 (0.61, 0.93)). Interactions by sex, ethnicity, diabetes and high-sensitivity C-reactive protein were not significant.

Conclusions—Adjusting for each other and LDL-P substantially attenuated associations of HDL-C, but not HDL-P, with cIMT and CHD. Potential confounding by related lipids or lipoproteins should be carefully considered when evaluating HDL-related risk.

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Keywords

Lipids; lipoproteins; high-density lipoprotein cholesterol; high-density lipoprotein particles; cardiovascular disease

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Introduction

There is great interest in raising levels of high-density lipoprotein cholesterol (HDL-C), given its well-established inverse association with atherosclerosis and coronary heart disease (CHD).(1) However, quantification of HDL-C, the cholesterol carried by HDL particles, may not fully capture HDL-related risk.(1,2) For example, some forms of genetically low, (3) or genetically high, HDL-C(4) do not correspond to expected differences in CHD risk. Recent failures of drugs that raised HDL-C without reducing cardiovascular (CVD) events (5,6) or atherosclerosis(7) have also fueled interest in alternative indices of HDL quantity, i.e., HDL particles (HDL-P) or apolipoprotein A-I (apoA-I), or possibly HDL "quality," i.e., particle size, subclass distribution,(8) or various measures of HDL functionality.(2)

The association of HDL-C with CHD risk is complicated by the inverse association of HDL-C with triglycerides, insulin resistance, obesity, high-sensitivity c-reactive protein (hsCRP), and atherogenic lipoprotein particles, i.e., apolipoprotein B (apoB) and LDL particle concentration (LDL-P).(1) Recent reports showed that adjusting for apoB and apoA-I abolished the inverse association of HDL-C with CHD risk,(9) but HDL-P remained inversely associated with CHD, adjusted for apoB, triglycerides, and HDL particle size.(10) Therefore, we hypothesized that HDL-C associations with carotid intima-media thickness (cIMT) and incident CHD events partly reflect correlated lipid, apolipoprotein or lipoprotein concentrations, particularly LDL-P, but that HDL-P associations are less affected by these metabolic risk factors, including HDL-C. Since HDL functionality has been reported to be altered in diabetes, (11,12), with inflammation,(13), by ethnicity(14), or sex,(15) we also evaluated potential interactions by baseline diabetes, hsCRP, sex and ethnicity.

Methods

Participants and Risk Factor Measurement

Participants eligible for the current study were 6814 men and women enrolled at baseline (2000-2002) in the NHLBI-sponsored multi-center community-based cohort, MESA, the design and objectives of which have been described.(16) Briefly, MESA participants were community-dwelling men and women ages 45-84, of African-American, Hispanic, White, and Chinese-American ethnicity. Baseline exclusion criteria included self-reported CVD (heart attack, angina, coronary or any other arterial revascularization procedure, pacemaker or defibrillator implantation, valve replacement, heart failure or cerebrovascular disease), pregnancy, cancer, cognitive impairment, or weight >136 kg. The current study excluded participants with baseline lipid-lowering medication use (n=1100), triglycerides >400 mg/dl (n=57), or missing values for lipid-lowering medication use, HDL-C, HDL-P, or smoking (n=59). Of the remaining 5598 participants, cIMT was missing for 57 (1.0%) and incident CHD data was missing for 1, leaving 5541 for the cIMT analyses and 5597 for the CHD analyses. Participants provided informed written consent at their field centers. The study was approved by the institutional review boards of the participating institutions and the University of Pittsburgh.

Height, weight, blood pressure, and medications were collected at the baseline MESA examination. Smoking was defined as never, former (smoked 100 cigarettes in lifetime) or current (smoked cigarettes in last 30 days). Hypertension was defined as systolic blood

pressure   140 mmHg, or diastolic blood pressure   90 mmHg, or self-reported hypertension and anti-hypertensive medication use. Hormone therapy use was defined as current user (yes/no.) Type 2 diabetes was defined as fasting glucose >125 mg/dL or use of anti-diabetic medication. HOMA-IR (homeostasis model assessment of insulin resistance) was calculated as insulin (μIU/L) $\times$ (glucose [mg/dL] $\times$ 0.055)/22.5,(17) for those not on anti-diabetic medication.

Lipid, Lipoprotein and other Laboratory Assays

Blood was drawn following a 12 hour fast, and samples were stored at -70 degrees C. Lipids, insulin and glucose were measured at a central laboratory (Collaborative Studies Clinical Laboratory at Fairview-University Medical Center, Minneapolis, MN). Lipids were assayed on thawed EDTA plasma within 2 weeks of the sample collection, using CDC/NHLBI standards. HDL-C was measured using the cholesterol oxidase method (Roche Diagnostics) after precipitation of non-HDL-C with magnesium/dextran (coefficient of variation (CV) = 2.9%)). LDL-C was calculated using the Friedewald equation.(18) Plasma lipoprotein particle concentrations were measured at LipoScience, Inc. (Raleigh, N.C.) by NMR spectroscopy using the LipoProfile-3 algorithm. HDL-P and LDL-P (CVs<4%) are the sums of the particle concentrations of their respective subclasses, which are quantified based on particle size using the amplitudes of their lipid methyl group NMR signals, and mean particle sizes are the weighted average of related subclasses.(19)

Endpoints

Carotid atherosclerosis was measured using high-resolution B-mode ultrasound as previously described for the Cardiovascular Health Study.(20) CIMT was calculated from maximal thickness measured at 8 sites (right and left, near and far walls of the common and internal carotid arteries), as previously described.(21) Incident CHD events (myocardial infarction, CHD death, resuscitated cardiac arrest, or definite or probable angina (followed by revascularization)) were ascertained and adjudicated for MESA as previously described. (22) Hypotheses were also tested using secondary endpoints of 1) "hard" CHD, which excluded angina, 2) "all CVD", which was "all CHD" plus stroke, stroke death, other atherosclerotic death or other CVD death, and 3) "hard" CVD, which included "hard CHD" plus stroke and stroke death, and finally, 4) cIMT considered separately for the common vs. the internal (more susceptible to plaque) carotid artery.

Statistical Analysis

Analyses were performed using SAS 9.2 (SAS Institute, Cary, NC). Two-tailed p values <0.05 were considered significant. Spearman-rank correlations were adjusted for age, sex, and ethnicity. HDL-C and HDL-P were analyzed as continuous variables (results reported per 1 standard deviation (1-SD) increment) or categorized as tertiles or quartiles. Triglycerides were log-transformed. Associations with cIMT were modeled using analysis of covariance and linear regression, and with incident CHD events using Cox proportional hazards regression. All models were adjusted for a base set of covariates: age, sex, ethnicity, hypertension and smoking. P for linear trend was calculated using contrasts. Hypothesized differences by sex, ethnicity, diabetes and hsCRP were tested by including interaction terms with HDL-C and HDL-P for each of the models, with the main effect included in the model. Sensitivity analyses included excluding hormone therapy users (n=837), stratifying by sex, and also testing hypotheses using secondary endpoints described above, i.e., "hard" CHD, "all CVD", "hard" CVD, internal carotid artery (ICA) cIMT and common carotid artery (CCA) cIMT.

To illustrate multivariable regression results, a stratified analysis was used to calculate adjusted mean cIMT for HDL-C tertiles within HDL-P tertiles, and then further stratified by

above/below median LDL-P. Tertiles (HDL-C and HDL-P) were used rather than quartiles, to allow for a sufficient number of individuals in discordant cells (i.e., high HDL-C/low HDL-P or low HDL-C/high HDL-P). Finally, we sought to replicate a report of an increased risk of CHD for very high HDL-C (80 mg/dl) relative to low HDL-C (<40 mg/dl) in adjusted models,(9) and also evaluated CHD risk for very high vs. low HDL-P, using the corresponding 95th and 25th percentiles of HDL-P (45.7 and <29 µmol/l).

Results

Study participants were multi-ethnic men and women ages 45-84 from MESA, without baseline clinical CVD or lipid-lowering medication use (Table1). HDL-C and HDL-P concentrations were positively correlated (Fig 1). HDL-C and HDL-P were inversely correlated with LDL-C, weakly, (ρ= –0.08, –0.13, respectively), and LDL-P, more strongly, (ρ= –0.38, –0.25) and with other metabolic risk factors (e.g., small LDL-P, triglycerides, BMI, waist circumference, and HOMA-IR), but for all, correlations were stronger for HDL-C than for HDL-P (Table 2).

LDL-C and LDL-P

Positive associations of LDL-C and LDL-P with cIMT and CVD events in MESA have been published(21,23) and when LDL-P and LDL-C differ, associations are stronger for LDL-P(23). In our study, adjusted for base covariates, the cIMT difference (95%CI) per 1-SD increment was 28.8(13.4, 44.3) µm for LDL-C, and 36.5 (20.8, 52.1) µm for LDL-P. The HR (95%CI) for CHD per 1-SD increment was 1.24 (1.09, 1.42) for LDL-C and 1.29 (1.13, 1.47) for LDL-P, adjusted for base covariates. LDL-C and LDL-P each remained associated ($p < 0.05$) with cIMT and incident CHD in the models reported below that also adjusted for HDL-P, HDL-C or both.

Associations with Carotid Atherosclerosis

Adjusted for base covariates (age, sex, ethnicity, hypertension and smoking) mean cIMT was lower with higher quartiles of HDL-C (Figure 2, Panel A) or HDL-P (Figure 2, Panel B). The inverse linear association between HDL-C and cIMT was attenuated by adjusting for LDL-P or for HDL-P, and was abolished when adjusted for both. In contrast, HDL-P remained inversely associated with cIMT after adjusting for LDL-P, HDL-C, or both (p for trend <0.05 for all).

Additional models are shown in Table 3, which reports adjusted mean cIMT difference for a 1-SD increment in HDL-C or HDL-P. Separately, higher HDL-C and HDL-P were each associated with lower mean cIMT, and were modestly attenuated by adjusting for LDL-C or HDL particle size. However, adjusting for LDL-P substantially attenuated cIMT estimates, more for HDL-C (12.2 (–21.4,–3.1) µm), than for HDL-P (–20.7 (–29.6, –11.8) µm). Further adjustment for LDL-C and triglycerides had little effect.

In joint models, (HDL-C and HDL-P adjusted for each other and base covariates; Table 3 lower panel), HDL-C and HDL-P associations were only mildly attenuated by adjusting for LDL-C. However, adjusted for HDL-P, HDL-C was no longer significantly associated with cIMT (–11.1 (–22.7, 0.42) µm, and became positive, but not statistically significant, in models that also adjusted for mean HDL size or LDL-P, with or without LDL-C and triglycerides. Conversely, HDL-P remained inversely associated with cIMT (–22.2 (–33.8, –10.6) µm) when adjusted for HDL-C, and also for LDL-P, or HDL size, or LDL-P, LDL-C and triglycerides.

To visualize the multivariable model results, Figure 3 shows adjusted mean cIMT for cross-classified tertiles of HDL-C and HDL-P (Fig 3: A), and further stratified by median LDL-P

(Fig 3: B, C). Power is limited for these analyses due to small numbers in the discordant groups, e.g., high HDL-C/low HDL-P. Within HDL-P tertiles, HDL-C was generally not inversely associated with cIMT (p HDL-C trend= not significant for all) (Fig 3: A), but within HDL-C tertiles, HDL-P was inversely associated with cIMT in both low and high HDL-C tertiles (p trend <0.05 for both). When further stratified by median LDL-P (Fig 3: B, C), trends were generally not statistically significant, but HDL-C was positively associated with cIMT within 5 of the 6 HDL-P tertiles, whereas HDL-P remained inversely associated with cIMT within 5 of the 6 HDL-C tertiles.

Sensitivity Analyses for cIMT Associations

Interaction terms for sex, ethnicity, diabetes status, or hsCRP were not significant. In sex-stratified models, adjusted for LDL-P and each other, HDL-C was not inversely associated with cIMT for women: −1.2 (−15.0, 12.7) µm, or men: 8.3 (−13.4, 30.1) µm, whereas HDL-P was significantly inversely associated with cIMT for women: −17.8 (−31.4, −4.2) µm, and men: −27.1 (−47.7, −6.4) µm. Evaluated separately, the internal and common carotid artery showed similar results to our combined cIMT measure, although associations were slightly stronger for the internal carotid artery, which is more prone to plaque.

Associations with Incident Coronary Heart Disease Events

Among the 5597 participants with incident CHD data, 227 CHD events occurred during mean(SD) =6.0(1.4) years follow-up. The proportionality assumption appeared valid, i.e., interactions of time with HDL-C or HDL-P were not significant. CHD risk is reported for quartiles and for a 1-SD increment in HDL-C and HDL-P, for separate and joint models (Table 4). Separately, higher HDL-C and HDL-P were similarly associated with lower CHD risk, adjusted for base covariates. Adjusting for LDL-C or HDL particle size had little effect. Adjustment for LDL-P attenuated the association for HDL-C to a greater extent than for HDL-P. In joint models (adjusted for HDL-P), HDL-C HRs were not statistically significant and became weakly positive when adjusted for LDL-P, LDL-C, and (log) triglycerides. In contrast, HDL-P's inverse association with CHD remained statistically significant when adjusted for HDL-C, LDL-P, LDL-C, and triglycerides.

Sensitivity Analysis for CHD Associations

In multivariable models, interaction terms for sex, ethnicity, diabetes or hsCRP were not statistically significant. Results were similar if adjusted for baseline diabetes status, or if current hormone users were excluded, or if stratified by sex. With few cases among women (n=66), CHD risk estimates for women were not statistically significant, but the base covariate-adjusted HR (95%CI) for (1-SD) higher HDL-C (0.81 (0.63, 1.05) became 1.00 (0.71, 1.43) when adjusted for HDL-P and LDL-P, whereas for HDL-P, the base covariate-adjusted HR(95%CI) of 0.77 (0.59, 1.00), became 0.81 (0.57, 1.14) when adjusted for HDL-C and LDL-P. Results were also similar for the secondary outcomes of all CVD or hard CHD or hard CVD (Online Table 1.)

Finally, we evaluated CHD risk associations at very high levels of HDL-C and HDL-P. Adjusted for base covariates, LDL-P, HDL-P, and log-triglycerides the HR(95%CI) for very high HDL-C (80 mg/dl) compared with low HDL-C (<40 mg/dl) became positive and statistically significant (2.59 (1.11, 6.02). Conversely, adjusted for base covariates, LDL-P, HDL-C and log-triglycerides, the HR(95%CI) for analogous very high vs. low HDL-P (45.7 vs. <29 µmol/l) was 0.50 (0.19, 1.35).

Discussion

Among multi-ethnic men and women without clinical CVD or lipid-lowering medication use at baseline, HDL-C associations with cIMT and incident CHD were substantially attenuated by adjusting for atherogenic lipoproteins, particularly LDL-P. In contrast, HDL-P associations with cIMT and incident CHD were relatively unaffected by adjusting for atherogenic lipoproteins, HDL-C, and mean HDL particle size. Results were similar for secondary outcomes of "all CVD," and "hard" CHD or CVD events.

Few studies have evaluated HDL-P associations with CHD risk, and we know of none that evaluated it jointly with HDL-C and LDL-P. Low HDL-P levels predicted CHD death over 18 years of follow-up among men with metabolic syndrome in the MRFIT cohort.(24) In EPIC-Norfolk, lower HDL-P levels predicted incident events independent of age, sex, apoB, triglycerides, mean HDL particle size, smoking, myeloperoxidase, paraoxonase-1, and hsCRP.(10) Lower HDL-P also predicted CVD events among HIV patients.(25) In the VA-HIT Study, lower levels of baseline and on-trial HDL-P predicted CHD events among men with low HDL-C randomized to gemfibrozil vs. placebo.(26) In the large Women's Health Study, the inverse association of HDL-P with incident CVD over an 11 year follow-up was not significant.(27) However, HDL-P was inversely associated with incident CHD among postmenopausal women in the Women's Health Initiative Hormone Trial, adjusted for treatment arm,(28) and the inverse association of HDL-P with cIMT was statistically significant for women in the current study. Future studies may help to reconcile these results.

In this study, HDL-C was not inversely associated with cIMT or CHD after adjusting for LDL-P and HDL-P, similar to reported attenuation of HDL-C associations when adjusted for apoB, as an index of atherogenic lipoproteins, and apoA-I.(9) In our study, very high HDL-C (80 mg/dl) became positively associated with CHD risk ($p < 0.05$), when adjusted for LDL-P, HDL-P and triglycerides, as reported in IDEAL (adjusted for apoB and apoA-I.)(9) In contrast, very high HDL-P (45.7 μmol/l) remained inversely associated with CHD, in models adjusting for LDL-P, HDL-C and triglycerides. However, the results of this study suggest that, adjusted for LDL-P and HDL-P, HDL-C loses its inverse association with atherosclerotic CHD across the range of HDL-C, not just at very high HDL-C.

Potential Mechanisms

These results suggest that since HDL-C, the cholesterol content of HDL, varies inversely with triglycerides, LDL-P or apoB, and other metabolic risk factors, the risk attributed to HDL-C may come from several sources other than low levels of particles (i.e. HDL-P). In contrast, HDL-P remained inversely associated with atherosclerotic risk relatively independently of both atherogenic lipoprotein levels, and its own cholesterol content (i.e. HDL-C). More HDL particles may equal higher reverse cholesterol transport capacity. Cholesterol efflux, an index of HDL's capacity for reverse cholesterol transport, was inversely correlated with cIMT and angiographic coronary disease independent of HDL-C, and cholesterol efflux was also associated with higher levels of apoA-I (a rough measure of HDL particles) independent of HDL-C.(29) Furthermore, in a study of diabetic patients, cholesterol efflux was positively associated with total HDL-P but not with HDL-C or apoA-I.(30) Anti-atherogenic benefits of higher HDL-P (anti-oxidation, anti-inflammation, etc.) may also be related to HDL's protein or other cargo (e.g., apoA-I, paraoxonase-1, myeloperoxidase) rather than to its cholesterol cargo.(1,2) Indeed, in EPIC-Norfolk, the HDL-associated anti-oxidant paroxonase-1 was positively correlated with HDL-P more strongly than with HDL-C or apoA-I.(10) However, given the complexity of HDL, many potential mechanisms require further investigation.(2)

Terapeutic Interventions

Lifestyle and pharmacologic interventions to increase low HDL-C have been reviewed elsewhere,(31) but few studies have evaluated intervention effects on HDL-P compared with HDL-C. A few studies have reported that HDL-C and HDL-P are both higher with hormone therapy(28) and alcohol intake.(32) Active smokers have lower levels of both HDL-C and HDL-P, (33) and both increased with smoking cessation in a recent randomized clinical trial. (34) The limited existing data suggest that physical activity(35) and diet/exercise interventions may increase HDL-C but not HDL-P,(36) which would occur if large cholesterol-rich particles increase at the expense of smaller particles.

Niacin also raises HDL-C with little effect on HDL-P.(37) Torcetrapib reportedly raised HDL-P by only 1% despite a 53% increase in HDL-C.(38) In contrast, gemfibrozil increased HDL-P more than HDL-C in VA-HIT, and as noted, on-treatment HDL-P predicted lower CHD events.(26) Statins also increase HDL-P more than HDL-C, as well as decreasing LDL-P less than LDL-C.(39) Among diabetic individuals, effects of vitamin E intake on HDL function (i.e., cholesterol efflux) differ by haptoglobin genotype, increasing it for Hp2-2 and decreasing it for Hp 2-1.(12) Whether effects on HDL-P levels would parallel these effects is unknown. Additional research is needed to quantify differential effects of pharmacological interventions as well as types of diet, omega-3 fatty acids, haptoglobin genotype(12) and other influences on HDL in relation to outcomes.

Potential implications

These results show that associations of HDL-C with CHD risk may be partially due to metabolic correlations with atherogenic lipoprotein concentrations. In contrast, associations of HDL-P with CHD risk were substantially independent of atherogenic lipoprotein concentrations, and of HDL-C. HDL-C is one measured parameter of HDL. It may be important to assess parameters other than HDL-C in clinical trials of interventions to raise "HDL." HDL-P may be an alternative to HDL-C as a marker of HDL-related cardiovascular risk, if these findings are confirmed in other studies and found to be cost-effective.

Potential limitations

These results, while robust, are observational, do not prove causality, and may be subject to bias and confounding, measurement error or unmeasured confounders. Lipid and lipoprotein levels are dynamically metabolically interrelated,(8,11) and statistical modeling in a large observational study is only one tool to investigate potential effects of these interrelationships, which warrant additional metabolic studies. However, despite statistical adjustment for several correlated lipoproteins, multicollinearity was not a problem in this study, as assessed by variance inflation factors. ApoA-I and apoB are not currently available in MESA, so their influence could not be evaluated in the current study. Finally, there were very few events at the high end of the HDL parameter ranges and few among women, limiting our ability to evaluate independent effects on CHD risk in those groups.

Conclusion

Among multi-ethnic men and women, associations of HDL-C with carotid IMT and incident CHD were substantially attenuated by adjusting for atherogenic lipoproteins and HDL-P, whereas HDL-P remained significantly inversely associated with cIMT and incident CHD, independent of atherogenic lipoproteins (LDL-P, triglycerides and LDL-C) and HDL-C. These results may have implications both for risk assessment and for evaluation of therapeutic interventions, particularly pharmacologic interventions which may differentially affect several lipid and lipoprotein parameters concurrently. Quantitative and metabolic interrelationships between lipids and the lipoprotein particles that carry them should be

considered when evaluating associations between single parameters, e.g., HDL-C, and atherosclerotic cardiovascular risk.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

Abbreviations

CVD	cardiovascular disease
CHD	coronary heart disease
HDL-P	High-density lipoprotein particles
LDL-P	Low-density lipoprotein particles
LDL-C	Low-density lipoprotein cholesterol
HDL-C	High-density lipoprotein cholesterol
TG	Triglycerides
hsCRP	high sensitivity c-reactive protein
cIMT	carotid intima-media thickness
apoA-I	apolipoprotein A-I
apoB	apolipoprotein B
MESA	Multi-Ethnic Study of Atherosclerosis
HOMA-IR	homeostasis model of insulin resistance
SD	standard deviation

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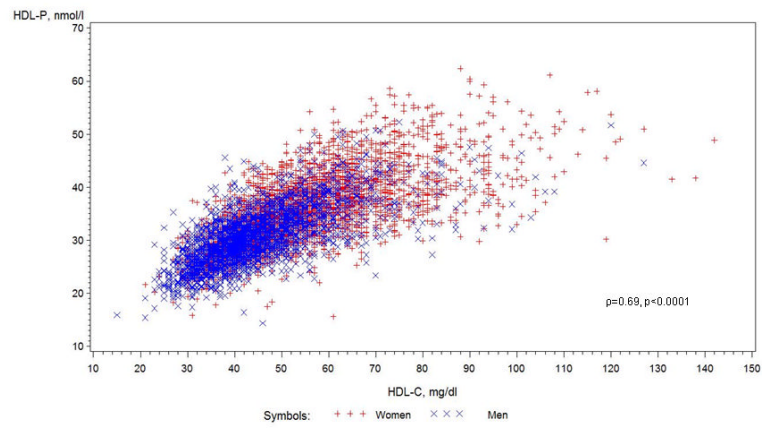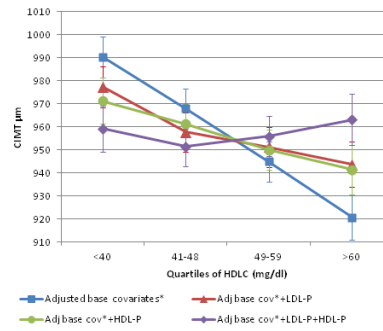
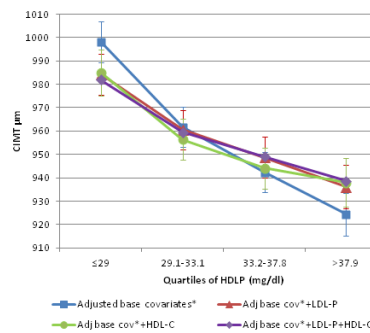**Figure 1.**
Scatterplot of HDL-C with HDL-P
All results except Fig 1 are adjusted for base covariates (base cov.): age, sex, ethnicity, hypertension, and smoking. Error bars are standard errors; p values are for linear trend.

A. Mean cIMT by Quartiles of HDL-C

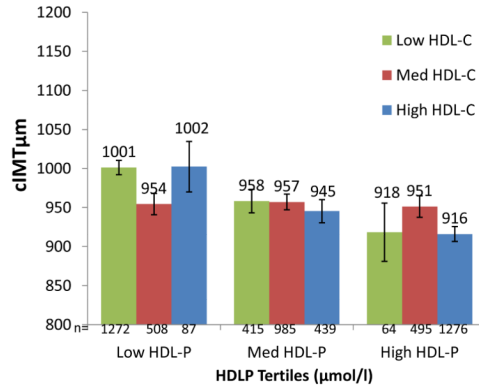B. Mean cIMT by Quartiles of HDL-P

Figure 2.
Mean cIMT (µm) across quartiles of HDL-C or HDL-P, before and after adjusting for LDL-P and each other (n=5541).
A. Mean cIMT (µm) across HDL-C quartiles, adjusted for base covariates, p <0.001; adjusted for base covariates plus LDL-P, p=0.01; adjusted for base covariates plus HDL-P, p=0.051, and adjusted for base covariates plus LDL-P and HDL-P, p=0.75.
B. Mean cIMT (µm) across HDL-P quartiles; adjusted for base covariates, p <0.0001; adjusted for base covariates plus LDL-P, p=0.0001; adjusted for base covariates plus HDL-C, p=0.002; adjusted for base covariates plus LDL-P and HDL-C, p<0.001
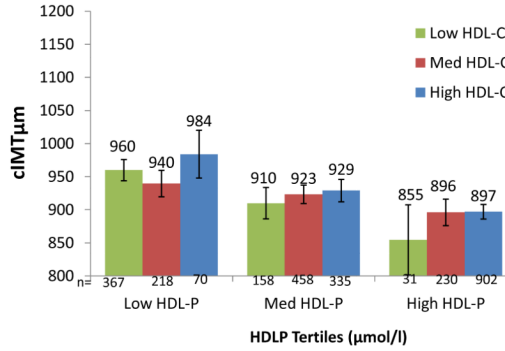All results except Fig 1 are adjusted for base covariates (base cov.): age, sex, ethnicity, hypertension, and smoking. Error bars are standard errors; p values are for linear trend.

A. Mean cIMT by HDL-C and HDL-P Tertiles, Entire Sample

B. Mean cIMT by HDL-C and HDL-P Tertiles, LDL-P < 1236.5 nmol/l

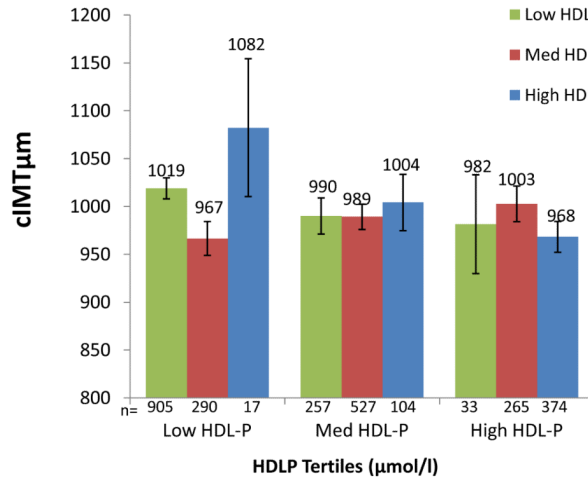C. Mean cIMT by HDL-C and HDL-P Tertiles, LDL-P ≥ 1236.5 nmol/l

Figure 3.
Mean cIMT (μm) by joint tertiles of HDL-C and HDL-P, entire group, and stratified by median LDL-P (1236.5 nmol/l). All models adjusted for base covariates.
HDL-C tertiles (mg/dl): low HDL-C (42), medium HDL-C (43-54), high HDL-C: (55.)
HDL-P tertiles (μmol/l): low HDL-P (30.5), medium HDL-P (30.6-36.1), high HDL-P: (30.6).

A. Entire study sample.
P for HDL-C trend within each HDL-P tertile= not significant.
P for HDL-P trend <0.05 for both low and high HDL-C tertiles, not significant for medium HDL-C tertiles. B. Participants with LDL-P below the median.
P for HDL-C trend within each HDL-P tertile: not significant.
P for HDL-P trend within HDL-C tertile= 0.06, 0.09, 0.02 for low, medium and high HDL-C tertiles, respectively.
C. Participants with LDL-P above the median.
Neither HDL-C trends within each HDL-P tertile, nor HDL-P trends within each HDL-C tertile were significant.
All results except Fig 1 are adjusted for base covariates (base cov.): age, sex, ethnicity, hypertension, and smoking. Error bars are standard errors; p values are for linear trend.

Table 1

Characteristics of MESA Participants in Study (n=5598), 2000-2002

Characteristic	Mean(SD) or %
Age, years	61.5(10.3)
Women, %	52.9
Ethnicity, %	
White	37.6
Chinese-American	12.1
African- American	27.7
Hispanic	22.7
Body Mass Index, kg/m^2	28.2(5.5)
Waist Circumference, cm	97.6 (14.4)
Smoker, %	
Current	13.5
Former	36.0
Hypertension, %	41.3
Diabetes, %	10.7
Carotid Intima-Media thickness, μm	946 (339)
Lipids, mg/dL	
HDL-C	51.2 (15)
LDL-C	119.6 (31.3)
Total Cholesterol	195.8 (34.6)
Triglycerides	124.5 (64.8)
Lipoprotein Particle Concentrations	
HDL-P, μmol/L	
Total	33.8 (6.6)
Large	6.1 (3.5)
Medium	13.4 (6.8)
Small	14.3 (5.6)
LDL-P, nmol/L	
Total	1261 (342)
Large	607 (253)
Small	524 (381)
IDL-P	130.5(98.5)
Lipoprotein Particle Size(nm)	
LDL	20.8 (0.6)
HDL	9.3 (0.5)

Values are mean (SD) or n, %. Abbreviations: HDL: High density lipoprotein, HDL-C: HDL cholesterol, HDL-P: HDL particles, IDL-P: intermediate density lipoprotein particles LDL: Low density lipoprotein, LDL-C: LDL cholesterol, LDL-P: LDL particles.

Table 2

Spearman correlations of the concentrations of HDL cholesterol (HDL-C) and HDL particles (HDL-P) among MESA participants in study (n=5598), 2000-2002

	HDL-C	HDL-P
Mean HDL particle size, nm	0.68	0.34
LDL-C, mg/dl	−0.08	−0.13
Triglycerides, mg/dl	−0.48	−0.09
Total Cholesterol, mg/dl	0.14	0.12
Large HDL-P, μmol/L	0.87	0.53
Medium HDL-P, μmol/L	0.45	0.58
Small HDL-P, μmol/L	−0.28	0.07
Mean LDL Particle Size, nm	0.69	0.32
LDL-P, nmol/L	−0.38	−0.25
Large LDL-P, nmol/L	0.53	0.22
Small LDL-P, nmol/L	−0.69	−0.38
Body Mass Index, kg/m^2	−0.26	−0.12
Waist Circumference, cm	−0.27	−0.12
HOMA-IR [*]	−0.38	−0.16
C-reactive protein, mg/L	−0.15	−0.05

All correlations (Spearman-rank) are adjusted for age, sex, ethnicity and all are statistically significant (p<0.0001)

[*] calculated for participants reporting no anti-diabetic medication (n=5157)

Abbreviations: HDL: High density lipoprotein, HDL-C: HDL cholesterol, HDL-P: HDL particles, HOMA-IR: Homeostasis model of insulin resistance, LDL: Low density lipoprotein, LDL-C: LDL cholesterol, LDL-P: LDL particles.

Table 3

Predicted difference in carotid intima-media thickness (μm) for a 1 standard deviation (SD) greater HDL cholesterol (HDL-C) or HDL particle (HDL-P) concentration among MESA participants (n=5541), 2000-2002

	Difference (95%CI) in cIMT (μm)	
HDL-C and HDL-P in separate models, covariates	**HDL-C (SD=15 mg/dl)**	**HDL-P (SD= 6.64 μmol/l)**
Basic Model [*]	−26.1 (−34.7, −17.4)	−30.1 (−38.8, −21.4)
plus LDL-C	−22.7(−31.4,−14.1)	−25.5(−34.2,−16.7)
plus HDL size	−19.3 (−31.2, −7.4)	−25.5 (−34.6, −16.3)
plus LDL-P	−12.2 (−21.4, −3.1)	−20.7 (−29.6, −11.8)
plus LDL-P, LDL-C and (log) TG	−13.9 (−23.9,−3.9)	−21.6 (−30.5,−12.7)
Joint models [†] of HDL-C, HDL-P, covariates	**HDL-C (SD=15 mg/dl)**	**HDL-P (SD= 6.64 μmol/l)**
Basic Model	−11.1 (−22.7, 0.42)	−22.7 (−34.3, −11.0)
plus LDL-C	−10.9 (−22.4, 0.6)	−18.2 (−29.8, −6.5)
plus HDL size	4.9 (−11.0 20.8)	−27.9 (−40.1, −15.8)
plus LDL-P	2.3 (−9.5, 14.2)	−22.2 (−33.8, −10.6)
plus LDL-P, LDL-C and (log) TG	5.7 (−8.2, 19.7)	−25.2 (−37.6, −12.8)

[*] Basic model and all substantial models are adjusted for: age, sex, ethnicity, hypertension, and smoking.

[†] Joint models report estimates with HDL-C and HDL-P together in the same model.

Abbreviations: HDL: High density lipoprotein, HDL-C: HDL cholesterol, HDL-P: HDL particles, LDL: Low density lipoprotein, LDL-C: LDL cholesterol, LDL-P: LDL particles, TG: Triglycerides

Table 4

Risk of incident CHD across higher HDL-C and HDL-P quartiles and for 1-SD increment among MESA participants (n=5597), baseline 2000-2002

	Hazard Ratio (95%CI) by HDL-C Quartile: range, mg/dl					HR(95% CI) for 1 SD* HDL-C
	Q1:15-40	Q2: 41-48	Q3: 49-59	Q4: 60-142	P trend	
Basic Model	1.0	0.67 (0.48, 0.94)	0.56 (0.38, 0.81)	0.45 (0.29, 0.69)	<0.0001	0.74 (0.63, 0.88)
plus LDL-C	1.0	0.64 (0.46,0.89)	0.53 (0.37, 0.77)	0.45 (0.29, 0.70)	<0.0001	0.74 (0.62, 0.88)
plus HDL size	1.0	0.67 (0.48,0.94)	0.55 (0.37, 0.83)	0.44 (0.25,0.76)	0.0002	0.74 (0.60, 0.92)
plus LDL-P	1.0	0.67 (0.48, 0.94)	0.61 (0.42, 0.89)	0.53 (0.34, 0.84)	0.002	0.80 (0.67, 0.95)
plus HDL-P	1.0	0.77 (0.54, 1.09)	0.71 (0.47, 1.09)	0.67 (0.39, 1.16)	0.10	0.92 (0.74, 1.14)
plus HDL-P+LDL-P	1.0	0.76 (0.54, 1.08)	0.77 (0.50, 1.18)	0.77 (0.44, 1.36)	0.25	0.97 (0.77, 1.22)
plus HDL-P, LDL-P, LDL-C,TG	1.0	0.83 (0.57,1.20)	0.85 (0.52,1.40)	0.92 (0.48,1.77)	0.69	1.12 (0.86,1.46)

	Hazard Ratio (95%CI) by HDL-P Quartile (range, µmol/l)					HR(95% CI) for 1 SD* HDL-P
	Q1:14.4-29	Q2: 29.1-33.1	Q3: 33.2-37.8	Q4: 37.9-62.4	P trend	
Basic Model	1.0	0.79 (0.57, 1.08)	0.49 (0.33, 0.73)	0.46 (0.30, 0.71)	<0.0001	0.70 (0.59, 0.82)
plus LDL-C	1.0	0.81 (0.59, 1.12)	0.50 (0.34, 0.75)	0.49 (0.31, 0.75)	<0.0001	0.71 (0.60, 0.84)
plus HDL size	1.0	0.81 (0.59,1.12)	0.52 (0.35,0.77)	0.49 (0.31,0.77)	0.0007	0.74 (0.62, 0.87)
plus LDL-P	1.0	0.84 (0.61, 1.16)	0.55 (0.37, 0.81)	0.52 (0.33, 0.82)	0.0006	0.74 (0.63, 0.87)
plus HDL-C	1.0	0.84 (0.60,1.16)	0.55 (0.36, 0.85)	0.56 (0.33, 0.97)	0.009	0.74 (0.60, 0.92)
plus HDL-C+LDL-P	1.0	0.89 (0.64, 1.23)	0.59 (0.38, 0.91)	0.57 (0.33, 0.98)	0.015	0.75 (0.61, 0.93)
plus HDL-C, LDL-P, LDL-C,TG	1.0	0.82 (0.58, 1.14)	0.52 (0.34, 0.81)	0.49 (0.27, 0.86)	0.003	0.68 (0.54, 0.85)

*
Basic model and all substantial models are adjusted for: age, sex, ethnicity, hypertension, and smoking.

*
1 SD = 15 mg/dl for HDL-C, and 6.64 µmol/l for HDL-P.

Abbreviations: HDL: High density lipoprotein, LDL-P: LDL particles, LDL-C: HDL cholesterol, HDL-P: HDL particles, LDL.: Low density lipoprotein, LDL-C: LDL cholesterol, LDL-P: LDL particles, TG: Triglycerides