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Effects of dietary saturated fat on LDL subclasses and apolipoprotein CIII in men

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Abstract

Background/Objectives—Small dense LDL particles and apolipoprotein (apo) CIII are risk factors for cardiovascular disease (CVD) that can be modulated by diet, but there is little information regarding the effects of dietary saturated fat on their plasma levels. We tested the effects of high vs. low saturated fat intake in the context of a high beef protein diet on levels and composition of LDL subclasses and on apoCIII levels in plasma and LDL.

Subjects/Methods—Following consumption of a baseline diet (50% CHO, 13% protein, 38% total fat, 15% saturated fat) for 3 wk, 14 healthy men were randomly assigned to two reduced carbohydrate high beef protein diets (31% CHO, 31% protein, 38% fat) that differed in saturated fat content (15% vs. 8%) for 3 wk each in a crossover design.

Results—The high saturated fat diet resulted in higher mass concentrations of buoyant LDL I, medium density LDL II and dense LDL III, but not the very dense LDL IV; and significant increases in plasma and LDL apoCIII concentration of 9.4% and 33.5%, respectively. The saturated fat-induced changes in LDL apoCIII were specifically correlated with changes in apoCIII content of LDL IV.

Conclusions—Taken together with previous observations, these findings suggest that, at least in the context of a lower carbohydrate high beef protein diet, high saturated fat intake may increase CVD risk by metabolic processes that involve apoCIII.

Keywords

saturated fat; low-density lipoprotein; apolipoprotein CIII; cardiovascular disease; diet; LDL subfractions

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Introduction

It is well established that increased saturated fat intake raises LDL cholesterol (1). However, LDL is comprised of a spectrum of distinct particle subclasses differing in metabolic properties and atherogenic potential (2), and effects of saturated fat on these subclasses have been less extensively evaluated. Lipoprotein profiles characterized by smaller, denser LDL particles are associated with higher CVD risk than those with predominately larger LDL (2–5). Small dense LDL have a number of properties that may contribute to atherosclerosis risk, including lower affinity for LDL receptors, greater binding to arterial proteoglycans and higher oxidative susceptibility than more buoyant LDL (6). It has been reported that very small LDL are particularly strongly associated with coronary atherosclerosis progression (7), a property consistent with the recent finding that a single nucleotide polymorphism (SNP) strongly associated with plasma LDL cholesterol and risk of myocardial infarction is specifically related to plasma levels of very small LDL (8).

Intake of saturated fat, particularly myristic (14:0) and palmitic (16:0) acids, but not stearic acid (18:0), has been found to correlate with levels of large LDL I (9). Moreover, in the setting of a low carbohydrate mixed protein diet, the higher levels of LDL that resulted from increased saturated fat intake were found to be entirely due to increases in larger LDL particles (10). Yet we recently found that consumption of increased saturated fat in conjunction with a high beef protein, low carbohydrate diet resulted in an increase in LDL particles across the full size spectrum, raising the possibility that the effects of saturated fat on LDL particles may depend on dietary context (11). We here describe analyses of lipid and protein composition of LDL subfractions isolated from a subset of participants in that study, with the aim of determining whether higher intake of saturated fat resulted in compositional changes in specific LDL particles. We also examined the effects of higher vs. lower saturated fat on LDL content of apoCIII, an important regulator of lipoprotein metabolism by virtue of its capacity to inhibit lipoprotein lipase (LPL) activity and reduce receptor-mediated clearance of apoB-containing particles (12, 13). The association of apoCIII with apoB-containing lipoproteins has repeatedly been linked with increased CVD risk (14–18), an effect that may be related to the metabolic effects and/or its pro-inflammatory properties of apoCIII (19–21). Finally, we tested for diet-induced changes in LDL content of apoE, a determinant of receptor-mediated plasma lipoprotein clearance (22, 23).

Subjects and methods

Study design and diets

These analyses were performed in a subgroup of 14 healthy male participants in a controlled, randomized, crossover dietary intervention trial aimed at evaluating plasma lipid and lipoprotein responses to high vs. low saturated fat intake in the context of replacement of carbohydrate with protein derived to a large extent from beef (11). All participants consumed a baseline diet for 3 weeks, after which they were assigned in random order to a low carbohydrate high beef protein diet that was either high (15%) or low (8%) in saturated fat for 3 weeks. Each 3 week experimental diet period was separated by a 2 week washout during which participants resumed consumption of their habitual diet. The experimental

diets were designed to be low in carbohydrate and high in beef protein, and to differ in saturated fat content (Table 1).

Diets were prepared in the metabolic kitchen of the University of California, San Francisco (UCSF) Clinical and Translational Studies Institute (CTSI). After preparation, entrees were frozen and delivered to participants on a weekly basis. Participants were provided with lunch and dinner meals and were asked to prepare their own breakfasts and snacks according to menus developed in accordance with their assigned diet. Participants were weighed weekly and energy intake was adjusted if weight deviated by more than ± 3%. Dietary adherence was promoted through regular telephone contacts and weekly meetings with the dietitians. The nutrient composition of the diets was assessed using the Nutrition Data System for Research (NDSR) 2007 MN Food and Nutrient Database (Nutrition Coordinating Center, University of Minnesota, Minneapolis, MN) and confirmed by compositional analysis of daily menus (Covance Inc, Kalamazoo, MI). Daily use of a pedometer allowed monitoring of participants' physical activity, which was required to remain stable for the duration of the study.

Study participants

Participants were recruited from our extensive recruitment database and the community primarily through internet advertisements. They were healthy, non-smoking men 18 years of age, free of chronic disease in the last 5 years, and not taking medications known to affect lipid metabolism. Other eligibility criteria included a BMI between 20–35 kg/m^2, total cholesterol and LDL cholesterol less than the 95th percentile for their age and gender, triglyceride < 500 mg/dL, fasting blood glucose < 126 mg/dL, resting blood pressure < 150/90 mmHg and hematocrit 36%. Participants were asked to refrain from alcohol and dietary supplements for the duration of the study. All participants gave informed consent under a protocol approved by the Institutional Review Board of Children's Hospital and Research Center Oakland. The study is registered at clinicaltrials.gov (NCT00852267). One participant was eliminated from the present analysis due to a triglyceride level > 2 SD above the mean for all diets (baseline: 270 mg/dL, low saturated fat (LSF): 171 mg/dL and high saturated fat (HSF): 451 mg/dL), since this may affect apoCIII metabolism.

Laboratory measurements

Fasting plasma samples were prepared from venous blood collected in tubes containing Na$_2$EDTA (1.4 g/L) and a preservative cocktail of protease and bacterial inhibitors. Blood and plasma were kept at 4oC throughout processing. Plasma total cholesterol and triglyceride concentrations were determined by enzymatic procedures on an Express 550 Plus analyzer (Ciba Corning, Oberlin, OH). HDL cholesterol was measured after dextran sulfate precipitation of plasma (24), and LDL cholesterol was calculated from the formula of Friedewald et al (25). Measurements were regularly monitored by the standardization program of the Centers for Disease Control-National Heart, Lung and Blood Institute. Plasma apoB concentrations were measured by an immunoturbidometric assay (Bacton Assay Systems, San Marcos, CA, and Express 550 Plus analyzer, Ciba Corning, Oberlin, OH). ApoE and apoCIII were measured in triplicate by sandwich-style ELISA with commercially available goat anti-human apoE and apoCIII, respectively (International

Immunology Corp, Murrieta, CA). HDL apoCIII was measured in the supernatant obtained after dextran sulfate precipitation (24) of apoB-containing lipoproteins. The concentration of apoCIII in apoB-containing lipoproteins (non-HDL apoCIII) was calculated by difference (plasma – HDL apoCIII concentration). Glucose concentrations were measured enzymatically (Express 550 Plus Analyzer, Ciba Corning, Oberlin, OH). Insulin concentrations were measured with ELISA (Millipore, Billerica, MA). The Homeostasis Model Assessment of Insulin Resistance (HOMA-IR) was calculated from glucose and insulin concentrations (26): HOMA-IR = [Insulin (µU/mL) * Glucose (mmol/L)]/22.5.

Isolation of LDL and LDL subfractions

LDL were isolated from the serum of each participant by ultracentrifugation under standard conditions (27). The LDL fractions were grouped into four major LDL subclasses, based on density, designated LDL I (d < 1.028), LDL II (1.028–1.034), LDL III (1.034–1.044) and LDL IV (1.044–1.060) (6).

Chemical composition of LDL subfractions

Fractions were analyzed for concentrations of total cholesterol (TC) (Siemens Medical Solutions Diagnostics, Tarrytown, NY), triglyceride (Sigma-Aldrich, St. Louis, MO), free cholesterol (FC) (Wako Chemicals USA, Inc., Richmond, VA) and phospholipids (PL) (Wako Chemicals USA, Inc., Richmond, VA) using commercially available enzymatic kits. Cholesterol ester (CE) concentrations were calculated as $[(TC – FC) \times 1.68]$. Protein concentrations were determined by a modification of the method of Lowry et al. using BSA as the standard (28). The lipoproteins were read on an ELx 808 Ultra Microplate Reader (Bio-Tek Instruments, Inc., Winooski, VT). Assays were carried out in duplicate with standards, controls, and unknowns on the same 96-well plate, and with coefficients of variation within 10%. On average over 90% of the plasma apolipoproteins (B, CIII and E) in LDL subclasses were recovered from density gradient ultracentrifugation.

Statistical analysis

JMP statistical software (version 7.0; SAS Institute, Cary, NC) was used for statistical analysis. Data are presented as means ± SD. Shapiro-Wilk test was used to test the skewness of the distributions. Non-normally distributed variables were log-transformed before analysis. Paired t-tests were used to evaluate differences in lipids, lipoproteins and apolipoproteins between the diets and across LDL subclasses. Spearman's correlation coefficients were used to evaluate the relationship between diet-induced changes in lipids, lipoproteins and apolipoproteins. P values < 0.05 were considered statistically significant.

Results

Participant characteristics at baseline

The mean (± SD) age of the men was 44.5 ± 14.4 y (range: 24 to 67 years), weight was 78.6 ± 10.8 kg, BMI was 25.0 ± 2.4 kg/m^2 and body fat was 20.9 ± 6.3%. The participants had normal fasting blood glucose (89.3 ± 8.0 mg/dL), plasma insulin concentrations (5.1 ± 2.4 µU/mL) and HOMA-IR (1.1 ± 0.6).

Plasma lipid and lipoprotein response to dietary intervention

In the context of moderate carbohydrate intake, consumption of LSF vs. HSF intake resulted in an overall more favorable lipid profile with lower plasma TC (P = 0.007), LDL cholesterol (P = 0.007), TC/HDL cholesterol (P = 0.0004), apoB (P = 0.06) and apoCIII concentrations (P = 0.03) (Table 2).

Compositional characteristics of LDL and LDL subclasses at baseline

Analyses of lipid and apolipoprotein content were performed on total LDL and LDL subfractions (Table 3). The baseline compositional trends of LDL subclasses for percent free cholesterol, cholesterol esters, phospholipids and protein across LDL subclasses were consistent with previous findings (6, 27, 29). Percent triglyceride at baseline was greater in LDL I versus LDL II–IV.

LDL I and LDL IV particles were significantly lower in number (as measured by apoB) and total mass concentration compared to LDL II and III, with LDL III being the most abundant subfraction (Table 3). ApoCIII and apoE levels were highest in LDL IV (Table 3). We also calculated the relative enrichment of apoCIII and apoE per apoB molecule of total LDL and across LDL subclasses (Table 4). ApoCIII/apoB and apoE/apoB ratios were significantly higher in LDL I and IV than in LDL II and III with no significant difference between LDL I and IV.

Effects of dietary saturated fat on compositional characteristics of LDL and LDL subclasses

In the setting of moderate carbohydrate intake, HSF vs. LSF intake significantly increased LDL total mass concentration by 15.2% as well as LDL apoCIII content by 33.5% (Table 5). The change in LDL mass was attributable to significant increases in the mass concentrations of LDL subclasses I, II and III by 19.0, 20.1 and 16.5%, respectively, but was significantly correlated only with changes in LDL III (r = 0.65, P = 0.01) (data not shown). The relative enrichment of apoCIII per apoB in total LDL tended to increase with HSF vs. LSF intake (P = 0.07) (data not shown). Saturated fat-induced increases in plasma and LDL apoCIII were positively and significantly correlated with changes in LDL IV apoCIII (Table 6 and Figure 1), but not LDL I–III apoCIII. No significant changes in apoE content were observed in total LDL or LDL subclasses with HSF vs. LSF intake (Table 5).

Discussion

Our results show that in the context of a reduced carbohydrate, high beef protein diet, the increase in plasma and LDL cholesterol with high vs. low saturated fat is associated with concurrent increases in the concentration of plasma and LDL apoCIII and the mass of large, medium and small LDL particles. The change in total LDL apoCIII was correlated with changes in apoCIII in the LDL IV fraction ($\rho = 0.66$). Consistent with previous reports, the concentration of apoCIII was significantly higher in LDL IV compared to other LDL subfractions (Table 3) (30, 31). Increased LDL apoCIII has been shown to be an important marker of CVD risk that is independent of standard lipid risk factors and in fact much of the risk ordinarily attributed to LDL appears to be due to LDL particles that contain apoCIII

(18, 32). The increased apoCIII content of small dense LDL and greater affinity of these particles for arterial proteoglycans (31) may increase interaction with the arterial wall and facilitate oxidative modification. Furthermore, clinical observations suggest that apoCIII may play a direct role in the development of atherosclerosis by activating inflammatory responses through monocyte-endothelial interactions (33).

Consistent with the results of the larger study, we found that saturated fat increased the mass of LDL I–III with no change in LDL IV (Table 5). This contrasts with results from an earlier study, where high (15%) vs. low (9%) saturated fat intake in the context of a diet high in protein from mixed sources increased LDL cholesterol primarily through elevations in mass of larger LDL without changes in levels of small LDL particles (10). While the basis for the differing findings is unclear, one explanation may be related to differences in dietary protein source between the two studies.

To our knowledge, this study is the first to show significant increases in plasma and LDL apoCIII with saturated vs. monounsaturated fat feeding, with all other macronutrients held constant. Others (34–37) have reported diet-induced increases in plasma apoCIII and concurrent elevations in plasma triglycerides, mainly as a result of increased dietary carbohydrate intake. In contrast to the current study, the saturated fat-mediated changes in plasma and LDL apoCIII were independent of changes in triglycerides. Studies in cynomolgus monkeys have shown significantly higher hepatic apoCIII mRNA levels with saturated vs. monounsaturated fat intake (38). Consistent with our observations, the increase in hepatic apoCIII in the monkey studies was not paralleled by an increase in plasma triglycerides. Thus, the saturated fat induced rise in plasma and LDL apoCIII in our study may be due to the increase in production of apoCIII at the transcriptional level.

ApoCIII enriched apoB-containing lipoproteins may be particularly atherogenic by virtue of their decreased affinity for the LDL receptor (12) and increased binding to extracellular vascular proteoglycans (39). LDL with apoCIII strongly predicts coronary events in diabetic patients independent of other lipids (18). Notably, in plasma isolated from patients with metabolic syndrome or type 2 diabetes mellitus, the content of apoCIII in small dense LDL has been shown to be significantly correlated with an enhanced affinity of these particles for arterial proteoglycans (31). A recent report in 141 healthy middle-aged men showed a significant positive association between plasma apoCIII concentration and levels of the smallest of LDL subclasses (LDL IV) independent of plasma triglycerides and concluded that enrichment of apoCIII in small dense LDL may contribute to the metabolic and pathologic properties of these particles (40). A recent kinetic study (41) showed accelerated hepatic lipase-mediated conversion of light LDL to dense LDL when precursor particles were enriched with apoCIII. It is possible that *in vivo*, apoCIII positively modulates hepatic lipase to increase dense LDL particle concentrations. Based on the results from the total set of individuals participating in the current dietary intervention (n = 40) (11), higher saturated fat intake combined with high beef intake tended to increase the activity of hepatic lipase activity (P = 0.08) supporting the role of hepatic lipase in promoting the formation of small dense LDL.

Limitations of the current study include a small sample size, providing limited power for multivariable statistical adjustments, and the potential for type I error as a result of multiple testing. Nonetheless, the present findings suggest that increased levels of small, apoCIII-containing LDL may contribute to the adverse effects of a diet high in beef and saturated fat on CVD risk.

Acknowledgments

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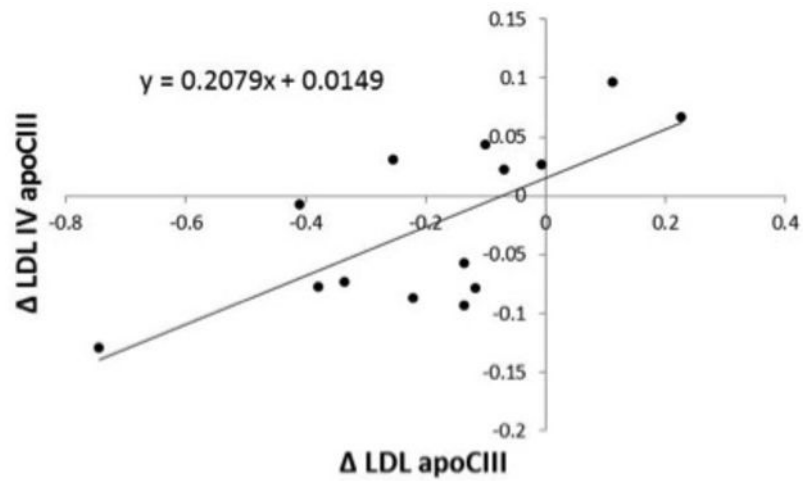
FIGURE 1.
Spearman's correlation between saturated fat-induced changes in total LDL apoCIII (mg/dL) and LDL IV apoCIII (mg/dL) in men. , low saturated fat – high saturated fat. N = 14, $\rho = 0.66$, P = 0.01.

TABLE 1

Macronutrient composition of the diets

	Baseline	HSF	LSF
	% of energy		
Carbohydrate	50	31	31
Protein	13	31	31
Beef protein	0	10	11
Total Fat	38	38	38
Saturated fat	15	15	8
Monounsaturated fat	15	15	21
Polyunsaturated fat	6	5	6
Cholesterol, mg/d	468	463	467

Diets were formulated to provide similar amounts of *trans* fat (1.5% daily E), linoleic acid (0.5% E), linolenic acid (0.5% E), cholesterol (465 mg/3000 kcal), and fiber (25 g/2000 kcal plus 2.5 g/500 kcal above this level), and to have a comparable ratio of sugars to starches (1:1).

E, energy; HSF, high saturated fat; LSF, low saturated fat.

TABLE 2

Plasma measurements following consumption of baseline (B), high saturated fat (HSF) and low saturated fat (LSF) diets

| | 50% CHO, 13% protein | 31% CHO, 31% protein | | |
	B	HSF	LSF	SF
Triglyceride (mg/dL)	97.7 ± 47.9	85.4 ± 33.4	82.5 ± 33.8	-2.7 ± 14.9
Total cholesterol (TC) (mg/dL)	177.6 ± 32.0	170.4 ± 30.0	156.7 ± 23.3	-13.6 ± 15.8**
LDL cholesterol (mg/dL)	108.9 ± 29.1	106.2 ± 31.2	93.3 ± 22.0	-12.9 ± 15.3**
HDL cholesterol (mg/dL)	49.2 ± 10.9	47.2 ± 12.8	46.9 ± 13.4	-0.3 ± 2.7
TC/HDL cholesterol (mg/dL)	3.8 ± 1.2	3.9 ± 1.4	3.6 ± 1.1	-0.3 ± 0.3**
ApoA-I (mg/dL)	109.1 ± 13.1	103.1 ± 10.6	103.2 ± 14.6	0.1 ± 8.6
ApoB (mg/dL)	70.5 ± 17.7	69.5 ± 15.3	64.1 ± 15.2	-5.0 ± 9.4
ApoCIII (mg/dL)	8.8 ± 2.0	8.7 ± 2.0	8.0 ± 2.3	-0.8 ± 1.2*
HDL apoCIII (mg/dL)	5.9 ± 1.0	5.8 ± 1.2[#]	5.5 ± 1.2[#]	-0.1 ± 0.9
Non-HDL apoCIII (mg/dL)	2.9 ± 1.4	2.9 ± 1.2[#]	2.3 ± 1.4[#]	-0.6 ± 1.2
Ratio of HDL to non-HDL apoCIII	2.5 ± 1.2	2.3 ± 1.2[#]	3.1 ± 1.9[#]	0.9 ± 2.3

Values are means $\pm$ SD. n=14.

[#] n=13.

CHO, carbohydrate;   SF, LSF − HSF.

* $P < 0.05$,

** $P < 0.01$, LSF vs. HSF (paired t-test).

TABLE 3

Composition and apolipoprotein content of LDL subclasses at baseline

	total mass mg/dL	TG	FC	CE	PL	protein	apoB	apoCIII	apoE
				% composition				mg/dL plasma	
LDL total	251.6 ± 67.5	3.7 ± 0.9	8.0 ± 1.2	41.0 ± 2.4	21.1 ± 1.2	26.2 ± 1.4	54.47 ± 21.57	0.59 ± 0.31	0.47 ± 0.17
LDL I	26.5 ± 9.1[a]	7.7 ± 2.2[a]	9.6 ± 1.3[a]	39.5 ± 2.9[a]	21.8 ± 0.6[a]	21.4 ± 1.8[a]	4.51 ± 1.62[a]	0.09 ± 0.09[ab]	0.06 ± 0.05[a]
LDL II	46.6 ± 14.9[b]	3.7 ± 1.4[b]	8.8 ± 1.3[a]	43.8 ± 3.2[b]	21.6 ± 1.1[a]	22.1 ± 1.6[a]	9.30 ± 2.99[b]	0.06 ± 0.08[a]	0.02 ± 0.02[b]
LDL III	117.6 ± 55.1[c]	2.8 ± 0.8[c]	8.8 ± 2.5[ab]	40.4 ± 11.4[abc]	21.7 ± 4.5[ab]	26.2 ± 5.3[b]	27.38 ± 12.45[c]	0.09 ± 0.07[b]	0.07 ± 0.04[a]
LDL IV	28.5 ± 18.5[ab]	3.3 ± 1.7[bc]	7.7 ± 0.4[b]	33.8 ± 3.1[c]	20.0 ± 1.2[b]	35.2 ± 2.6[c]	6.29 ± 4.23[ab]	0.18 ± 0.09[c]	0.13 ± 0.07[c]

Values are means ± SD. n=10. Means in a column without a common letter differ, $P < 0.05$. Paired t-test across LDL subclasses.

FC, free cholesterol; CE, cholesterol ester; PL, phospholipid.

TABLE 4

Apolipoprotein ratios of LDL and LDL subfractions at baseline

	apoCIII/apoB	apoE/apoB
LDL total	0.012 ± 0.005	0.009 ± 0.004
LDL I	0.019 ± 0.014[a]	0.013 ± 0.006[a]
LDL II	0.006 ± 0.007[b]	0.003 ± 0.002[b]
LDL III	0.003 ± 0.001[b]	0.003 ± 0.002[b]
LDL IV	0.034 ± 0.018[a]	0.026 ± 0.013[a]

Values are means ± SD. n=10. Means in a column without a common letter differ, P < 0.005. Paired t-test across LDL subclasses.

TABLE 5

Effects of dietary saturated fat on composition and apolipoprotein content of LDL and LDL subclasses

	total mass mg/dL	TG	FC	CE	PL	protein	apoB	apoCIII	apoE
				% composition				mg/dL plasma	
LDL total									
HSF	259.6 ± 60.2	3.9 ± 1.0	7.6 ± 1.1	41.5 ± 5.2	19.7 ± 2.0	27.3 ± 5.7	51.13 ± 17.55	0.73 ± 0.36	0.40 ± 0.18
LSF	225.3 ± 48.8**	4.1 ± 0.9	8.2 ± 0.9	39.9 ± 2.3	21.0 ± 1.2*	26.8 ± 2.2	47.08 ± 14.84	0.55 ± 0.20*	0.39 ± 0.24
LDL I									
HSF	35.2 ± 12.7	7.2 ± 2.1	9.1 ± 1.2	39.2 ± 4.3	20.2 ± 1.8	24.3 ± 5.9	5.19 ± 1.44	0.13 ± 0.12	0.06 ± 0.04
LSF	29.6 ± 9.8*	7.8 ± 1.5	9.6 ± 1.4	39.0 ± 2.5	21.8 ± 1.2*	21.8 ± 1.8	4.75 ± 1.73	0.08 ± 0.07	0.06 ± 0.04
LDL II									
HSF	47.9 ± 16.2	4.1 ± 1.6	9.3 ± 0.9	44.0 ± 5.0	20.8 ± 1.9	21.9 ± 3.5	8.62 ± 3.28	0.04 ± 0.02	0.02 ± 0.01
LSF	39.8 ± 13.2**	4.3 ± 1.4	9.5 ± 1.1	41.7 ± 2.9	22.2 ± 1.5	22.3 ± 2.7	6.90 ± 3.24	0.03 ± 0.03	0.02 ± 0.02
LDL III									
HSF	121.9 ± 44.9	2.9 ± 0.9	8.6 ± 0.9	41.3 ± 3.7	20.1 ± 1.6	27.0 ± 4.9	25.56 ± 10.81	0.12 ± 0.10	0.06 ± 0.02
LSF	104.6 ± 34.6**	3.0 ± 0.8	9.1 ± 0.7	41.1 ± 2.0	21.5 ± 0.9*	25.4 ± 1.9	22.72 ± 10.00	0.08 ± 0.05	0.06 ± 0.03
LDL IV									
HSF	27.6 ± 14.2	2.7 ± 1.4	7.1 ± 1.2	35.8 ± 5.1	19.5 ± 2.2	34.8 ± 7.1	5.60 ± 2.61	0.18 ± 0.09	0.10 ± 0.03
LSF	25.2 ± 11.8	2.9 ± 1.1	7.7 ± 0.5	34.5 ± 3.1	20.9 ± 2.1	34.0 ± 3.2	5.40 ± 3.54	0.16 ± 0.07	0.10 ± 0.05

Values are means ± SD. n=14.

FC, free cholesterol; CE, cholesterol ester; PL, phospholipid; HSF, high saturated fat; LSF, low saturated fat.

Significantly different from HSF:

*
 P < 0.05,

**
 P < 0.01 (paired t-test).

TABLE 6

Spearman's correlations between saturated fat-induced changes in plasma & LDL apoCIII and changes in apoCIII in LDL subclasses

apoCIII in LDL subclasses	plasma apoCIII	LDL apoCIII
LDL I	0.34	0.17
LDL II	0.17	0.10
LDL III	0.07	0.42
LDL IV	0.54[*]	0.66[*]

low saturated fat – high saturated fat.

[*] P < 0.05.