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Lipoprotein Particle Number & Size Predict Vascular Structure & Function Better than Traditional Lipids in Adolescents & Young Adults

EM Urbina*, CE McCoy*, Z Gao*, PR Khoury*, AS Shah*, LM Dolan*, and TR Kimball*

*Cincinnati Children's Hospital and University of Cincinnati, Cincinnati, OH

Abstract

Background—In adults, dyslipidemia is associated with higher carotid thickness and arterial stiffness, predictors of CV events. In young subjects, lipid concentrations have not been consistently associated with vascular measures.

Objective—To compare nuclear magnetic resonance (NMR) measures of lipoprotein particle number (LDL-P, HDL-P, VLDL-P) and size (LDL-S, HDL-S, VLDL-S) to determine if they were associated with vascular measures more strongly than lipid concentrations (LDL-C, HDL-C, TG).

Methods—We evaluated 214 lean (L), 228 Obese (O) and 214 diabetic (T2DM) subjects age 10–24 years (33% male, 39% Caucasian). CV risk factors, vascular structure and arterial stiffness were measured. General linear models were constructed including demographics, risk factors and traditional or NMR lipid parameters. A composite vascular function score was developed as the outcome in receiver operator characteristic scores (ROC) for determining which lipid parameter was superior in predicting vascular damage.

Results—Risk factors worsened from L to O to T. However, LDL-C was similar in O and T while LDL-S differentiated the 3 groups ($T > O > L$, $p = 0.0001$). Models demonstrated the superiority of NMR values which entered for all but one vascular outcome and explained more of the variance than traditional lipid concentrations. ROC curves demonstrated that NMR values were superior in predicting vascular outcomes. Models stratified by race were similar but cut-points predicting vascular outcomes differed by race for TG, TG/HDL and VLDL.

Conclusion—Lipoprotein particle number and size are more strongly related to vascular structure and function than traditional lipid values. NMR lipid measures may be a better indicator of risk for target organ damage than traditional lipid measures in adolescents and young adults.

Address for correspondence and reprint requests: Elaine M. Urbina, M.D. M.S., Cincinnati Children's Hosp. Med. Ctr., Director of Preventive Cardiology, 3333 Burnet Ave., MLC 7002, Cincinnati, OH 45229, Office: (513) 636-8265, Fax: (513) 636-0162, Elaine.urbina@cchmc.org.

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Keywords

carotid arteries; arterial stiffness; lipoproteins; dyslipidemia; pediatrics

Introduction

In the late 1950s, the Framingham Heart Study established the relationship between elevated cholesterol levels and cardiovascular (CV) disease.¹ Subsequent data demonstrates a substantial number of individuals who develop CVD have normal low density lipoprotein cholesterol (LDL-C) levels.² Recently, direct measurement of lipoprotein particle size with nuclear magnetic resonance (NMR) has become available for clinical and research use.³ The use of NMR allows for measurement of number and size of LDL particles which may be better predictors of CV events⁴ and early non-invasive atherosclerotic target organ damage (carotid intima media thickness, cIMT)⁵ in adults than traditional cholesterol concentration.

Few studies have measured lipoprotein particle size and number in adolescents and young adults^{6, 7} but none have compared their usefulness in predicting target organ damage in young subjects. Therefore, we measured traditional and NMR lipid values and cIMT and arterial stiffness in adolescents and young adults. We hypothesized that NMR lipid values would be more strongly associated with target organ damage than traditional lipids even after adjusting for other CV risk factors.

Materials and Methods

Population

The population consisted of 674 subjects mean age 18 years (10–24 years, 33% male, 39% Caucasian) recruited for a study of the effects of type 2 diabetes mellitus (T2DM) on CV health. There were 12 Hispanics: 6 subjects who identified as white and Hispanic, 1 black and Hispanic, 4 other Hispanic and 1 more than one race Hispanic. Of the non-Hispanics, 259 were white, 394 black, 1 American Indian, 8 more than one race. Since there were so few Hispanics and races other than white or black, for analyses, the races/ethnicities were compressed into white and black without regards to ethnicity. Subjects with T2DM were matched by age, race (Caucasian or African-American), and sex to both lean and obese controls. T2DM (T=214) was determined using American Diabetes Association criteria.⁸ Obese controls (O=228) had BMI ≥ 95th percentile by CDC criteria with normal oral glucose tolerance test. Lean subjects (L=214) had BMI < 85th percentile. Pregnant women were excluded. Written informed consent was obtained from individuals over 18 years old or the parent or guardian for individuals less than 18 years old. Written assent was also obtained for individuals less than 18 years old according to the guidelines established by the Institutional Review Board at Cincinnati Children's Hospital Medical Center.

Laboratory

After a minimum 10 hour overnight fast, participants had questionnaire, anthropometric, blood pressure (BP), laboratory and vascular structure and function data collected. Experienced personnel obtained two measures of height using a calibrated stadiometer

(Veeder-Rood, Elizabethtown, North Carolina, USA) and two measures of weight using a Health-O-Meter electronic scale. The average of each was used. Three measures of BP were obtained with a mercury sphygmomanometer according to published pediatric standards⁹ and the average was used. Fasting plasma glucose, insulin, glycosylated hemoglobin (HbA1c) and high sensitivity C-reactive protein (CRP) were measured by standard techniques.¹⁰ Assays of fasting plasma lipids were carried out in a NHLBI-CDC standardized laboratory. LDL cholesterol concentration was calculated using the Friedewald equation as previously described.¹⁰

The lipoprotein particle analysis was performed with a 400-MHz proton NMR analyzer at Liposcience (Raleigh, North Carolina) as previously described.³ In brief, the number of particles of lipoprotein subclasses of different size are derived from the measured amplitudes of the distinct lipid methyl group NMR signals they emit. The intensity of each signal is proportional to the quantity of the subclass which is converted to particle concentration units (nmol/L for LDL-P and very low density lipoprotein cholesterol (VLDL-P) and $\mu\text{mol/L}$ for high density lipoprotein cholesterol (HDL-P)). Although this technique can separate VLDL, LDL and HDL into 10 subclasses, average particle numbers were counted (VLDL-P, LDL-P, HDL-P) and sizes (VLDL-S, LDL-S and HDL-S) were computed as the sum of the diameter of each subclass multiplied by its relative mass percentage as estimated from the amplitude of its methyl NMR signal. NMR lipoprotein particle analyses were done on frozen samples that had been stored at -80°C . Previous studies have demonstrated that NMR lipoprotein particle analyses are unaffected by frozen storage and multiple freeze-thaw cycles.¹¹ Reproducibility of the NMR-measured lipoprotein particle parameters determined by replicate analyses of plasma pools found between-run coefficients of variability for low-normal concentrations were $<4\%$ for total LDL and HDL particle concentrations, $<0.5\%$ for LDL and HDL size, $<8\%$ for large and small LDL subclasses, and $<5\%$ for large and small HDL subclasses.¹¹ For all NMR analyses, samples were handled in a fully blinded fashion such that investigators had no knowledge of subject characteristics.

Vascular testing

Carotid ultrasound was performed using B-mode ultrasonography with a GE Vivid 7 ultrasound imaging system (GE Medical Systems, Wauwatosa, WI) with a high-resolution linear array vascular ultrasound centered at 7.5 MHz. For each subject, the far wall of each carotid segment bilaterally was examined independently from continuous angles to identify the thickest cIMT for the right and left common, bulb (bifurcation) and internal carotid arteries. Multiple digital image loops were digitally transmitted using the Amicas Vericys Medical System (Chicago, IL) for offline reading and analyses. All images were read by a single experienced and research-trained ARDMS Registered Vascular Sonographer who was blinded to subject group. The mean of right and left carotid segments were used in analyses for all parameters. A manual tracing technique was employed to measure the maximum carotid thickness from the leading edge (lumen-intima) to the leading edge (medial-adventitia). This technique has coefficients of variation for repeat readings of 5.3% to 8.0%.¹² M-mode measurements of the distal common carotid were also performed 1 cm proximal to the beginning of the carotid bulb. The maximal and minimal lumen diameters

were read from the M-mode tracing for calculation of carotid stiffness (Peterson's elastic modulus (PEM)).¹³

Non-ultrasound based vascular testing was conducted after 5 min of rest in the supine position. Three measures of brachial artery distensibility (BrachD), and mean arterial blood pressures (MAP), were obtained with a DynaPulse Pathway instrument (Pulse Metric, Inc., San Diego, California, USA) as previously described.¹⁰ DynaPulse derives brachial artery pressure curves from distensibility arterial pressure signals obtained from a standard cuff sphygmomanometer assuming a straight tube brachial artery and T-tube aortic system. Evaluation of repeated measures in our laboratory show excellent reproducibility with coefficients of variability less than 9%.¹⁰ Three measures of carotid-femoral pulse wave velocity (PWV) were obtained with tonometry with a SphygmoCor SCOR-PVx System (Atcor Medical, Sydney, Australia) according to the manufacturer's protocol. The coefficient of variability for repeat measures is less than 7%.¹⁰ Augmentation Index (AIx), which is influenced by arterial stiffness but is also a measure of wave reflections was also collected in triplicate using tonometry on the radial artery using the generalized transfer function applied by the device and normalized to a HR of 75 beats per minute. Reproducibility studies in our laboratory demonstrated intraclass correlation coefficients between 0.7 and 0.9 for all variables.¹⁰

Statistical methods

All analyses were performed with Statistical Analyses Software (SAS, version 9.3; Cary, North Carolina, USA). Mean values for demographic, anthropometric, and laboratory data were obtained by group. Variance stabilizing procedures were employed as needed. Analysis of variance was performed to look for differences by group and Chi squared analyses were performed for categorical variables. Bivariate correlations were calculated between vascular outcome variables and lipid parameters overall and by group. Separate general linear models using stepwise selection process were constructed using important either traditional or NMR lipid parameters and other important covariates from correlation analyses to elucidate if traditional or NMR lipid parameters were independent determinates of CV outcomes after correcting for other CV risk factors. Variables that were available to enter the models included age, race, sex, group, BMI z-score (height for AIx only), systolic BP and diastolic BP z-score for cIMT or MAP for arterial stiffness, heart rate (HR), log fasting glucose, insulin, HbA1c, CRP and either traditional or NMR lipid parameters. Regression diagnostics were performed and no model had a condition index > 3.2 with values <10 indicating lack of significant collinearity. Highest Variance Inflation Factor was 2.48 with values < 5 considered lack of significant collinearity. Model fit was deemed robust with Mallow's Cp no more than 1 greater than P (number of regression parameters).

In an effort to compare traditional lipids to NMR lipid parameters in predicting vascular structure and function in our cohort, we developed a composite measure of vascular target organ damage. We calculated z-scores using means and standard deviations from the lean, healthy subjects in our population for all 3 carotid IMT measures, PWV, AIx and BrachD. The z-scores of all measures were summed to produce the vascular score (VS). A higher score representing thicker and or stiffer vessels. Receiver operator characteristic scores were

then developed for each of the traditional and NMR lipid parameters to determine which had the most sensitivity and specificity in predicting elevated VS (90th% for lean). The non-parametric approach of DeLong¹⁴ was then used to determine if differences in area under the curve (C-statistic) were statistically significant between traditional and NMR lipid values.

Results

The study included more non-Caucasians in the obese non-diabetic control group (O) but there were no differences in age or sex among groups (Table 1). Lean subjects (L) had lower BMI than O & type 2 diabetic group (T) with other CV risk factors increasing in severity across the groups. Traditional lipid parameters also tended to worsen across groups (Table 1) with significantly increasing Total Cholesterol, triglycerides and significantly declining High density lipoprotein cholesterol (HDL-C) from L to O to T (all $p < 0.05$). LDL-C did not differ between O & T. In contrast, LDL particle number (LDL-P) increased and LDL size (LDL-S) decreased from L to O to T such that subjects with type 2 diabetes mellitus (T2DM) had the largest number of the smallest and most dense LDL particles. Very low density lipoprotein cholesterol (VLDL) particle number (VLDL-P) was only higher in T although size increased from L to O to T. HDL particle number (HDL-P) did not differ but HDL size (HDL-S) did decline across the groups. These results demonstrate an increasingly more atherogenic lipid profile across groups.

Subjects with T2DM had evidence for target organ damage with higher cIMT, PEM, AIx, PWV and lower BrachD (Table 1). O had worse vascular structure and function compared to L for all parameters except common cIMT (all $p < 0.05$). Independent determinants of vascular outcomes using NMR lipid parameters are presented in Table 2.

The most consistent covariates were age, sex, race, and blood pressure (BP). Presence of T2DM was important for the AIx and PWV. In the carotid bulb, there was a group by LDL-P interaction such that LDL-P was only significant in T2DM. BMI was significant for PWV and BrachD; metabolic variables for common carotid IMT, PEM and BrachD. An NMR lipid variable entered the models for all outcomes except for common carotid. LDL-P was the most consistent contributor (bulb, internal cIMT, AIx). The only traditional lipid parameter that entered any model was HDL-C for BrachD with a lower amount of the variance explained by the model with HDL-C than with NMR variables (BrachD = $1.9 + 0.067 \times \text{Female} + 0.083 \times \text{BMI z-score} - 0.0024 \times \text{MAP} - 0.0025 \times \text{Heart Rate} - 0.032 \times \log \text{insulin} + 0.08 \times \text{HDL-C}$; $R^2=0.43$, $p < 0.0001$). Regression of internal carotid IMT (Figure 1) and PWV (Figure 2) on NMR and traditional lipid variables demonstrates the stronger relationship with NMR lipid measures. As seen in previous studies, only a small amount of the variance in carotid IMT was explained by the model¹⁰ and the contribution on NMR lipid values was small. To further explore potential race differences in the relationship between NMR lipid values and vascular score, we repeated the models stratified by race (data not shown). The models for white and black were similar to each other although it appeared that diabetes might be more important in blacks and SBP more important in whites. However, the stratified models had fewer significant terms and lower R^2 than the model with races combined which may indicate lower power of the stratified analyses. When we explored differences in optimum cut-points to predict vascular dysfunction by race, not

surprisingly, cut-points for TG, TG/HDL ratio and VLDL particle number differed by for whites (103 mg/dl, 2.63 and 60.1 nmol/L) and blacks (82 mg/dl, 1.8 and 36.7 nmol/L) respectively but the LDL and HDL parameters and non-HDL were similar.

The prevalence of elevated vascular score (VS, 90th for lean subjects) was 30% overall (9.5% for L, 35.5% for O, 46.3% for T). When the C-statistics for the area under the curve (AUC) for the receiver operator characteristic curves were compared by the method of Delong¹⁴ (Table 3), HDL-S had greater sensitivity and specificity compared to HDL-C with a significantly greater AUC ($p = 0.003$). Similarly, LDL-P was superior to LDL-C ($p = 0.001$) with a trend for superiority for LDL-S ($p=0.11$). Triglyceride concentration had a greater AUC than VLDL-P or VLDL-S. Triglyceride to HDL ratio (TG/HDL-C) was greater than TG alone but performed similarly to HDL-S and there was no significant difference in AUC between TG/HDL-C and HDL-S.

Discussion

Our data demonstrated that fewer ideal levels of CV risk factors are present in adolescents and young adults with obesity and T2DM as compared to lean adolescents and young adults. As previously demonstrated, the CV risk factors are associated with non-invasive measures of atherosclerotic target organ damage. Not surprisingly, lean subjects have the lowest LDL-C and triglycerides and the highest HDL-C levels. However, LDL-C levels were similar between O & T2DM groups, whereas NMR lipid evaluation was more sensitive, demonstrating significantly higher LDL-P in diabetic as compared to both obese and lean groups. The most novel finding from our data is that NMR lipid parameters are stronger independent predictors of vascular damage than traditional lipid parameters. These data suggest the utility of using advanced lipid testing to assess risk for target organ damage in high risk adolescents and young adults with obesity and T2DM. This may be especially important in young subjects with T2DM since LDL-P was an important determinant of thickness in the carotid bulb but only in the presence of diabetes.

In 1957, the Framingham Heart Study established the relationship between elevated serum total cholesterol level and incident coronary heart disease.¹ Since then, there have been many refinements in lipid measurements including the recent development of techniques that determine lipoprotein particle number and size. These improvements in technique are important because the total cholesterol and triglyceride concentrations reported with traditional lipid testing include the total amount of cholesterol found in a variety of different lipoprotein particles.² Since the cholesterol content and atherogenic potential of lipoproteins may differ by lipoprotein class and size, the latter may be more strongly related to atherosclerosis than traditional cholesterol concentration. Therefore, it is not surprising that longitudinal adult studies have found that LDL-P had a higher hazard ratio for predicting incident CV events than LDL-C.⁵ Furthermore, in subjects with discordant lipid values (i.e. acceptable LDL-C but abnormal LDL-P), LDL-P, not LDL-C predicted events.^{4, 5} In fact, one study found that LDL-P was related to coronary artery disease even after adjustment for Framingham risk score and LDL-C.¹⁵ Similarly, small HDL size was also independently associated with coronary events for both sexes retaining significance after adjustment for lipid concentration and other CV risk factors in men.¹⁶

LDL-P has proven useful in predicting sub-clinical atherosclerosis, with a stronger relationship to carotid plaque than LDL-C in former NFL football players known to be at high risk for atherosclerosis.¹⁷ LDL-P was also proven useful in healthy individuals. In the Multi-Ethnic Study of Atherosclerosis (MESA) study, multivariable models containing LDL-P predicted increased cIMT and addition of LDL-C did not improve the results.¹⁸ In the Bogalusa Heart Study, models using NMR lipoprotein size had a larger area under the curve for the receiver-operating characteristic curve predicting elevated cIMT (0.782, [0.718–0.841]) although the result was statistically similar to the model using traditional lipid values (0.754 [0.690–0.812]).¹⁹ In the Atherosclerosis Risk in Communities study, both LDL-C and LDL-P were significant predictors of carotid plaque volume in older adults (mean age 71 years), but the odds ratio of greater plaque burden was slightly higher for LDL-P.²⁰ LDL size is also important. LDL-S is inversely related to cIMT independent of CV risk factors in healthy middle age men.²¹ LDL-S also predicted higher cIMT in subjects with familial combined hyperlipidemia where traditional lipids were not significant.²² Small LDL size is also associated with thicker cIMT in middle age subjects with metabolic syndrome²³ and in patients with coronary artery disease.^{24, 25} In subjects with T2DM, small LDL was more highly associated with cIMT than LDL-C²⁶ retaining significance even after adjustment for other risk factors.²⁷ The importance of LDL particle number and size are not entirely clear for subjects with T1DM as the DCCT/EDIC study found stronger relationship between cIMT and LDL-C than LDL-P or LDL-S.²⁸ Similar to our data, however, they found all lipid parameters related more strongly to the internal than the common carotid artery.

Few studies have evaluated the relationship between lipids and arterial stiffness. Specifically, in adults with a mutation in the lecithin:cholesterol acyl transferase gene, affected subjects with very low HDL-C had higher PWV than non-affected controls.²⁹ In healthy Japanese subjects, low HDL-C was also associated with higher brachial-ankle PWV.³⁰ No studies to date have evaluated the effect of lipoprotein particle size on arterial stiffness, we found that a higher TG/HDL-C ratio was associated with increased PWV and lower BrachD in adolescents and young adults.³¹ Although measurement of HDL size was not available in that study, since TG/HDL-C ratio is highly correlated with lipoprotein particle size in adolescents and young adults,³² it appears these results are concordant.

Limitations

This study has a number of limitations. First, our study is cross-sectional. Therefore, we cannot determine if the more adverse sized lipoprotein particles are initiating the target organ damage. Also, by design, our study was enriched with individuals of non-Caucasian race, obesity and T2DM. Although our statistical models controlled for these factors, the results of our study may not be generalizable to the general population. We also only measured traditional and NMR values at one time point so we cannot control for biologic or seasonal variation.³³ Since only a small amount of the variance in carotid IMT was explained by the model and the contribution on NMR lipid values was small it is possible that our cohort was imaged at too young an age to have a sufficient exposure to high LDL particles to demonstrate a strong effect on vascular parameters. Other limitations include that NMR measures were performed on frozen samples while the traditional lipids were

performed on fresh blood. However, there was a correlation of 0.92 between TG measured by both techniques and a previous study found no difference in results after multiple freeze/thaw cycles¹¹ suggesting validity of the frozen samples. We also did not measure oxidized LDL, a biomarker known to have a stronger relationship to target organ damage than the traditional LDL-C measurement.³⁴ Since there are no longitudinal studies using BrachD to predict heart attack and stroke, we were unable to weight our vascular dysfunction score based on strength of prediction of hard CV events. Finally, lack of widespread availability (only a few laboratories in the U.S. perform the testing) and increased cost may limit generalizability and clinical application of NMR lipid testing in the near future.

Conclusions

Adult studies demonstrate the superiority of lipoprotein particle size and number over traditional cholesterol concentration in predicting degree of atherosclerosis. Our study extends these observations to a younger population for the first time. We found that lipoprotein particle number and size are more strongly related to vascular structure and function than traditional lipid values in adolescents and young adults although the effect size for the lipid parameters was small. Since diet,³⁵ exercise programs,³⁶ and drugs^{37–41} may affect lipoprotein particle size in different ways, particle size may become important in assessing the efficacy of treatment of lipids to reduce CV risk. This may be why one recent analysis found that managing LDL-C and LDL-P in comparison to LDL-C alone resulted in reduced costs and fewer CV events.³⁹ For this reason, we may see increased use of measures of lipoprotein particle size in the future to prevent heart attack and stroke.

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Drs. Urbina, Dolan and Kimball designed the study, collected the data, participated in analyses and contributed to the manuscript. Ms. McCoy analyzed the carotid ultrasounds, participated in analyses and contributed to the manuscript. Mr. Khoury and Dr. Gao performed data analyses and contributed to the manuscript. Dr. Shah contributed to the analysis and completion of the manuscript. All authors have approved this version and agree to be accountable for all aspects of the work. We would like to thank the entire T2CVD team and the participants of the T2DVD study and their families, without whose support, this study would not be possible.

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Highlights

- Adolescents and young adults with obesity or T2DM have equally adverse lipid concentrations
- NMR lipid measures can differentiate diabetic versus obese subjects
- NMR measures are more sensitive in identifying subclinical atherosclerosis.

Figure 1.
Regression of Internal cIMT on LDL parameters (solid line = regression, dashed line = confidence interval around the mean).

Figure 2.
Regression of PWV on HDL parameters (solid line = regression, dashed line = confidence interval around the mean).

Table 1Description of the study population (mean $\pm$ SD or frequency).

Variable	group=L		N = 232		group=O		N=228		group=T		N=214	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Age (years)	17.6	3.5	18.0	3.4			18.1	3.1				
Race (%Caucasian) ^a	43		32		44							
Sex (%Male)	36		30		33							
Height (cm) ^b	165.5	10.6	166.3	9.7	168.7	10.0						
Weight (kg) ^c	59.0	11.6	103.1	21.5	106.0	28.5						
BMI (kg/m ²) ^c	21.3	2.6	37.2	7.0	37.1	9.0						
SBP (mmHg) ^d	107.6	10.3	116.6	11.3	121.5	12.1						
DBP (mmHg) ^c	59.4	12.8	66.1	12.3	67.2	13.1						
HR (beats/min) ^d	63.4	10.5	66.2	9.7	70.2	11.8						
Glucose (mg/dl) ^b	89.5	6.2	92.4	7.6	150.5	79.9						
Insulin (mU/ml) ^d	11.5	4.6	22.4	15.1	26.9	19.8						
Glycosylated Hemoglobin (%) ^b	5.4	0.5	5.5	0.4	8.1	2.9						
High sensitivity C-reactive protein (mg/L) ^c	1.0	1.7	4.4	4.2	4.6	4.1						
Total Cholesterol (mg/dl) ^d	160.7	28.4	171.6	32.5	179.8	40.6						
LDL-C (mg/dl) ^c	89.8	23.6	105.2	28.6	107.0	33.3						
HDL-C (mg/dl) ^e	56.5	13.1	47.4	10.1	44.4	11.3						
Triglycerides (mg/dl) ^d	72.5	34.9	99.4	61.1	137.4	87.9						
VLDL-P (nmol/L) ^b	42.7	24.8	46.5	25.3	56.5	37.3						
LDL-P (nmol/L) ^d	849.7	248.7	1102.6	326.8	1257.9	437.9						
HDL-P (nmol/L)	33.1	5.1	32.5	5.1	32.7	5.7						
VLDL-S (nm) ^d	45.5	4.6	48.4	6.2	52.5	8.0						
LDL-S (nm) ^e	21.3	0.5	21.0	0.5	20.7	0.6						

Variable	group=L	N = 232	group=O	N=228	group=T	N=214
	Mean	SD	Mean	SD	Mean	SD
HDL-S (nm) ^e	9.5	0.5	9.0	0.4	8.8	0.4
Common IMT (mm) ^b	0.50	0.08	0.49	0.08	0.53	0.10
Bulb IMT (mm) ^d	0.47	0.09	0.49	0.10	0.53	0.12
Internal IMT (mm) ^d	0.39	0.08	0.41	0.09	0.43	0.11
Peterson Elastic Modulus (mmHg) ^c	175.0	49.6	195.5	60.8	206.5	71.6
Augmentation Index (%) ^d	-0.8	10.9	2.8	11.3	6.5	12.0
Brachial Distensibility (% /mmHg) ^f	6.9	1.2	5.5	1.1	5.3	1.0
Pulse Wave Velocity (m/sec) ^d	5.3	0.7	6.3	1.0	6.6	1.1

P 0.05 for:

^aO>L&T,

^bT>L&O,

^cT&O>L,

^dT>O>L,

^eL>O>T,

^fL>O&T

Table 2

Independent determinants of vascular outcomes.

Parameter	Common	Bulb	Higher worse				PWV	BrachD
			Internal	PEM	AIx	Lower worse		
Intercept	-0.87	-0.89	-1.22	4.8	20.81	1.13	1.32	
Age	0.0097	0.012	0.016	0.021	2.5	0.02		
Race (Caucasian reference)	0.041		0.048	0.05		0.079		
Sex (Male reference)	-0.095	-0.054	-0.11			0.022	0.68	
Group (T2DM reference)								
Lean		0.068 ^a			-3.59	-0.098		
Obese		0.023 ^a			-2.02 ^a	-0.039		
Group by LDL-P interaction								
Lean		-0.00012						
Obese		-0.00056 ^a						
BMI z-score						0.025	-0.085	
Height					-0.31			
SBP z-score		0.024	0.031					
MAP	0.014			0.0047	0.18	0.0032	-0.0021	
HR						0.0028	-0.0024	
Insulin log				0.038			-0.39	
Glycosylated Hemoglobin	0.015							
LDL-P		0.000072	0.00011		0.0035			
LDL-S							0.041	
HDL-S				-0.58		-0.033		
VLDL-S					0.15			
R²	0.18	0.11	0.22	0.17	0.17	0.57	0.44	

All model p 0.0001. All beta parameter estimates p 0.05 except where indicated by ^a. Non-significant terms not involved in an interaction were removed from the models and are represented by blank cells.

Table 3

Area under the curve (AUC) of ROC curves for prediction of Vascular Score >90th% of Lean Subjects

Lipid value	AUC	Sensitivity	Specificity	Best Cut Point
HDL-C	0.68	0.65	0.64	47
HDL-P	0.55	0.67	0.42	33.6
HDL-S ^a	0.73	0.68	0.70	8.9
LDL-C	0.63	0.68	0.53	94
LDL-P ^b	0.70	0.77	0.54	925
LDL-S	0.70	0.54	0.76	20.8
TG	0.69	0.67	0.63	83
VLDL-P	0.56	0.34	0.78	60.1
VLDL-S	0.67	0.55	0.76	49.2
TG/HDL-C	0.72	0.66	0.67	1.8

P 0.003 for

^a AUC significantly different from HDL-C;^b AUC significantly different from LDL-C (by method of DeLong et al¹⁴).