

Original Research Article

Common Set of Genes Regulates Low-Density Lipoprotein Size and Obesity-Related Factors in Alaskan Eskimos: Results from the GOCADAN Study

V. SAROJA VORUGANTI,^{1*} GUOWEN CAI,¹ SHELLEY A. COLE,¹ JEANNE H. FREELAND-GRAVES,² SANDRA LASTON,¹ CHARLOTTE R. WENGER,¹ JEAN W. MACCLUER,¹ BENNETT DYKE,¹ RICHARD DEVEREUX,³ SVEN O.E. EBBESSON,⁴ RICHARD R. FABSITZ,⁵ BARBARA V. HOWARD,⁶ AND ANTHONY G. COMUZZIE¹

¹Department of Genetics, Southwest Foundation for Biomedical Research, San Antonio, Texas 78227-5301

²Division of Nutritional Sciences, University of Texas at Austin, Austin, Texas 78712

³Greenberg Division of Cardiology, Cornell Medical Center, New York, New York 10021

⁴Norton Sound Health Corporation, Nome, AK 99709

⁵National Heart, Lung, and Blood Institute, National Institutes of Health, Bethesda, Maryland 20892-7934

⁶Medstar Research Institute, Hyattsville, Maryland 20783

ABSTRACT Increasing incidence of cardiovascular disease in traditionally low-risk Alaskan Eskimos is a cause for concern. The purpose of this study was to examine the genetic and environmental correlations of low-density lipoprotein (LDL) subfractions with obesity-related factors in Alaskan Eskimos, using data from the first 954 participants of the Genetics of Coronary Artery Disease in Alaska Natives Study. Estimates of genetic and environmental influence were calculated using a maximum likelihood variance component method implemented in SOLAR. Mean values of weight, body mass index (BMI), and waist were 73.4 ± 0.5 kg, 27.6 ± 0.2 kg/m², and 88.0 ± 0.4 cm, respectively. LDL, and its small (LDL1), medium (LDL2), and large (LDL3) subfractions, had mean values of 115.8 ± 1.2 mg/dl, 8.3 ± 0.4 mg/dl, 19.6 ± 0.8 mg/dl, and 71.5 ± 1.5 mg/dl, respectively. Bivariate analysis displayed significant genetic correlations between LDL subfractions and obesity-related factors: LDL1 with BMI ($\rho_G = 0.67$, $P < 0.05$), waist ($\rho_G = 0.80$, $P < 0.001$), and subscapular and tricep skinfolds ($\rho_G = 0.93$, $P < 0.005$, and $\rho_G = 0.78$, $P < 0.05$, respectively); LDL2 with BMI ($\rho_G = 0.52$, $P < 0.05$), waist ($\rho_G = 0.46$, $P < 0.05$), and tricep skinfold ($\rho_G = 0.60$, $P < 0.05$); and mean LDL size with BMI ($\rho_G = -0.36$), waist ($\rho_G = -0.42$), and subscapular and tricep skinfolds ($\rho_G = -0.44$ and -0.43 , respectively) ($P < 0.005$). These results show that a common set of genes is influencing LDL size and obesity-related factors in Alaskan Eskimos. *Am. J. Hum. Biol.* 18:525–531, 2006. © 2006 Wiley-Liss, Inc.

Mortality rates due to cardiovascular disease (CVD) among Alaskan Eskimos have historically been low. This lower incidence was attributed to a greater dietary intake of fish oils, which are excellent sources of omega-3 fatty acids (Kromhout et al., 1985). In the past decade, this trend has changed with the Westernization of diet and lifestyle. Heart disease is now the third leading cause of death in Alaskan Eskimos, behind accidental injuries and cancer (Day and Lanier, 2003).

Obesity, particularly in the abdominal region of the body, is one of the major risk factors for CVD (Rexrode et al., 1998), as an obese person is 2–3 times more likely to exhibit CVD than a nonobese person (Stein and Colditz, 2004). Approximately 33% and 16% of Alaskan Eskimo women and men are obese,

respectively. This is of great concern, particularly for women, as it was observed that, in comparison to Americans of other races, Alaskan Eskimo women have a greater prevalence of obesity (Risica et al., 2000a). Also, women from this region have a visceral adiposity comparable to the world's highest reported averages (Risica et al., 2000b). Increased fat deposition in the abdominal area enhances the flow

Grant sponsor: National Institutes of Health; Grant numbers: U01 HL 64244, MH 59490.

*Correspondence to: V. Saroja Voruganti, Department of Genetics, Southwest Foundation for Biomedical Research, PO Box 760549, San Antonio, TX 78245-0549. E-mail: svorugan@darwin.sfbr.org

Received 31 October 2005; Revision received 8 February 2006; Accepted 13 February 2006

Published online in Wiley InterScience (www.interscience.wiley.com). DOI 10.1002/ajhb.20527

of free fatty acids into the liver via the portal vein, thus increasing the production of triglycerides in the liver (Ginsberg, 2000). An elevation in triglycerides occurs with the simultaneous suppression of high-density lipoprotein (HDL) production and low-density lipoprotein (LDL) particle size (Sonnenberg et al., 2004).

Low-density lipoproteins transport triglycerides and cholesterol esters in the plasma (Nabel, 2003), and are composed of well-defined and different-sized subfractions. As per nuclear magnetic resonance (NMR) spectroscopy, these particles are differentiated into LDL1–LDL3 based on their size, with LDL1 being the smallest (Otvos et al., 1992). Small, dense LDLs are established CVD risk factors, and are associated strongly with other lipid and obesity-related phenotypes. The significance of smaller LDL particles is that they have less affinity for their receptors (Herron et al., 2004), and remain longer in the plasma, providing a greater chance of being deposited on arterial walls (Rainwater et al., 2003). Furthermore, small LDLs are more susceptible to oxidation than other forms. Oxidized LDL demonstrates greater atherogenicity because of its increased uptake by macrophages (Herron et al., 2004).

LDL size may be a better tool for measuring individual CVD risk than total LDL (Johnson et al., 2004). Generally, the plasma level of LDL is used as one of the primary criteria for the diagnosis of CVD. However, two individuals may have the same level of LDL but differ in their susceptibility to atherosclerosis, due to the different concentrations of small and large LDLs. The size of LDL is under substantial genetic influence. Heritabilities in family-based studies showed that genetic factors are responsible for 30–60% of the variation in LDL size (Bosse et al., 2004). On the other hand, the influence of other phenotypes such as weight, body fat, triglycerides, and HDL cholesterol on LDL size cannot be ignored (Kang et al., 2002). For example, LDL size is sensitive to changes in triglyceride and HDL concentrations (James et al., 1997), and these two phenotypes are considered to be independent predictors of LDL size (Kang et al., 2002). Adiposity measures such as body mass index (BMI), waist-to-hip ratio (WHR), and subscapular-to-triceps ratio (STR) were negatively associated with LDL size (Rainwater et al., 1999). In Rainwater et al. (1999), the strongest correlation of LDL size was with WHR (an indicator of central fat deposition). Thus, the purpose of this study was to investigate ge-

netic and environmental correlations between LDL and its subfractions and obesity phenotypes in Alaskan Eskimos.

METHODS

Experimental design

The Genetics of Coronary Artery Disease in Alaska Natives (GOCADAN) Study involves populations from villages in the Norton Sound region on the northwestern coast of Alaska. Two GOCADAN investigators visited each village, conducted interviews, and explained the nature of the study to household members. Participants ($n = 1,214$) over 18 years of age were recruited and demographics were collected during the first visit. Participants attended clinics for a medical examination and blood draw after a 12-hr fast. Also, information regarding medical history, dietary intake, current physical activity, and anthropometrics was obtained. Blood was drawn by venipuncture, and samples were stored in aliquots at -80°C for future analyses. This study was approved by Institutional Review Boards from all participating institutions, and informed consent was obtained from participants.

Study population

Participants resided in the villages of Teller, Golovin, Elim, Koyuk, Shaktolik, Unalakleet, White Mountain, Brevig Mission, and Nome, and were primarily Inupiat Eskimo (Howard et al., 2005). Relative pairs used in this study are listed in Table 1. For these analyses, data were available for 954 individuals (420 men and 534 women) ranging in age from 17–92, with an average age of 43 ± 0.5 years.

Demographic and phenotypic data

Demographic and genealogical data collected during the surveys included names, genders, dates, and places of birth, current home of the participant and his/her spouse, and first-degree relatives of all household members. This information was sent to the Southwest Foundation for Biomedical Research (SFBR, San Antonio, TX) and entered into the PEDSYS pedigree database system (Dyke, 1994).

Anthropometric measurements included height, weight, and waist and hip circumference. Height was measured to the nearest quarter inch while a participant was standing, using a vertical mounted ruler. Weight was determined to the nearest tenth of a pound, using a balance scale (model 683-P, Detecto).

TABLE 1. Relative pairs in this study

Relationship	Number of pairs
Unrelated	26,712
Self	1,033
Parent-offspring	436
Siblings	328
Grandparent-grandchild	141
Avuncular	396
Half-siblings	274
Great grandparent-grandchild	6
Grand avuncular	20
Half-avuncular	458
First cousins	215
Half-grand avuncular	89
First cousins, once removed	72
Half-first cousins	391
Half-first cousins, once removed	299
Half-second cousins	84
Double-first cousins	1
Half-second cousins, once removed	39
Half-first cousins, twice removed	23
Half-great-grand avuncular	8
Double-half grand avuncular	1
Half-third cousins	3
First cousins and half-second cousins	2
Half-first cousins and half-first cousins, once removed	2
Unknown relationship	16
Grandparent-grandchild and half-grand avuncular	1
Self-unknown inbred mating type	1
Half-avuncular and half-first cousins, once removed	1
Half-first cousins and half-second cousins	2
Half-siblings and half-second cousins	2
Total	31,056

Waist and hip circumferences were measured to the nearest quarter inch at the level of the umbilicus with the subject in supine position and at the level of maximal protrusion of the gluteal muscles, respectively. Measurements were converted to International System of Units (SI) units.

Skinfolds (subscapular and triceps) were measured to the nearest millimeter with a Lange caliper. The subscapular measurement was taken 1 cm inferior to the angle of the right scapula while the participant was standing with shoulders relaxed and arms hanging loosely at his/her sides. The triceps skinfold was determined directly over the right triceps muscle, halfway between the acromial and olecranon processes, with the arms hanging comfortably at the participant's side.

Total cholesterol, HDL cholesterol, LDL cholesterol, very-low-density lipoprotein (VLDL) cholesterol, and triglycerides were measured by an autoanalyzer (Hitachi 717, Amposta, Spain). Lipoprotein subfractions type, size,

and concentrations were measured by NMR spectroscopy (Otvos et al., 1992). This method is based on the NMR signal emitted by the terminal methyl groups on the lipids contained in the particle core and the shell. Subfractions of LDL were categorized based on their sizes: small (18.3–19.7 nm), medium (19.8–21.2 nm), or large (21.3–23 nm) LDL.

Statistical genetic methods

Quantitative genetic analyses were performed utilizing the maximum likelihood-based variance decomposition method, implemented in the computer program SOLAR (Almasy and Blangero, 1998). According to classical quantitative genetics principles, the total phenotypic variance (σ_P^2) can be partitioned into its genetic components (σ_G^2) and nongenetic or environmental components (σ_E^2):

$$\sigma_P^2 = \sigma_G^2 + \sigma_E^2.$$

The heritability of a phenotype refers to the ratio of the variance contributed by additive genetic effects to the total phenotypic variance, and is denoted by $h^2 = \sigma_G^2/\sigma_P^2$ (Hopper and Mathews, 1982). For heritabilities of traits related to LDL size and obesity, likelihood ratio tests were used to obtain P -values. In the process, the null hypothesis, in which the additive genetic variance (σ_G^2) equals zero, was tested against an alternate hypothesis in which the additive genetic variance was estimated. Twice the difference in logarithmic likelihoods was distributed asymptotically as a $\frac{1}{2}\chi^2$ mixture of a χ^2 variable, with 1 degree of freedom and a point mass at zero (Self and Liang, 1987).

Univariate quantitative genetic analysis was used to estimate residual heritability, using sex, sex-specific age, and age squared as covariates. Bivariate genetic analysis was conducted to examine the genetic correlations between LDL, its subfractions, and obesity-related factors.

The phenotypic correlation between phenotypes can be expressed in terms of core genetic and environmental correlations:

$$\rho_P = \rho_G(\sqrt{h_1^2}\sqrt{h_2^2}) + \rho_E(\sqrt{(1-h_1^2)} \times \sqrt{(1-h_2^2)})$$

where h_1^2 and h_2^2 are the heritabilities of the two phenotypes being studied, and ρ_G and ρ_E are the additive genetic and environmental correlations between traits, respectively (Lange and Boehnke, 1983).

TABLE 2. Descriptive statistics for anthropometrics and lipids in GOCADAN ($n = 954$)¹

Phenotype	Men ²	Women ²	P-value
Obesity			
Weight (kg)	76.3 (0.73)	71.1 (0.69)	<0.01
BMI (kg/m ²)	26.4 (0.23)	28.5 (0.26)	<0.01
Waist circumference (cm)	87.4 (0.59)	88.6 (0.58)	NS
Skinfold (mm)			
Subscapular	14.6 (0.36)	20.8 (0.40)	<0.01
Triceps	12.8 (0.30)	21.7 (0.33)	<0.01
LDL concentration (mg/dl)			
Small	10.5 (0.69)	6.6 (0.51)	<0.01
Medium	22.0 (1.23)	17.7 (1.13)	<0.01
Large	65.4 (2.16)	76.3 (2.00)	<0.01
Total	117.2 (1.74)	114.7 (1.52)	NS
Mean LDL size	21.2 (0.034)	21.5 (0.03)	NS
Other lipids (mg/dl)			
Chylomicrons	0.16 (0.02)	0.13 (0.02)	NS
IDL	2.9 (0.25)	3.8 (0.25)	<0.05
HDL	54.7 (0.79)	64.6 (0.75)	<0.01
Triglycerides	121.5 (2.93)	123.0 (2.54)	NS

¹BMI, body mass index; LDL, low-density lipoprotein; IDL, intermediate-density lipoprotein; HDL, high-density lipoprotein.

²Mean \pm SEM.

An unrestricted model, where all parameters are estimated, is compared with two restricted models: one in which the genetic correlation is constrained to zero ($\rho_G = 0$, or no shared gene effects), and the other in which the genetic correlation is constrained to 1 ($\rho_G = 1$, or complete pleiotropy). The basic premise of this model is that the genes controlling expressions of the two traits completely overlap with each other. The alternative hypothesis is that some genes influencing trait 1 do not affect trait 2, and vice versa. Additive genetic correlations that are significantly different from zero might suggest pleiotropy (same set of genes influencing two or more traits).

An independent Student's *t*-test was applied to evaluate comparisons between men and women, using SPSS (version 10.0, SPSS, Inc., Chicago, IL).

RESULTS

Descriptive statistics of anthropometrics and lipids are shown in Table 2. Sex-specific comparisons showed that men weighed more but had lower BMI, and smaller skinfold thicknesses, than women. Lipoprotein measurements displayed higher circulating small and medium LDL in men compared to women. Women, on the other hand, had greater plasma concentrations of large LDL, intermediate density lipoprotein (IDL), and HDL cholesterol than men. Plasma triglyceride levels

TABLE 3. Heritabilities (h^2) of anthropometric and lipid-related measurements

Phenotype	h^2	SEM	P-value	Effects of covariates
Obesity				
Weight (kg)	0.64	0.06	<0.0001	0.044
BMI (kg/m ²)	0.57	0.07	<0.0001	0.037
Waist circumference (cm)	0.55	0.07	<0.0001	0.030
Skinfold (mm)				
Subscapular	0.53	0.07	<0.0001	0.124
Triceps	0.47	0.07	<0.0001	0.269
LDL concentration (mg/dl)				
Small	0.20	0.06	<0.0001	0.039
Medium	0.31	0.08	<0.0001	0.026
Large	0.30	0.07	<0.0001	0.058
Total	0.36	0.07	<0.0001	0.108
Mean LDL size (nm)	0.45	0.09	<0.0001	0.075
Other lipids (mg/dl)				
HDL	0.51	0.07	<0.0001	0.117
Triglycerides	0.31	0.08	<0.0001	0.020

did not differ significantly between genders. In the univariate analysis, significant heritabilities were exhibited by all obesity-related (47–64%) and LDL-related (20–45%) phenotypes (Table 3).

Genetic correlations between LDL, its subfractions, and obesity-related anthropometric measures are shown in Table 4. Significant and positive genetic correlations were observed between small to medium LDL and obesity-related factors. Large LDL was not genetically correlated with any obesity-related factors. Table 5 shows the genetic correlations between LDL size and obesity-related traits. Mean LDL size was significantly correlated with all obesity-related traits. When these correlations were further analyzed, adjusted for effects of triglycerides, the correlations remained significant, but ρ_G values decreased. Genetic correlations between LDL subfractions, their mean size, and other lipids are depicted in Table 6. Small LDL displayed negative correlations with HDL. Medium LDL had significant associations with HDL (negative) and triglycerides (positive). Large LDL was correlated positively with HDL, and with a nonsignificant negative relationship with triglycerides. Mean LDL size was correlated positively with HDL cholesterol, and negatively with triglycerides. All tests for complete pleiotropy were significantly different from 1, suggesting that no two traits tested were governed by completely overlapping genes, and that additional nonshared genes were also responsible for the variation in each of these traits.

TABLE 4. Genetic correlations (ρ_G) between plasma concentrations of LDL subfractions and obesity-related measures

LDL concentration (mg/dl)	Phenotype	ρ_G^1	P-value
Small	Weight	0.65 (0.27)	<0.05
	BMI	0.67 (0.25)	<0.01
	Waist	0.80 (0.22)	<0.01
	Subscapular skinfold	0.93 (0.27)	<0.01
	Triceps skinfold	0.78 (0.22)	<0.01
Medium	Weight	0.46 (0.22)	<0.05
	BMI	0.52 (0.22)	<0.05
	Waist	0.46 (0.23)	<0.07
	Subscapular skinfold	0.44 (0.22)	NS
	Triceps skinfold	0.60 (0.19)	<0.05
Large	Weight	-0.29 (0.18)	NS
	BMI	-0.21 (0.19)	NS
	Waist	-0.13 (0.19)	NS
	Subscapular skinfold	-0.21 (0.20)	NS
	Triceps skinfold	-0.27 (0.20)	NS
Total	Weight	0.22 (0.16)	NS
	BMI	0.34 (0.16)	<0.05
	Waist	0.49 (0.16)	<0.005
	Subscapular skinfold	0.23 (0.17)	NS
	Triceps skinfold	0.07 (0.18)	NS

¹Mean \pm SEM.

DISCUSSION

This study demonstrated significant genetic correlations between small-to-medium-sized LDL particles and obesity-related factors, indicating that a common set of genes might be regulating these phenotypes in Alaskan Eskimos. Obesity, its related factors, and LDL are strong cardiovascular risk factors. However, the mode of action is complex. Weisberg et al. (2003) suggested that macrophage infiltration into the adipose tissue might be the basis for the development of several obesity-related pathologies. According to their study, increasing adiposity tends to raise the number of macrophages in adipose tissue (Weisberg et al., 2003). These bone marrow-derived cells induce a rise in proinflammatory markers and fat accumulation in blood vessels (Lehrke and Lazar, 2004), thereby increasing the risk of CVD. In addition, visceral adiposity results in a greater flow of free fatty acids to the liver, where these fatty acids tend to alter lipid levels, such as increase triglycerides, lowered HDL levels, and reduction in LDL size (Sonnenberg et al., 2004).

TABLE 5. Genetic correlations (ρ_G) between mean LDL size and obesity-related traits (with and without triglyceride as covariate)

LDL size (nm)	Phenotype	ρ_G^1	P-value
Age and sex as covariates	Weight	-0.37 (0.11)	<0.005
	BMI	-0.36 (0.11)	<0.005
	Waist	-0.42 (0.11)	<0.005
	Subscapular skinfold	-0.44 (0.11)	<0.005
Age, sex, and triglycerides as covariates	Triceps skinfold	-0.43 (0.13)	<0.005
	Weight	-0.26 (0.13)	<0.06
	BMI	-0.27 (0.13)	<0.06
	Waist	-0.34 (0.13)	<0.05
	Subscapular skinfold	-0.37 (0.13)	<0.01
Triceps skinfold	-0.35 (0.14)	<0.05	

¹Mean \pm SEM.TABLE 6. Genetic correlations (ρ_G) between plasma concentrations of LDL subfractions and other lipids

LDL concentration (mg/dl)	Other lipids (mg/dl)	ρ_G^1	P-value
Small	HDL	-0.49 (0.24)	<0.07
	Triglycerides	0.24 (0.34)	NS
Medium	HDL	-0.65 (0.20)	<0.01
	Triglycerides	0.88 (0.19)	<0.01
Large	HDL	0.58 (0.16)	<0.01
	Triglycerides	-0.35 (0.23)	NS
Total	HDL	-0.07 (0.18)	NS
	Triglycerides	0.18 (0.25)	NS
Mean LDL size	HDL	0.50 (0.10)	<0.001
	Triglycerides	-0.72 (0.07)	<0.001

¹Mean \pm SEM.

The size of LDL is a major determinant in the risk for CVD, and varies with age, sex, and central obesity (Austin et al., 2003). Small, dense LDL is considered the most crucial, as it is more atherogenic than other forms (Berneis et al., 2005). Studies by Freedman et al. (2004), Nikkila et al. (1996), and Johnson et al. (2004) observed that men had lower levels of large LDL and higher concentrations of small-to-medium LDL than women, indicating an increased risk of CVD. These same results were echoed in our study, with men exhibiting elevated levels of smaller LDLs. In contrast, women had higher levels of both small and large LDL subfractions in a study by Friedlander et al. (2000).

In this study, the additive genetic heritability for LDL subfractions and their mean size ranged from 0.20–0.45, indicating a substantial influence of genetic factors on LDL size. Similar estimates for LDL cholesterol, ranging

from 0.38–0.62, were reported in other populations that included Caucasian-American families from the Take Off Pounds Sensibly (TOPS) Study (Sonnenberg et al (2004), African American and Hispanic families (Hokanson et al., 2003), and Amish families (Pollin et al., 2004). Mean LDL size was also shown to be heritable in several studies, mainly, families from the Genetic Epidemiology of Hypertriglyceridemia (GET) Study (Edwards et al., 1999; Austin et al., 2004), participants in the Genetic Epidemiology Network of Arteriopathy (GENOA) Study (Kullo et al., 2005), Ashkenzi Jews from the Longevity Genes Project (Barzilai et al., 2003), and families from San Antonio Family Heart Study (SAFHS) (Rainwater et al., 2001).

Small LDL had strong genetic correlations with weight and BMI, suggesting that 42% ($\rho_G^2 = (0.65)^2 = 0.42$) to 47% ($\rho_G^2 = (0.67)^2 = 0.47$) of additive genetic variance in small LDL is shared with weight and BMI. Abdominal obesity poses a greater CVD risk than other types, irrespective of ethnicity and gender. Individuals with large waist circumferences have a greater risk of developing CVD than those with a normal or smaller waist (Vega, 2002). In the present study, the genetic correlations between small LDL and waist circumference was 0.80, indicating that 64% ($\rho_G^2 = (0.80)^2 = 0.64$) of the additive genetic contribution to small LDL is shared with waist circumference. Other studies also found that abdominal adiposity, as measured by waist circumference, was positively associated with CVD, despite controlling for BMI (Rexrode et al., 1998). Similar results were presented by James et al. (1997) in a group of Caucasian men and women. Their study observed positive correlations between small LDL and abdominal adiposity, as measured by waist-to-hip ratio. Medium LDL showed modest genetic correlations with weight, BMI, and waist circumference, yet approximately 25% of the additive genetic variance in these phenotypes could be attributable to shared genes. However, large LDL showed negative, but nonsignificant, genetic and environmental correlations with obesity-related factors. Rainwater et al. (1999), in the SAFHS, also reported a negative relationship of large LDL with obesity-related phenotypes.

Obesity is associated with high triglycerides and low HDL (James et al., 1997). These altered lipid levels, coupled with decreased LDL size, are thought to result from the overproduction of VLDL in the liver, which is regulated by the flow of free fatty acids from vis-

ceral adipose tissue. The negative genetic correlation between small-to-medium LDL and HDL in this study may indicate substantial genetic influence on altered lipid levels.

LDL size also is influenced, to a large extent, by plasma triglycerides (Wallace et al., 2000). In Edwards et al. (1999) and Kullo et al. (2005), significant positive genetic correlations were observed between LDL size and HDL cholesterol, and a negative correlation between LDL size and triglycerides. In the present study, positive genetic correlations between triglycerides and medium LDL were observed ($\rho_G^2 = (0.88)^2 = 0.77$), suggesting that 77% of the additive genetic variance in medium LDL is shared with triglycerides. However, mean LDL size was positively correlated with HDL cholesterol, and negatively with triglycerides, indicating that the same set of genes responsible for the increase in triglycerides may be responsible for the decrease in LDL size. The relationship between triglycerides and LDL size is further corroborated by the fact that quantitative trait loci (QTLs) for both of these phenotypes were linked to the same region on human chromosome 7q35–q36 in the TOPS Study (Sonnenberg et al., 2004). Additionally, we investigated genetic correlations between LDL size and obesity-related traits, with triglycerides as a covariate. This is important, as both LDL size and adiposity are highly sensitive to changes in triglycerides (James et al., 1997). The genetic correlations remained significant despite a decrease in ρ_G , suggesting that the correlations may be independent of the effect of triglycerides.

Significant genetic correlations of LDL subfractions with obesity-related factors and other lipids demonstrate considerable genetic influence on these phenotypes. Understanding the core of these genetic relationships and the identification of chromosomal regions containing genes that control variation in these phenotypes will help gain insights into the complex nature of cardiovascular disease.

ACKNOWLEDGMENTS

We thank the participants of the Genetics of Coronary Artery Disease in Alaska Natives (GOCADAN) Study for their generous participation.

LITERATURE CITED

- Almasy L, Blangero J. 1998. Multipoint quantitative-trait linkage analysis in general pedigrees. *Am J Hum Genet* 62:1198–1211.

- Austin MA, Edwards KL, Monks SA, Koprowicz KM, Brunzell JD, Motulsky AG, Mahaney MC, Hixson JE. 2004. Genome-wide scan for quantitative trait loci influencing LDL size and plasma triglycerides in familial hypertriglyceridemia. *J Lipid Res* 44:2161–2168.
- Barzilai N, Atzmon G, Schechter C, Schaefer EJ, Cupples AL, Lipton R, Cheng S, Shuldiner AR. 2003. Unique lipoprotein phenotype and genotype associated with exceptional longevity. *JAMA* 290:2030–2040.
- Berneis K, Jeanneret C, Muser J, Felix B, Miserez AR. 2005. Low-density lipoprotein size and subclasses are markers of clinically apparent and non-apparent atherosclerosis in type 2 diabetes. *Metabolism* 54:227–234.
- Bosse Y, Perusse L, Vohl MC. 2004. Genetics of LDL particle heterogeneity: from genetic epidemiology to DNA-based variations. *J Lipid Res* 45:1008–1026.
- Day GE, Lanier AP. 2003. Alaskan mortality, 1979–1998. *Public Health Rep* 118:518–530.
- Dyke B. 1994. PEDSYS, a pedigree data management system user's manual. Population Genetics Laboratory technical report no. 2, 2nd ed. San Antonio: Southwest Foundation for Biomedical Research. 226 p.
- Edwards KL, Mahaney MC, Motulsky AG, Austin MA. 1999. Pleiotropic genetic effects on LDL size, plasma triglyceride, and HDL cholesterol in families. *Arterioscler Thromb Vasc Biol* 19:2456–2464.
- Freedman DS, Otvos JD, Jeyarajah EJ, Shalaurova I, Cupples LA, Parise H, D'Agostino RB, Wilson PWF, Schaefer EJ. 2004. Sex and age differences in lipoprotein subclasses measured by nuclear magnetic resonance spectroscopy: The Framingham Study. *Clin Chem* 50:1189–1200.
- Friedlander Y, Kidron M, Caslake M, Lamb T, McConnell M, Bar-On H. 2000. Low density lipoprotein particle size and risk factors of insulin resistance syndrome. *Atherosclerosis* 148:141–149.
- Ginsberg HN. 2000. Insulin resistance and cardiovascular disease. *J Clin Invest* 106:453–458.
- Herron KL, Lofgren IE, Sharman M, Volek JS, Fernandez ML. 2004. High intake of cholesterol results in less atherogenic low-density lipoprotein particles in men and women independent of response classification. *Metabolism* 53:823–830.
- Hokanson JE, Langefeld CD, Mitchell BD, Lange LA, Goff DC Jr, Haffner SM, Saad MF, Rotter JI. 2003. Pleiotropy and heterogeneity in the expression of atherogenic lipoproteins: the IRAS Family Study. *Hum Hered* 55:46–50.
- Hopper JL, Mathews JD. 1982. Extensions to multivariate normal models for pedigree analysis. *Ann Hum Genet* 46:373–383.
- Howard BV, Devereux RB, Cole SA, Davidson M, Dyke B, Ebbesson SOE, Epstein SE, Robinson DR, Jarvis, Kaufman DJ, Laston S, MacCluer JW, Okin PM, Roman MJ, Romnesko T, Ruotolo G, Swenson M, Wenger CR, Williams-Blangero S, Zhu J, Saccheus C, Fabsitz RR, Robbins DC. 2005. A genetic and epidemiologic study of cardiovascular disease in Alaska Natives (GOCADAN): design and methods. *Int J Circumpolar Health* 64:206–221.
- James RW, Brulhart-Meynet MC, Lehmann T, Golay A. 1997. Lipoprotein distribution and composition in obesity: their association with central adiposity. *Int J Obes* 21:1115–1120.
- Johnson JL, Slentz CA, Duscha BD, Samsa GP, McCartney JS, Houmard JA, Kraus WE. 2004. Gender and racial differences in lipoprotein subclass distributions: the STRRIDE study. *Atherosclerosis* 176:371–377.
- Kang HS, Gutin B, Barbeau P, Litekar MS, Allison J, Le N-A. 2002. Low-density lipoprotein particle size, central obesity, cardiovascular fitness, and insulin resistance syndrome markers in obese youths. *Int J Obes* 26:1030–1035.
- Kromhout D, Bosscheiter EB, Coulander CL. 1985. The inverse relation between fish consumption and 20-year mortality from coronary heart disease. *N Engl J Med* 312:1205–1209.
- Kullo IJ, Andrade MD, Boerwinkle E, McConnell JP, Kardia SLR, Turner ST. 2005. Pleiotropic genetic effects contribute to the correlation between HDL cholesterol, triglycerides and LDL particle size in hypertensive sibships. *Am J Hypertens* 18:99–103.
- Lange K, Boehnke M. 1983. Extensions to pedigree analysis. IV. Covariance components models for multivariate traits. *Am J Med Genet* 14:513–524.
- Lehrke M, Lazar MA. 2004. Inflamed about obesity. *Nat Med* 10:126–127.
- Nabel EG. 2003. Cardiovascular disease. *N Engl J Med* 349:60–72.
- Nikkila M, Pitkajarvi T, Koivula T, Solakivi T, Lehtimäki T, Laippala P, Jokela H, Lehtomäki E, Seppä K, Sillanauke P. 1996. Women have a larger and less atherogenic low density lipoprotein particle size than men. *Atherosclerosis* 119:181–190.
- Otvos JD, Jeyarajah EJ, Bennett DW, Krauss RM. 1992. Development of a proton nuclear magnetic resonance spectroscopic method for determining plasma lipoprotein concentrations and subspecies distributions from a single, rapid measurement. *Clin Chem* 38:1632–1638.
- Pollin TI, Hsueh WC, Steinle NI, Snitker S, Shuldiner AR, Mitchell BD. 2004. A genome-wide scan of serum lipid levels in the Old Order Amish. *Atherosclerosis* 173:89–96.
- Rainwater DL, Mitchell BD, Comuzzie AG, Haffner SM. 1999. Relationship of low-density lipoprotein particle size and measures of adiposity. *Int J Obes* 23:180–189.
- Rainwater DL, Martin LJ, Comuzzie AG. 2001. Genetic control of coordinated changes in HDL and LDL size phenotypes. *Arterioscler Thromb Vasc Biol* 21:1829–1833.
- Rainwater DL, Kammerer CM, Mahaney MC, Rogers J, Cox LA, Schneider JL, Vandeberg JL. 2003. Localization of genes that control LDL size fractions in baboons. *Atherosclerosis* 168:15–22.
- Rexrode KM, Carey VJ, Hennekens CH, Walters EE, Colditz GA, Stampfer MJ, Willett WC, Manson JE. 1998. Abdominal adiposity and coronary heart disease in women. *JAMA* 280:1843–1848.
- Risica PM, Schraer C, Ebbesson SOE, Nobmann ED, Caballero B. 2000a. Overweight and obesity among Alaskan Eskimos of the Bering Straits region: the Alaska Siberia Project. *Int J Obes* 24:939–944.
- Risica PM, Ebbesson SOE, Schraer CD, Nobmann ED, Caballero BH. 2000b. Body fat distribution in Alaskan Eskimos of the Bering Straits region: the Alaskan Siberia Project. *Int J Obes* 24:171–179.
- Self SG, Liang KY. 1987. Asymptotic properties of maximum likelihood estimators and likelihood ratio tests under non-standard conditions. *J Am Stat Assoc* 82:605–610.
- Sonnenberg GE, Krakower GR, Martin LJ, Olivier M, Kwitek AE, Comuzzie AG, Blangero J, Kissebah AH. 2004. Genetic determinants of obesity-related lipid traits. *J Lipid Res* 45:610–615.
- Stein CJ, Colditz GA. 2004. The epidemic of obesity. *J Clin Endocrinol Metab* 89:2522–2525.
- Vega GL. 2002. Cardiovascular outcomes for obesity and metabolic syndrome. *Obes Res* 10:27–32.
- Wallace AJ, Humphries SE, Fisher RM, Mann JI, Chisholm A, Sutherland WHF. 2000. Genetic factors associated with response of LDL subfractions to change in the nature of dietary fat. *Atherosclerosis* 149:387–394.
- Weisberg SP, McCann D, Desai M, Rosenbaum M, Leibel RL, Ferrante AW Jr. 2003. Obesity is associated with macrophage accumulation in adipose tissue. *J Clin Invest* 112:1796–1808.