Low density lipoprotein metabolism in hypertriglyceridemic and normolipidemic patients with coronary heart disease

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Abstract The turnover rates of low density lipoprotein-apolipoprotein B (LDL-apoB) were determined in 32 men with coronary heart disease (CHD) and 11 control men with normal plasma lipids. Thirty patients with CHD had normal levels of LDL-cholesterol (LDL-C); of these patients, 9 had hypertriglyceridemia and 21 had normal plasma lipids. Mean concentrations of total cholesterol and LDL-C were similar among the control subjects and CHD patients, although the latter had significantly lower HDL-C. In control subjects, transport rates and fractional catabolic rates (FCR) of LDL-B were 10.6 ± 0.5 (SEM) mg/kg-day and 0.31 ± 0.01 pools/day, respectively. In 10 hypertriglyceridemic patients with CHD, transport rates were 21.7 ± 1.7 mg/kg-day, and FCRs averaged 0.56 ± 0.06 pools/day; both were significantly higher than normal ($P < 0.05$). Six normolipidemic patients also had abnormally high transport rates of LDL-apoB (19.4 ± 2.8 mg/kg-day) and FCRs (0.51 ± 0.03 pools/day); again both were higher than normal. The remaining 16 normolipidemic patients with CHD had normal transport rates (9.9 ± 0.6 mg/kg-day) and FCRs (0.28 ± 0.01 pools/day). Thus, hypertriglyceridemic patients with CHD and a portion of normolipidemic patients with CHD were characterized by increases in both transport and fractional catabolic rate of LDL-apoB; these abnormalities in LDL metabolism may have contributed to their coronary heart disease. However, the majority of normolipidemic patients with CHD did not show a distinct defect in their LDL metabolism.—**Vega, G. L., W. F. Beltz, and S. M. Grundy.** Low density lipoprotein metabolism in hypertriglyceridemic and normolipidemic patients with coronary heart disease. *J. Lipid Res.* 1985. **26:** 115–126.

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Elevations of plasma cholesterol and low density lipoproteins (LDL) are known risk factors for coronary heart disease (CHD) (1–4). As LDL levels increase, more LDL presumably filters into subintimal regions (5), and the cholesterol carried in LDL accumulates within the arterial wall. The contribution of LDL to atherosclerosis, however, may not be determined solely by plasma concentrations. Other abnormalities in metabolism of LDL also may affect the atherogenicity of this lipoprotein. For example, it has been proposed that normal LDL must be modified before it can induce the development of foam cells (6). Other metabolism abnormalities may also render LDL more atherogenic.

There are several reports of such abnormalities (7–17). Defects both in composition and metabolism of LDL have been noted in patients with CHD who have relatively normal levels of LDL. The present study, therefore, was undertaken to examine further whether alterations in metabolism of LDL can be identified in patients with CHD who do not have definite hypercholesterolemia. Two types of patients were chosen for investigation: *a*) those with hypertriglyceridemia and *b*) those with normolipidemia. The results demonstrate that most hypertriglyceridemic patients with CHD have defects in metabolism of LDL as do a portion of normolipidemic patients.

METHODS

Patients

Forty-three men were studied; most of the studies were carried out on the metabolic ward of the Veterans Administration Medical Center and the General Clinical Research Center of the Parkland Memorial Hospital,

Abbreviations: CHD, coronary heart disease; LDL-apoB, low density lipoprotein-apolipoprotein B; LDL, low density lipoprotein; VLDL, very low density lipoprotein; HDL, high density lipoprotein; FCR, fractional catabolic rate; IDL, intermediate density lipoprotein; C, cholesterol; TG, triglyceride; HTG, hypertriglyceridemia.

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Dallas, Texas. Thirty-two patients were selected for the study on the basis of having coronary heart disease (CHD) and normal LDL-cholesterol levels. Ten patients had persistent hypertriglyceridemia (HTG). Six patients had occasional HTG, but they were consistently normolipidemic during the study. The remaining 16 patients were persistently normolipidemic. HTG was defined as a plasma triglyceride (TG) level greater than the 95th percentile for the population of the same age and sex as reported in the Lipid Research Clinic (LRC) Population Prevalence Survey (18, 19). CHD was defined as documented myocardial infarction or coronary artery bypass graph. Neither myocardial infarction nor coronary surgery had occurred during the 6 months before study. None of the patients had marked obesity. For all patients, percent ideal body weight (% IBW) varied between 97 and 129% (mean % IBW was 115 ± 3%) (20). None of the patients had received hypolipidemic drugs for 6 months prior to the study, and none of the patients had diabetes mellitus, gastrointestinal or liver diseases, or other endocrine disorders. Family screening for hyperlipidemia was carried out on as many patients as possible as described by Beil et al. (21).

Eleven normolipidemic subjects were studied as controls (**Table 1**). They were all men of ages 38 to 68 years (mean = 55 ± 3 yr). None had a history of CHD or angina pectoris. Their mean weight was 76 ± 3 kg (ideal weight 106 ± 14%). All of these subjects were healthy, and none had a history of significant illness. In four of these controls (Nos. 5, 6, 7, 8), turnover studies were carried out at the San Diego Veterans Administration Medical Center, under essentially identical conditions (22).

The clinical characteristics of each patient with HTG are shown in **Table 2**. Mean age for this group was 50 ± 2 (SE) yrs at time of study. Six of ten gave a history of CHD in first degree relatives. Hyperlipidemia, either hypercholesterolemia or hypertriglyceridemia, was documented in other family members for seven patients, while in two patients (Nos. 17 and 21), levels could not be obtained on other family members.

TABLE 1. Clinical data: normolipidemic subjects without coronary heart disease

Patient	Age	Weight	% Ideal Weight
	yr	*kg*	*%*
1	61	84	112
2	60	75	112
3	45	58	84
4	58	98	118
5	62	75	114
6	54	75	101
7	68	56	81
8	56	78	110
9	42	75	105
10	38	87	117
11	58	76	110
Mean ± SEM	55 ± 3	76 ± 3	106 ± 14

Clinical characteristics of the remaining 22 CHD patients with normolipidemia are given in **Table 3**. The mean age of this group was 61 ± 1 yr. A positive history for CHD in the family was reported in 11 patients. It was possible to obtain plasma lipid data on first degree relatives in only 15 patients, and of these only 9 families were found to have at least one member with hyperlipidemia. Patients number 22 through 27 had a history of intermittent HTG, but during the time of study, their plasma TG levels were not elevated. Patient 22 was the brother of patient 18.

None of the patients in the above groups had unstable angina pectoris, congestive heart failure, or disorders of the gastrointestinal or endocrine systems. Ten of the patients with HTG and 4 of the patients without HTG were taking beta adrenergic blocking agents at the time of study. All patients had a history of smoking, although only 16 were smokers at the time of study. Each patient gave informed consent to participate in the study.

Experimental design

Plasma lipid screening was carried out for each patient while they were on their home diet. Upon entrance into the current study the patients started on a metabolic diet. The majority of the patients underwent their whole study as inpatients on the metabolic ward of the VA Medical Center at Dallas. In the remainder, the study was initiated on the metabolic ward and patients remained hospitalized for 3 to 7 days; thereafter, the study was completed with subjects as outpatients. They followed the same diet whether they were inpatients or outpatients. All patients took a repetitive solid food diet consisting of 40% of calories as fat (18% saturated, 17% monounsaturated, and 5% polyunsaturated), 45% as carbohydrate, and 15% as protein. Daily intake of cholesterol was approximately 300 mg. This diet was designed to resemble the "typical American" diet. The inpatients were fed from the metabolic kitchen. A daily menu was given to outpatients, and they were counselled on food preparation and serving sizes. A daily diet diary was kept by these patients. Inpatients' weights were monitored daily, and serving sizes were adjusted to maintain constant weight. Outpatients were weighed frequently and advised if changes in caloric intake were needed. During the turnover study the patients' weights were relatively constant; their coefficient of variation in weight (SD of weights/mean weight) varied from 0.0029 to 0.0087 (mean ± SD = 0.0055 ± 0.0016). None of the patients showed a distinct trend toward weight gain or weight loss throughout the study.

TABLE 2. Clinical data: hypertriglyceridemic patients with coronary heart disease

Patient	Age	Weight	% Ideal Weight	Age of Onset of CHD	Family History of CHD	Hyperlipidemia in Family Members	
						Chol[a]	TG[a]
	yr	*kg*	*%*	*yr*			
12	36	84	113	34	0	3/3	0/3
13	41	86	118	36	+	4/8	2/8
14	47	84	106	46	+	1/7	4/7
15	56	77	111	51	+	1/4	0/4
16	51	88	114	40	+	0/3	0/3
17	57	80	120	47	n.a.	n.a.	n.a.
18	54	80	122	45	+	3/9	1/9
19	56	75	113	54	+	0/7	1/7
20	51	76	115	30	0	2/10	2/10
21	32	99	125	30	n.a.	n.a.	n.a.
Mean ± SEM	50 ± 2	81 ± 2	115 ± 2	43 ± 3			

[a]The numerator is the number of relatives of the patient who had either elevated plasma cholesterol (Chol) or triglycerides (TG). The denominator is the total of first degree relatives tested.

Abbreviations: Chol, cholesterol; TG, triglyceride; +, positive family history of premature atherosclerosis; 0, negative family history; n.a., history not available.

A measurement for turnover rates of LDL was initiated after 2 weeks on the metabolic diet. Plasmapheresis was carried out to obtain plasma for isolation of LDL by preparative ultracentrifugation; LDL was then radioiodinated and reinjected into the patient 5 days after plasmapheresis. Disappearance of radioactivity from the plasma was monitored for 20 days, and levels of plasma lipids, lipoprotein cholesterol, and LDL-apolipoprotein B (LDL-apoB) were measured repeatedly throughout the study. Patients received 0.5 to 0.9 g of KI orally in divided doses each day to suppress uptake of radioiodine by the thyroid.

Plasma total and lipoprotein lipids

Plasma lipids and lipoprotein-cholesterol were estimated every 3 days throughout the turnover study. Cholesterol was measured by the procedure of Roeschlau, Bernt, and Gruber (23). The cholesterol standard supplied by Behringer-Mannheim was calibrated using pooled reference plasma as standard. The cholesterol in

TABLE 3. Clinical data in coronary heart disease patients without hypertriglyceridemia

Patient	Age	Weight	% Ideal Weight	Age of Onset of CHD	Family History of CHD	Hyperlipidemia in Family Members	
						Chol[a]	TG[a]
	yr	*kg*	*%*	*yr*			
22[b]	57	70	108	48	+	3/9	1/9
23	59	100	126	34	+	0/3	0/3
24	70	82	116	40	0	0/8	1/8
25	55	90	120	54	n.a.	n.a.	n.a.
26	59	72	108	55	+	0/7	0/7
27	74	74	108	60	n.a.	n.a.	n.a.
28	56	83	115	45	0	0/6	1/6
29	64	86	117	62	+	0/4	1/4
30	61	82	116	41	n.a.	n.a.	n.a.
31	57	89	115	49	n.a.	n.a.	n.a.
32	59	80	127	55	n.a.	n.a.	n.a.
33	48	76	111	42	0	0/8	1/8
34	58	73	97	44	+	0/1	0/1
35	59	86	100	51	+	0/5	0/5
36	65	97	129	54	+	1/2	0/2
37	66	76	114	64	+	n.a.	n.a.
38	60	80	101	52	+	0/5	0/5
39	67	67	104	66	+	n.a.	n.a.
40	66	70	108	60	0	1/8	0/8
41	59	67	97	58	0	1/4	0/4
42	66	75	124	63	0	1/5	1/5
43	65	90	113	50	+	0/3	0/3
Mean ± SEM	61 ± 1	80 ± 2	112 ± 2				

[a]The numerator is the number of relatives of the patient who had either elevated plasma cholesterol (Chol) or triglycerides (TG). The denominator is the total of first degree relatives tested.

[b]Patient 22 is the brother of patient 18.

the latter was determined by gas–liquid chromatography. Triglycerides in whole plasma also were measured enzymatically (24).

Cholesterol (C) in VLDL, LDL, and HDL, and VLDL-TG were determined as previously described (17, 25). VLDL was isolated by ultracentrifugation at d < 1.006 g/ml. Cholesterol was determined in the infranatant [LDL-C + IDL-C + HDL-C]. LDL + IDL was precipitated by phosphotungstic acid and magnesium (26), and HDL-C was determined enzymatically in the supernatant. It has been reported that HDL-C levels determined with the phosphotungstic acid method average 3–5 mg/dl lower than those obtained with heparin-manganese precipitation (27), and this was confirmed in our laboratory. LDL-C + IDL-C was calculated by difference.

IDL-C and LDL-C were estimated by an equation developed from the data of Nichols (28); this worker presented values for IDL (total mass of S_f 12–20 lipoproteins) relative to LDL (total mass of S_f 0–12) in a large number of patients with varying levels of VLDL (total mass of S_f 20–400). Assuming that VLDL-TG = 0.6 VLDL (29), the IDL/LDL ratio (R_m) can be estimated from the data of Nichols (28) by the following equation: $R_m = (0.283 \times 10^{-3})$ (VLDL-TG) + 0.1144 ($r = 0.953$, $P < 0.004$). The chemical compositions of IDL and LDL were determined recently for 12 normolipidemic and 12 hypertriglyceridemic patients by Mattson and Grundy (15). For normotriglyceridemic subjects, IDL-C = 0.167 IDL and LDL-C = 0.326 LDL; for patients with HTG, IDL-C = 0.191 IDL and LDL-C = 0.311 LDL. Thus, for normotriglyceridemic patients, the IDL-C/LDL-C ratio (R_c) = (0.167/0.326) R_m = 0.5123 R_m, and for patients with HTG, R_c = 0.6141 R_m. Since (IDL-C + LDL-C) was measured, the following equations can be derived to estimate LDL-C:

for normotriglyceridemic patients,

$$\text{LDL-C} = (\text{IDL-C} + \text{LDL-C})/(0.5123\ R_m + 1) \quad \textit{Eq. 1}$$

and for hypertriglyceridemic patients,

$$\text{LDL-C} = (\text{IDL-C} + \text{LDL-C})/(0.6141\ R_m + 1). \quad \textit{Eq. 2}$$

Isolation of LDL for compositional analysis

LDL was isolated for compositional analysis every 3 days during the study. Four ml of plasma was used for isolation of LDL for compositional analysis. To this plasma was added 2 ml of a salt solution of 0.499 M NaBr and 0.195 M NaCl according to the method of Lindgren and Jensen (30). This mixture was made in 6-ml Ultra-Clear Beckman ultracentrifuge tubes. The resulting solution had a density of 1.019 g/ml. The tubes were spun in a fixed-angle Beckman 50.3 Ti rotor, at 40,000 rpm for 20 hr at 15°C (31). The top 2 ml was aspirated and designated VLDL. The infranatant was brought to a density of 1.070 g/ml in the same tube by addition of 2 ml of 2.099 M NaBr and 0.195 M NaCl. LDL was then isolated by ultracentrifugation as described above for VLDL; the top 2 ml was aspirated again.

Lipoproteins were dialyzed against 0.15 M NaCl containing 0.15 mM chloramphenicol, 1.5 mM sodium azide, and 0.27 mM EDTA at pH 7.4 (31). Extensive dialysis was carried out shortly after isolation. Total protein in LDL was measured by a modification of the Lowry procedure (17). ApoB concentration in the lipoproteins was calculated as the difference between total protein in the fractions and soluble proteins after precipitation of apoB by isopropyl alcohol (17, 32).

Bovine serum albumin (BSA), obtained from the National Bureau of Standards, was used as a protein standard. The stability of the standard was monitored routinely by measuring its absorbance at 280 nm and using $E^{1\%}_{cm} = 6.6$ (33). The protein mass of LDL was measured by the procedure of Markwell et al. (34) as described previously (17). ApoB was precipitated with isopropyl alcohol (17, 32). Since the chromogenicity of the small quantity of proteins soluble in isopropanol was not determined, a ratio of 1.0 compared to BSA was used for this fraction. Cholesterol concentration in isolated LDL was determined enzymatically as described above. Absolute concentration of LDL-apoB was estimated by taking the ratio of apoB-cholesterol in isolated LDL and multiplying by LDL-C estimated as described in the preceding section.

LDL-apoB kinetics

Radioiodination of autologous LDL-apoB. LDL (1.02–1.063 g/ml) was isolated from the plasma of each patient and the apoprotein was labeled with ^{125}I by the iodine-monochloride method of McFarlane (35) as modified by Langer, Strober, and Levy (36) and Bilheimer, Eisenberg, and Levy (37). Unbound ^{125}I was removed by extensive dialysis and the labeled LDL was diluted with 5% human serum albumin and sterilized by passage through a 0.22-μm pyrogen-free Millipore filter. The mean percent radioactivity in the lipid extract was 7.3 ± 3.9%; the average trichloroacetic acid-precipitable counts were 98 ± 0.12%. Usually 36–50 μCi of labeled LDL, and 5–10 mg of unlabeled LDL were injected intravenously. Blood samples were collected at 10 and 20 min, and 1, 4, 8, 12, and 24 hr. Thereafter samples were collected every 12 hr for the subsequent 3 days and, finally, every 24 hr from the 4th through the 20th day of the turnover study. Determination of radioactivity was made on samples obtained at each time interval, and plasma lipids, lipoprotein cholesterol, and LDL-apoB were measured twice weekly. All radioactivity was measured in a gamma counter.

The fractional catabolic rate (FCR) and transport

rates were determined as described by Matthews (38). Briefly, the die-away curve of the plasma radioactivity was bi-exponential. FCR of LDL-apoB was calculated using a two-pool mammillary model; transport rate of LDL-apoB was calculated by multiplying the pool size of LDL-apoB by the FCR; plasma volume was determined by isotope dilution at 10 min after injection of LDL. LDL-C transport was estimated by dividing the LDL-apoB transport by the LDL-apoB/C ratio. Data were normalized to kg of body weight.

Statistical analysis. Linear statistical procedures available as Interactive Statistical Programs (ISP) were used for data analysis. The analyses were carried out at the Medical Computing Resources Center, University of Texas Health Science Center at Dallas. One-way analysis of variance (ANOVA) was used for multiple comparison of all parameters for all groups.

RESULTS

The plasma lipids, lipoprotein-cholesterol, and kinetic parameters for LDL are summarized for all the groups in Tables 4 through 9. Results for normal subjects will be described first and data for other groups will be compared to these.

Normal subjects

In this group (**Table 4 and Table 5**), plasma cholesterol and TG ranged from 162 to 252 mg/dl and 66 to 244 mg/dl, respectively [mean cholesterol = 202 ± 9 mg/dl (SEM); mean TG = 128 ± 15 mg/dl]. HDL-cholesterol averaged 42 ± 5 mg/dl. The levels of LDL-C varied between 97 and 146 mg/dl (mean = 126 ± 4 mg/dl), and LDL-apoB between 67 and 95 mg/dl (mean = 76 ± 3 mg/dl), respectively. The LDL-apoB/C ratio averaged 0.61 ± 0.02; the FCR ranged from 0.26 to 0.36 pools/day (mean = 0.31 ± 0.01 pools/day) and the transport rate ranged from 7.9 to 12.9 mg/kg-day (mean = 10.6 ± 0.5 mg/kg-day).

Hypertriglyceridemic patients with CHD

The mean plasma cholesterol in this group was 245 ± 11 mg/dl; the TG levels averaged 589 ± 96 mg/dl (**Table 6**). The latter were significantly higher than in normal subjects. HDL-C levels averaged 26 ± 1 mg/dl, a mean significantly lower than that of the normal group. LDL-C levels in hypertriglyceridemic patients ranged from 75 to 191 mg/dl (mean = 117 ± 16 mg/dl); LDL-apoB concentrations varied between 54 and 209 mg/dl (mean = 99 ± 14 mg/dl) (**Table 7**). The concentrations of LDL-apoB but not LDL-C were significantly higher than those of the normal subjects. However, the FCR of LDL-apoB (mean = 0.56 ± 0.06 pools/day), the transport rates of LDL-apoB (mean = 21.0 ± 1.7 mg/kg-day) and LDL-apoB/C ratios (mean = 0.89 ± 0.06) were significantly higher than in normal subjects ($P < 0.05$). The LDL-apoB levels correlated negatively with FCR ($r = -0.58$; $P < 0.015$); in other words, when FCR was relatively low, LDL-apoB levels tended to be high. Linear regression analysis also demonstrated a positive correlation between LDL-apoB/C ratios and FCR ($r = 0.63$; $P < 0.07$).

Normolipidemic patients with CHD

The 23 patients of this group were of two types: *a*) those with a high FCR of LDL-apoB (FCR > 0.40 pools/day) and *b*) those with a relatively low FCR (FCR < 0.34 pools/day). These are designated subgroups A and B, respectively. The two subgroups appeared to differ on other grounds as discussed below; therefore, they are considered separately.

Subgroup A. In this subgroup of six patients, levels of plasma cholesterol and TG during the study averaged

TABLE 4. Plasma lipids and lipoprotein cholesterol: normal subjects

Patient	Plasma		Lipoprotein cholesterol			
	Chol	TG	VLDL	IDL	LDL	HDL
	mg/dl ± SD		*mg/dl ± SD*			
1	190 ± 12 (5)[a]	157 ± 38	27 ± 4	9 ± 2	123 ± 12	31 ± 2
2	179 ± 20 (7)	82 ± 19	12 ± 3	8 ± 2	125 ± 13	28 ± 3
3	162 ± 10 (3)	66 ± 10	9 ± 2	7 ± 1	114 ± 8	32 ± 4
4	186 ± 6 (7)	127 ± 28	20 ± 3	9 ± 1	127 ± 4	30 ± 2
5	201 ± 3 (7)	109 ± 6	15 ± 1	8 ± 1	122 ± 3	56 ± 1
6	162 ± 3 (8)	149 ± 8	28 ± 5	7 ± 1	97 ± 8	45 ± 4
7	221 ± 4 (8)	78 ± 3	11 ± 1	8 ± 1	120 ± 5	82 ± 1
8	252 ± 6 (9)	244 ± 15	41 ± 2	15 ± 1	145 ± 5	51 ± 3
9	206 ± 21 (6)	92 ± 22	18 ± 3	8 ± 3	129 ± 18	42 ± 5
10	217 ± 15 (6)	161 ± 17	25 ± 5	11 ± 2	146 ± 15	35 ± 2
11	251 ± 14 (5)	146 ± 25	24 ± 5	10 ± 3	143 ± 17	35 ± 4
Mean ± SEM	202 ± 9	128 ± 15	21 ± 3	9 ± 1	126 ± 5	42 ± 5

[a]Number of determinations.

TABLE 5. LDL kinetic parameters: normal subjects

Patient	LDL-apoB Conc.	LDL-apoB Pool Size	LDL-apoB/C	LDL-apoB FCR	LDL-apoB Transport Rate
	mg/dl ± SD	*mg/pool*	*ratio ± SD*	*pools/day*	*mg/kg-day*
1	73 ± 7	2247	0.59 ± 0.04	0.28	8.2
2	73 ± 14	2632	0.58 ± 0.08	0.28	9.8
3	67 ± 6	1745	0.59 ± 0.04	0.36	10.8
4	67 ± 7	2958	0.53 ± 0.07	0.34	10.3
5	87 ± 3	3179	0.71 ± 0.02	0.29	12.3
6	71 ± 3	2355	0.73 ± 0.07	0.32	10.0
7	70 ± 3	1818	0.58 ± 0.03	0.40	12.9
8	81 ± 4	2361	0.56 ± 0.05	0.26	7.9
9	83 ± 11	2767	0.64 ± 0.03	0.34	12.5
10	95 ± 9	3724	0.65 ± 0.03	0.28	11.9
11	72 ± 8	2432	0.50 ± 0.01	0.31	9.9
Mean ± SEM	76 ± 3	2583 ± 172	0.61 ± 0.02	0.31 ± 0.01	10.6 ± 0.5

209 ± 23 mg/dl and 166 ± 16 mg/dl, respectively (**Table 8**). However, before the study, some of these patients were noted to have elevated levels of plasma TG. Examples included isolated TG levels of 972, 779, 400, 480, and 600 mg/dl in patients 23–27, respectively. However, none of the patients demonstrated hypertriglyceridemia throughout the month of the turnover study; nonetheless, their HDL-C levels (26 ± 3 mg/dl) were significantly lower than normal ($P < 0.05$).

Concentrations of LDL-C and LDL-apoB in subgroup A averaged 138 ± 18 mg/dl and 87 ± 10 mg/dl, respectively (**Table 9**). The mean FCR was 0.51 ± 0.03 pools/day, and average transport rates of LDL-apoB (19.4 ± 2.8 mg/kg-day) were similar to those of the hypertriglyceridemic patients with CHD and significantly higher than normal ($P < 0.05$). Their LDL-apoB/C ratios (0.66 ± 0.03) were not significantly higher than normal.

Subgroup B. This subgroup included 16 patients. Plasma cholesterol and TG levels were 214 ± 7 mg/dl and 143 ± 13 mg/dl, respectively (Table 8). Mean HDL-C (31 ± 2 mg/dl) was lower than normal ($P < 0.05$). The mean FCR of LDL-apoB for this subgroup was 0.28 ± 0.01 pools/day; transport rates of LDL-apoB averaged 9.9 ± 0.6 mg/kg-day, and LDL-apoB/C ratios were 0.59 ± 0.02 (Table 9). None of these parameters differed from normal. In this subgroup, there was no correlation between LDL-apoB levels and FCR ($r = -0.14$); in contrast, a very high correlation existed between concentrations and transport rates of LDL-apoB ($r = 0.81$; $P < 0.001$).

DISCUSSION

Elevated levels of LDL are known to accelerate atherosclerosis. However, many patients with CHD do not have abnormally high concentrations of LDL. Most

TABLE 6. Plasma lipids and lipoprotein cholesterol: patients with coronary heart disease and hypertriglyceridemia

Patient	Plasma		Lipoprotein cholesterol			
	Chol	TG	VLDL	IDL	LDL	HDL
	mg/dl ± SD		*mg/dl ± SD*			
12	234 ± 32 (7)[a]	761 ± 168	156 ± 44	9 ± 2	49 ± 12	20 ± 3
13	225 ± 19 (7)	966 ± 148	73 ± 13	22 ± 3	104 ± 12	23 ± 3
14	183 ± 19 (6)	980 ± 117	111 ± 8	10 ± 2	45 ± 11	22 ± 4
15	246 ± 19 (5)	361 ± 92	91 ± 18	14 ± 1	114 ± 8	27 ± 5
16	302 ± 9 (5)	1014 ± 136	194 ± 22	16 ± 2	75 ± 11	24 ± 2
17	228 ± 14 (7)	307 ± 58	57 ± 20	15 ± 3	137 ± 23	28 ± 3
18	233 ± 25 (6)	279 ± 84	39 ± 3	16 ± 2	147 ± 22	31 ± 4
19	233 ± 10 (7)	339 ± 36	56 ± 22	15 ± 1	132 ± 13	31 ± 4
20	299 ± 69 (7)	438 ± 65	72 ± 14	23 ± 2	175 ± 17	28 ± 4
21	267 ± 31 (5)	449 ± 115	126 ± 41	28 ± 4	191 ± 29	21 ± 3
Mean ± SEM	245 ± 11[b]	589 ± 96[b]	98 ± 16[b]	17 ± 2[b]	117 ± 16[c]	26 ± 1[d]

[a]Number of determinations.
[b]Significantly greater than normal ($P < 0.05$).
[c]Not significantly different from normal.
[d]Significantly below normal ($P < 0.05$).

TABLE 7. LDL-apoB kinetics: patients with coronary heart disease and hypertriglyceridemia

Patient	LDL-apoB Conc.	LDL-apoB Pool Size	LDL-apoB/C	LDL-apoB FCR	LDL-apoB Transport Rate
	mg/dl ± SD	*mg/pool*	*ratio ± SD*	*pools/day*	*mg/kg-day*
12	54 ± 6	1880	1.1 ± 0.3	0.78	17.5
13	92 ± 5	3589	0.89 ± 0.03	0.37	15.4
14	54 ± 13	1920	1.2 ± 0.1	0.86	19.7
15	89 ± 8	3025	0.78 ± 0.06	0.61	23.9
16	71 ± 6	2461	0.94 ± 0.15	0.66	18.5
17	120 ± 13	4546	0.88 ± 0.02	0.31	17.6
18	112 ± 29	3805	0.76 ± 0.12	0.65	30.9
19	82 ± 7	2468	0.62 ± 0.03	0.50	16.5
20	103 ± 28	3537	0.59 ± 0.03	0.43	20.0
21	109 ± 34	6789	1.1 ± 0.02	0.44	30.2
Mean ± SEM	99 ± 14[a]	3402 ± 464[a]	0.89 ± 0.06[a]	0.56 ± 0.06[a]	21.0 ± 1.7[a]

[a]Significantly higher than normal ($P < 0.05$).

CHD patients with normal LDL levels are normolipidemic, but some have hypertriglyceridemia. Although their levels of LDL are normal, their LDL still might enhance atherogenesis because of a defect in its metabolism. For instance, high concentrations of LDL-apoB in CHD patients with normal levels of LDL-C, reported by Sniderman et al. (7, 10, 16), could be the result of abnormal metabolism of LDL. Furthermore, Kesäniemi and Grundy (13) described a small group of normolipidemic patients with CHD who had an abnormal turnover rate of LDL; the prevalence of this defect in CHD patients however was not determined. More recently, Vega and Grundy (17) reported that the LDL in many hypertriglyceridemic patients with CHD is abnormal

TABLE 8. Plasma lipids and lipoprotein cholesterol: patients with coronary heart disease and normolipidemia

Patient	Plasma		Lipoprotein cholesterol			
	Chol	TG	VLDL	IDL	LDL	HDL
	mg/dl ± SD		*mg/dl ± SD*			
Subgroup A						
22	229 ± 8 (7)[a]	184 ± 43	28 ± 4	20 ± 1	161 ± 7	30 ± 2
23	161 ± 22 (6)	182 ± 14	33 ± 4	15 ± 3	103 ± 21	18 ± 2
24	141 ± 10 (5)	164 ± 9	28 ± 1	13 ± 1	89 ± 9	17 ± 2
25	255 ± 24 (4)	124 ± 9	42 ± 8	24 ± 2	170 ± 13	31 ± 3
26	180 ± 4 (6)	119 ± 13	15 ± 3	15 ± 1	106 ± 4	35 ± 3
27	285 ± 31 (5)	225 ± 32	44 ± 7	31 ± 6	200 ± 35	24 ± 3
Mean ± SEM	209 ± 23[b]	166 ± 16[b]	31 ± 4[b]	20 ± 3[c]	138 ± 18[b]	26 ± 3[d]
Subgroup B						
28	247 ± 22 (5)	239 ± 21	77 ± 7	13 ± 1	156 ± 6	24 ± 2
29	203 ± 24 (7)	123 ± 20	21 ± 1	9 ± 1	126 ± 12	28 ± 3
30	235 ± 38 (7)	151 ± 19	25 ± 5	13 ± 2	173 ± 33	25 ± 2
31	246 ± 50 (7)	142 ± 21	23 ± 6	13 ± 3	186 ± 50	24 ± 1
32	194 ± 39 (7)	149 ± 54	26 ± 6	10 ± 5	135 ± 33	25 ± 3
33	223 ± 9 (7)	136 ± 26	32 ± 5	11 ± 1	152 ± 11	28 ± 2
34	183 ± 20 (7)	121 ± 10	28 ± 9	8 ± 1	118 ± 8	30 ± 3
35	248 ± 13 (6)	117 ± 7	31 ± 3	11 ± 1	164 ± 10	41 ± 5
36	173 ± 13 (6)	157 ± 32	21 ± 2	9 ± 1	116 ± 8	27 ± 2
37	252 ± 29 (6)	79 ± 12	22 ± 6	10 ± 3	162 ± 44	32 ± 9
38	161 ± 19 (6)	97 ± 14	19 ± 5	8 ± 1	106 ± 7	23 ± 2
39	181 ± 14 (5)	134 ± 32	14 ± 4	9 ± 1	124 ± 11	34 ± 4
40	217 ± 10 (6)	81 ± 5	11 ± 3	11 ± 1	165 ± 10	30 ± 1
41	200 ± 12 (5)	208 ± 29	33 ± 2	10 ± 1	125 ± 9	32 ± 2
42	234 ± 12 (5)	246 ± 59	48 ± 12	11 ± 1	135 ± 6	40 ± 3
43	224 ± 5 (7)	111 ± 17	17 ± 4	9 ± 1	135 ± 6	58 ± 3
Mean ± SEM	214 ± 7[b]	143 ± 12[b]	28 ± 4[b]	10 ± 1[b]	142 ± 6[b]	31 ± 2[d]

[a]Number of determinations in parentheses.
[b]Not significantly different from normal.
[c]Significantly higher than normal ($P < 0.05$).
[d]Significantly below normal ($P < 0.05$).

TABLE 9. LDL-apoB kinetics: patients with coronary heart disease and normolipidemia

Patient	LDL-apoB Conc.	LDL-apoB Pool Size	LDL-apoB/C	LDL-apoB FCR	LDL-apoB Transport Rate
	mg/dl ± SD	*mg/pool*	*ratio ± SD*	*pools/day*	*mg/kg-day*
Subgroup A					
22	109 ± 19	3099	0.68 ± 0.10	0.50	22.1
23	66 ± 7	2966	0.64 ± 0.12	0.49	14.5
24	69 ± 6	2220	0.77 ± 0.06	0.56	15.2
25	102 ± 12	4133	0.60 ± 0.06	0.40	18.4
26	61 ± 4	2316	0.58 ± 0.04	0.45	14.5
27	112 ± 8	3747	0.56 ± 0.02	0.63	31.9
Mean ± SEM	87 ± 10[a]	3080 ± 310[a]	0.66 ± 0.04[b]	0.51 ± 0.03[a]	19.4 ± 2.8[a]
Subgroup B					
28	92 ± 10	2793	0.59 ± 0.08	0.34	11.4
29	74 ± 11	2696	0.59 ± 0.06	0.26	8.1
30	127 ± 22	4580	0.73 ± 0.05	0.25	13.9
31	118 ± 11	4755	0.64 ± 0.08	0.23	12.3
32	70 ± 16	2587	0.52 ± 0.07	0.24	7.8
33	105 ± 18	2738	0.69 ± 0.12	0.29	10.4
34	59 ± 8	1944	0.50 ± 0.06	0.29	7.7
35	123 ± 25	3411	0.75 ± 0.09	0.27	10.7
36	58 ± 7	2524	0.50 ± 0.06	0.25	6.5
37	95 ± 14	3259	0.59 ± 0.06	0.25	11.2
38	55 ± 8	1978	0.52 ± 0.03	0.34	8.4
39	61 ± 5	1842	0.49 ± 0.04	0.25	6.9
40	94 ± 7	2967	0.57 ± 0.02	0.27	11.4
41	81 ± 5	2552	0.65 ± 0.03	0.33	12.6
42	85 ± 4	2562	0.63 ± 0.03	0.34	11.6
43	69 ± 7	2785	0.51 ± 0.04	0.22	6.8
Mean ± SEM	86 ± 6[a]	2878 ± 208[a]	0.59 ± 0.02[b]	0.28 ± 0.01[b]	9.9 ± 0.6[b]

[a]Significantly higher than normal ($P < 0.05$).
[b]Not significantly different from normal.

in composition, but this compositional change was not noted in most normolipidemic patients with CHD. The current study thus was designed to further examine the metabolism of LDL in patients with CHD who have essentially normal levels of LDL; its purpose was to determine whether defects in this metabolism might be implicated in atherogenesis.

Hypertriglyceridemia and CHD

A causal relation between elevated plasma TG and risk for CHD has been difficult to demonstrate. Most epidemiological studies have shown an association between high TG levels and prevalence of CHD (3, 39–41), although a high plasma TG per se does not appear to be an independent risk factor (42). Some forms of HTG, such as primary lipoprotein lipase deficiency (43) and familial HTG (44), may not be accompanied by increased risk for CHD. Other forms of HTG almost certainly do enhance risk (3, 44). The mechanisms whereby high TG levels could affect atherosclerosis are not clear. The plasma triglycerides themselves seemingly do not deposit in atherosclerotic lesions. Levels of LDL-C usually are not elevated in patients with HTG. On the other hand, these patients may have lipoprotein abnormalities that promote atherosclerosis. Atherogenic factors could be a low level of HDL (1, 2), abnormal VLDL (11), or elevated LDL-apoB (7, 10, 16). Another possibility is that defects in the metabolism of LDL may enhance atherogenicity.

The kinetics of LDL have been studied previously in a variety of patients with HTG that had not been well classified (45–48). A high FCR of LDL was reported in several studies (8, 45), but not in all (46–48). Increased production of LDL generally was not observed (45–48), but there were exceptions (8). Thus, no common pattern has been noted for HTG in general. However, there are two reports of LDL kinetics in patients being characterized as having familial combined hyperlipidemia. In this disorder multiple lipoprotein phenotypes occur in single families (3, 49, 50). Some affected family members have increases in plasma TG alone, others have high cholesterol levels, and still others have mixed hyperlipidemia. These patients seemingly are at increased risk for CHD regardless of their lipoprotein phenotype (44). Kinetic studies suggest that they have overproduction of both VLDL-TG and VLDL-apoB (21, 51–54). Janus et al. (51, 52) measured LDL turnover in 11 patients with HTG classified as familial

combined hyperlipidemia. In six patients with elevated LDL and VLDL (type 2b HLP), the mean LDL-apoB synthetic rate averaged about 80% above normal, while in the other five with normal LDL levels (type 4 HLP), production rates of LDL were inconsistently increased. Furthermore, in neither group was the FCR of LDL-apoB increased. Likewise, Kissebah, Alfarsi, and Adams (53) showed high production rates of LDL-apoB in patients with familial combined hyperlipidemia, but values for FCR of LDL-apoB were not reported.

Our patients with HTG and CHD frequently had three abnormalities in their LDL metabolism. *a*) Most had overproduction of LDL-apoB. This abnormality is likely the consequence of a high secretion of VLDL-apoB (51, 52, 54). Although patients with HTG may have increased hepatic clearance of VLDL remnants (55–57), excessive amounts of VLDL still are converted to LDL. *b*) FCRs of LDL were increased, frequently markedly so. This abnormality might be the result of an increase in LDL receptors which could have been triggered by the defective metabolism of cholesterol or bile acids in many hypertriglyceridemic patients (21, 58–61). Alternatively, a high FCR of LDL might be due to the third observed abnormality, *c*) a change in composition of LDL as reflected by an increase in the LDL-apoB/C ratio. Previously, Fisher et al. (8) reported that LDL of many hypertriglyceridemic patients are unusually heterogenous (polydisperse); this has been confirmed by others (16, 17, 62). Polydisperse low density lipoproteins have high LDL-apoB/C ratios which might mean that the conformation of apoB on the surface of LDL is altered to enhance affinity for LDL receptors. Another possibility is that the LDL from some hypertriglyceridemic patients retains traces of apoE that facilitate uptake by LDL receptors (63, 64).

Did our hypertriglyceridemic patients have familial combined hyperlipidemia? Most of them conformed to criteria for this condition (49). To classify patients according to their genetic patterns of lipoprotein concentrations however reveals little about their metabolic defect, particularly in this disorder. For example, the mechanisms for multiple lipoprotein phenotypes have not been defined. If the underlying defect is overproduction of VLDL-apoB, why do some patients have elevated TG levels while others do not? Those with HTG likely have a concomitant defect in TG metabolism, either overproduction or defective lipolysis of VLDL-TG (21). The marked HTG of some of our patients suggests an element of defective clearance. However, defective lipolysis cannot explain a high input of LDL-apoB, so our patients probably had increased secretion of VLDL-apoB as well.

Next we might consider whether these patients had hyperapobetalipoproteinemia. This condition was defined by Sniderman et al. (7, 10) as abnormally high levels of LDL-apoB (i.e., over 120 mg/dl) in the presence of normal LDL-C levels. Only one patient (No. 21) fitted these criteria. Most of our patients did not have elevated LDL-apoB because of a high clearance rate of LDL. Some patients however may fail to have as great a compensatory increase in LDL clearance, and their LDL levels will be higher. Since LDL-apoB/C ratios are high in many patients with HTG (17), a delay in LDL clearance should cause LDL-apoB levels to rise to abnormally high levels before LDL-C concentrations reach the abnormal range; such patients likely will demonstrate hyperapobetalipoproteinemia. However, our data indicate that many hypertriglyceridemic patients with CHD do not have such high LDL levels.

How might the abnormalities in LDL metabolism in our patients with HTG have contributed to their premature CHD? Several possibilities can be considered. A high influx of LDL most likely is associated with an increased transport of LDL-C. This excess cholesterol might either enter the arterial wall in some way or interfere with reverse cholesterol transport (65). Alternatively, compared to normal LDL, the LDL of some hypertriglyceridemic patients could be more atherogenic as suggested by their abnormal composition and high FCRs.

Normotriglyceridemic patients with premature CHD

Kesäniemi and Grundy (13) recently described a group of patients with premature CHD who had normal LDL levels but increased transport rates of LDL. Their LDL kinetics were similar to those of our current patients with HTG; in both there were increased production rates of LDL-apoB, raised FCRs of LDL-apoB, and high LDL-apoB/C ratios. The previous study (13) raised the question but did not answer it: how common is this abnormal pattern of lipoprotein kinetics in normolipidemic patients with CHD? Therefore in the current study we examined more patients. Although we cannot claim an accurate prevalence, the present results should provide a rough guide to the frequency of this abnormality.

Twenty-two normolipidemic patients with CHD were studied. Six (27%) (subgroup A, Table 9) had the same pattern reported before (13), i.e., high production rates and relatively high FCRs of LDL (over 0.40 pools/day). One difference however was that the current patients did not always have LDL-apoB/C ratios. Their overall kinetics nonetheless appeared to differ enough from the other normolipidemic patients with CHD to justify categorizing them as different. One characteristic of these patients was intermittent HTG which might provide a clue to their identification.

The current results strongly suggest that most normolipidemic patients with CHD do not have the above disorder. For example, the FCRs of LDL-apoB in the remaining normolipidemic patients (subgroup B, Table 9) were relatively low. Twelve of 16 patients had FCRs below 0.30 pools/day. Because of this tendency for low FCRs, some of the patients may have had higher levels of LDL-C before development of CHD. Lifestyle changes after a coronary event may have been altered to some extent to produce lower concentrations of LDL-C.

Although the FCRs of this subgroup were relatively uniform, their transport rates of LDL-B exhibited more variability, ranging from about 6.0 to 13.0 mg/kg-day. These differences in production rates were mainly responsible for the variability in LDL-apoB levels within this subgroup; these levels ranged from 52 to 118 mg/dl. Thus, as previously shown by Kesäniemi and Grundy (12), production rates of LDL-apoB are a major determinant of LDL-apoB levels in most patients without HTG. Factors regulating LDL-apoB production in these patients could be twofold: *a*) the production rates of VLDL-apoB, and *b*) the fractional conversion of VLDL-apoB to LDL-apoB. Since the patients of this subgroup in general were not overweight, it is unlikely that variability in input rates of VLDL-apoB was sizable. Thus, the fractional conversion of VLDL-apoB to LDL-apoB likely was the major factor regulating formation of LDL-apoB. The fraction of VLDL-apoB converted to LDL-apoB is determined in part by LDL receptors because the latter are responsible for removal of VLDL remnants (64). Consequently, variability in LDL receptor activity as it affects removal of VLDL remnants could have been an important regulator of LDL-apoB input.

The foregoing suggests the existence of two types of normolipidemic patients with CHD. One type has elevated transport rates of LDL, but they maintain normal LDL levels by a compensatory increase in FCR. This group is prone to intermittent HTG. In contrast, most patients with CHD appear to have relatively normal production rates of LDL, and their FCRs of LDL are normal or somewhat low. Concentrations of LDL in this latter group are determined mainly by variability in input rates of LDL, but in general this variability in input exists within the normal range; also, it probably is determined more by the fractional conversion of VLDL-apoB to LDL-apoB than by secretion rates of VLDL-apoB.

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