Clearance of chylomicron remnants in normolipidaemic patients with coronary artery disease: case control study over three years

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Abstract

Objective—To test the hypothesis that subjects who clear chylomicron remnants slowly from plasma may be at higher risk of coronary artery disease than indicated by their fasting plasma lipid concentrations.

Design—Case control study over three years.

Setting—An 800 bed general municipal hospital.

Subjects—85 normolipidaemic patients with coronary artery disease selected prospectively and matched with 85 normolipidaemic subjects with normal coronary arteries on angiography.

Interventions—All subjects were given a vitamin A fat loading test which specifically labels intestinal lipoproteins with retinyl palmitate.

Main outcome measure—Postprandial lipoprotein metabolism.

Results—The area below the chylomicron remnant retinyl palmitate curve was significantly increased in the coronary artery disease group as compared with the controls (mean 23·4 (SD 15·0) v 15·3 (8·9) μmol/l.h; 95% confidence interval of difference 4·37 to 11·82).

Conclusions—Normolipidaemic patients with coronary artery disease had significantly higher concentrations of chylomicron remnants in plasma than normolipidaemic subjects with normal coronary vessels. This may explain the mechanism underlying the susceptibility to atherosclerosis of coronary artery disease patients with normal fasting lipid values. As diet and drugs can ameliorate the accumulation of postprandial lipoproteins in plasma, the concentration of chylomicron remnants should be measured in patients at high risk of coronary artery disease.

Introduction

Measurements in fasting subjects show that plasma lipid and lipoprotein abnormalities are associated with coronary and peripheral atherosclerosis.[1-4] However, most of our lives are spent in the postprandial state, during which vessel walls are exposed to triglyceride rich lipoproteins—namely, chylomicrons and chylomicron remnants. Animal and human studies[5-10] suggest that these lipoproteins may be atherogenic. They are metabolised on the endothelial surface of arteries and their cholesterol becomes incorporated into the artery wall, where it may stimulate the formation of atherosclerotic lesions.[11-15] The accumulation of postprandial lipoproteins in plasma due to slowed catabolism may therefore enhance atherosclerosis in otherwise normolipidaemic subjects.

This picture is not detected by examining fasting state plasma samples only. Zilversmit was the first to publish this hypothesis,[5] which was followed by many confirmatory studies. Most, however, were small and provided only indirect support by showing correlations between postprandial lipoprotein metabolism and other well known risk factors for coronary artery disease.[16-20] Only one study found that coronary artery disease patients with normal fasting lipid values had

higher concentrations of postprandial lipoproteins than patients without coronary artery disease.[21] That study, however, included only 20 patients.

There are two main difficulties in testing Zilversmit's hypothesis. The first is in differentiating postprandial lipoproteins from endogenous very low density lipoproteins, which have similar physical and chemical properties. The second is the need to study a large number of coronary artery disease patients with normal fasting lipid concentrations and an age and sex matched control group. We overcame the first difficulty by using the vitamin A fat loading test, which specifically labels postprandial lipids with retinyl palmitate.[22-28] This method reliably detects both chylomicron and chylomicron remnant metabolism.[16 18-20 29] With respect to the size of the study we selected 85 normolipidaemic subjects with radiologically normal coronary arteries (controls) and matched them for age and sex with 85 normolipidaemic patients with advanced coronary artery disease. We investigated all participants for metabolic behaviour of postprandial lipoproteins.

Subjects and methods

The 85 patients with coronary artery disease and 85 controls were selected prospectively from the cardiology department of this hospital during February 1990 and April 1993. All patients and controls had normal fasting lipid and low density lipoprotein cholesterol concentrations. All had undergone coronary angiography because of chest pain suspected to be of coronary origin and all had stable cardiovascular and metabolic variables and were free of congestive heart failure, diabetes mellitus, chronic renal failure, and endocrine disease. No woman patient was taking contraceptive or hormone replacement drugs.

Selection criteria with regard to lipid and lipoprotein concentrations were: total cholesterol less than 6·2 mmol/l, triglycerides less than 2·0 mmol/l, and low density lipoprotein cholesterol less than 4·1 mmol/l. High density lipoprotein cholesterol concentrations were not used as a criterion for exclusion. Coronary vessels with less than 20% stenosis were defined as normal. Patients in the coronary artery disease group had two or three vessel disease with at least 80% occlusion.

MATCHING CRITERIA AND METHOD OF SELECTION

During the three years we selected 85 subjects with normal coronary vessels and 192 patients with coronary artery disease who fulfilled the study criteria. In most cases we successfully matched the two groups according to the following criteria with priority in descending order: sex, age, body surface area, hypertension, drugs being taken, and cigarette smoking. Hypertension was defined as systolic blood pressure >150 mm Hg and diastolic pressure >90 mm Hg treated for at least two years. Cigarette smoking was defined as consumption of more than 10 cigarettes daily for at least five years. All the participants claimed to have stopped smoking during the study. The two

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groups were matched for treatment with β blockers (treatment equivalent 60-160 mg propranolol daily), angiotensin converting enzyme inhibitors (treatment equivalent 12·5-50·0 mg captopril daily), and calcium inhibitors (verapamil 120-240 mg and nifedipine 30-60 mg daily) because of the known effects of these drugs on lipid metabolism. Patients taking diuretics or very high doses of the above drugs were excluded.

Finding normal coronary vessels on angiography is fairly rare. Whenever such patients were detected they were asked to continue the drugs they were taking for suspected angina until they could have a vitamin A fat loading test. This was never less than one week after the angiography. Thereafter, a patient with proved coronary artery disease who had undergone coronary angiography and who fulfilled the inclusion criteria was matched as closely as possible with a non-coronary artery disease patient and also given the vitamin A fat loading test. Of the 192 patients with coronary artery disease, 85 were so matched to controls during the three years. All participants in the study gave fully informed consent.

VITAMIN A FAT LOADING TEST

The vitamin A fat loading test was performed as described.[19] After a 12 hour overnight fast participants were given a fatty meal plus 60 000 units of aqueous vitamin A/m^2 body surface area. The meal contained 50 g fat/m^2 body surface area and 145 mg cholesterol/MJ 65% of the energy being taken as fat, 20% as carbohydrate, and 15% as protein. Blood samples were drawn before the meal, hourly thereafter for eight hours, and at 10 hours. Subjects tolerated the meal well and none had diarrhoea or other symptoms of malabsorption.

ANALYSIS OF SAMPLES

Venous blood was drawn from the forearm and transferred to a tube containing sodium EDTA. Samples were centrifuged immediately at 1500 g for 15 minutes and 1 ml plasma stored wrapped in foil at −20°C for retinyl ester assay. A further 0·5 ml was stored at 4°C for triglyceride determinations. An aliquot of 2·5 ml plasma was subjected to preparative ultracentrifugation to separate chylomicron fraction (>1000 S$_f$ units) from non-chylomicron fraction (<1000 S$_f$ units).[29-32]

RETINYL ESTER ASSAY AND LIPID AND LIPOPROTEIN DETERMINATIONS

The assays were carried out in subdued light with high performance liquid chromatography grade solvents as described.[16] [18-20] Cholesterol and triglyceride concentrations were measured enzymatically by using the reagents cholesterol 236991 and triglyceride 126012 (Boehringer Mannheim, Indianapolis, United States). High density lipoprotein cholesterol was measured after precipitation of whole plasma with dextran sulphate magnesium.

STATISTICAL ANALYSIS

The significance of differences between the groups in clinical characteristics and fasting plasma lipid and lipoprotein concentrations was determined by χ^2 test for discrete variables and a two sample t test for continuous variables. The significance of metabolic responses after the fatty meal was analysed by assuming the same behaviour of the curves of plasma triglyceride, chylomicron retinyl palmitate, and non-chylomicron retinyl palmitate values in the two groups. Areas under the curves were calculated for each patient and the Mann-Whitney test used to detect differences between the groups.[33] A non-parametric test was used because data were skewed. Spearman's test was used to evaluate the correlation between chylomicron retinyl palmitate and non-chylomicron plasma retinyl palmitate concentrations, triglyceride responses, and fasting lipid and lipoprotein concentrations, which were not normally distributed.

Stepwise multiple regression analysis was performed to adjust the metabolic response after the fatty meal for the difference in intake of nitrates and aspirin between the coronary artery disease patients and the controls. Analysis of covariance was performed to adjust the metabolic response for the difference in baseline triglyceride and high density lipoprotein cholesterol concentrations between the groups.
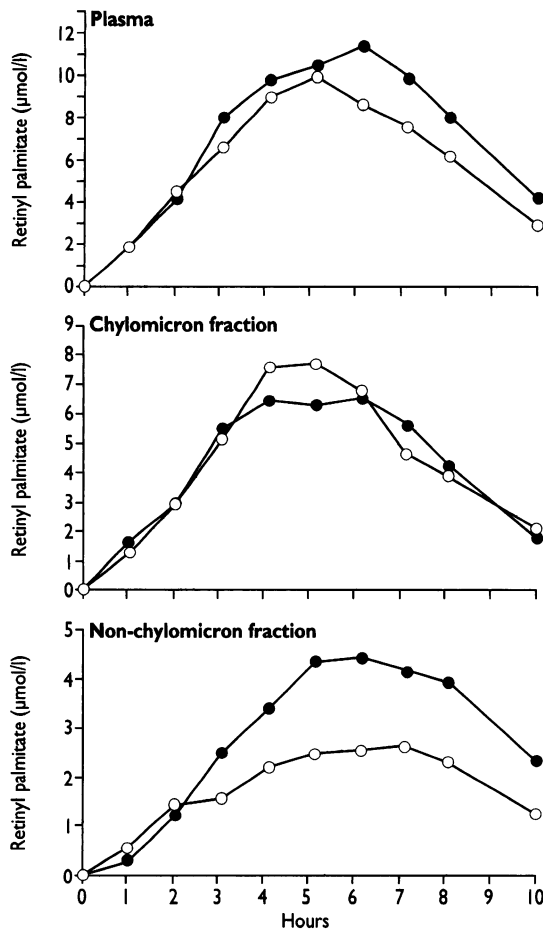
Results

CLINICAL FEATURES

Table 1 presents the clinical features of the coronary artery disease patients and controls. The groups were similar in age, body surface area, male to female ratio, fasting total cholesterol and low density lipoprotein cholesterol concentrations, cigarette smoking, and prevalence of hypertension. The coronary artery disease group was significantly different from the controls in fasting plasma triglyceride (mean 1·6 (SD 0·3) v 1·4 (0·4) mmol/l; P < 0·001) and high density lipoprotein cholesterol concentrations (1·1 (0·2) v 1·4 (0·4) mmol/l; P < 0·0001), though no patient was hypertriglyceridaemic. Significantly more coronary artery disease patients than controls were taking aspirin (77 v 43; P < 0·0001) and nitrates (46 v 21; P < 0·0001). Thirty two coronary artery disease patients had two vessel disease and 53 three vessel disease.

RESPONSES TO VITAMIN A FAT LOADING TEST

Plasma, chylomicron, and non-chylomicron retinyl palmitate and plasma triglyceride responses to the vitamin A fat loading test were determined for each participant. Figures 1 and 2 give the mean results in each group. Table 2 shows the mean areas below the retinyl palmitate and triglyceride curves of the different fractions in the two groups. The area below the plasma retinyl palmitate curve was significantly larger in the coronary artery disease patients than in the controls (mean 65·3 (SD 28·3) v 55·0 (27·5) μmol/l.h; P = 0·017). This increase in postprandial lipoprotein concentration in the coronary artery disease group was due mainly to the non-chylomicron fraction, whose

Table 1—*Characteristics of study patients. Except where stated otherwise, values are means (SD)*

	Coronary artery disease patients (n=85)	Controls (n=85)	P value (unmatched variables only)
Age (years) [range]	56·9 (8·4) [41-70]	59·1 (8·9) [46-68]	
Body surface area (m²) [range]	1·86 (0·2) [1·56-2·05]	1·74 (0·1) [1·58-2·09]	
Sex:			
No of men	63	63	
No of women	22	22	
Severity of coronary artery disease:			
No with two vessel disease	32		
No with three vessel disease	53		
Mean fasting lipids and lipoproteins (mmol/l):			
Total cholesterol [range]	5·3 (0·5) [4·4-6·0]	5·6 (0·5) [4·7-5·9]	0·235
Triglycerides [range]	1·6 (0·3) [0·8-2·0]	1·4 (0·4) [0·8-1·8]	<0·001
Low density lipoprotein cholesterol [range]	3·6 (0·3) [3·0-4·1]	3·6 (0·6) [2·7-4·0]	0·363
High density lipoprotein cholesterol [range]	1·1 (0·2) [0·8-1·5]	1·4 (0·4) [0·9-2·0]	<0·0001
No smoking > 10 cigarettes/day for five years	36	31	
No with hypertension (> 150/90 mm Hg)	38	42	
No receiving drugs:			
β blockers (propranolol 60-160 mg/day)	41	41	
Angiotensin converting enzyme inhibitors (capropril 12·5-50·0 mg/day)	25	26	
Verapamil 120-240 mg/day	19	20	
Nifedipine 30-60 mg/day	16	12	
Aspirin (not matched)	77	43	<0·0001
Nitrates (not matched)	46	21	<0·0001

Fig 1—Total plasma, chylomicron, and non-chylomicron retinyl palmitate concentration curves in 85 patients with coronary artery disease (●) and 85 controls (○). Points are means

area under the curve was highly significantly larger than in the controls (23·4 (15·0) v 15·3 (8·9) μmol/l.h; P<0·001).

There was also evidence of higher postprandial plasma triglyceride concentrations in the coronary artery disease patients than in the controls. The areas below the plasma triglyceride concentration curves in the two groups were 23·6 (SD 7·2) and 21·1 (9·2) mmol/l.h respectively (P=0·055). No significant differences were found in the chylomicron fractions between the two groups. Adjustment for baseline differences in triglyceride and high density lipoprotein cholesterol concentrations showed a non-significant difference in area below the plasma retinyl palmitate curve between the coronary artery disease patients and the controls (P=0·43 after adjustment for triglyceride concentrations; P=0·51 after adjustment for high density lipoprotein cholesterol concentrations). However, the significant difference in area below the non-chylomicron retinyl palmitate curve between the coronary artery disease patients and the controls was independent of baseline differences in triglyceride (P<0·001) and high density lipoprotein cholesterol concentrations (P<0·001).

To determine factors that might play a part in exogenous fat metabolism and atherosclerosis we sought correlations between chylomicron and non-chylomicron retinyl palmitate and plasma triglyceride responses and fasting lipid and lipoprotein concentrations (table 3). Significant positive correlations were found between fasting triglyceride concentrations and chylomicron as well as chylomicron remnant fractions in all participants (r=0·50 and r=0.42 respectively; P<0·001). Negative correlations were found between fasting high density lipoprotein cholesterol concentra-

Table 2—Plasma, chylomicron, and non-chylomicron retinyl palmitate and plasma triglyceride areas under curves in patients with coronary artery disease and controls. Values are means (SD)
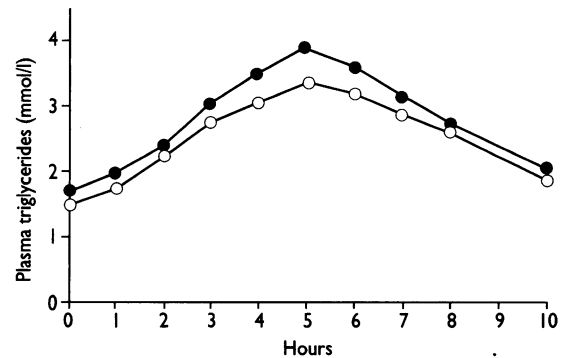
	Area under retinyl palmitate curves (μmol/l.h)			Area under triglyceride curve (mmol/l.h)
Group	Plasma	Chylomicron	Non-chylomicron	
Coronary artery disease patients (n=85)	65·3 (28·3)	41·9 (21·3)	23·4 (15·0)	23·6 (7·2)
Controls (n=85)	55·0 (27·5)	39·9 (21·1)	15·3 (8·9)	21·1 (9·2)
P	0·017	0·539	0·0001	0·055
P₁	0·43	0·25	0·001	0·044
P₂	0·51	0·24	0·001	0·59

P=Before adjustment for baseline differences in triglyceride and high density lipoprotein cholesterol concentrations. P₁=After adjustment for baseline difference in triglyceride values. P₂=After adjustment for baseline difference in high density lipoprotein cholesterol concentration.

Fig 2—Plasma triglyceride concentration curves in 85 patients with coronary artery disease (●) and 85 controls (○). Points are means

Table 3—Correlation coefficients between retinal palmitate areas below curves in chylomicron and non-chylomicron fractions; area below triglyceride curve; and total cholesterol, triglyceride, low density lipoprotein cholesterol, and high density lipoprotein cholesterol concentrations

Variable	Area below curve	Coronary artery disease patients	Controls	All participants
Fasting total cholesterol	Chylomicron fraction	0·30	0·05	0·15
	Non-chylomicron fraction	0·12	−0·14	−0·09
	Triglycerides	0·33	0·02	0·1
Fasting triglycerides	Chylomicron fraction	0·44*	0·57*	0·50*
	Non-chylomicron fraction	0·20	0·51*	0·42*
	Triglycerides	0·92*	0·95*	0·94*
Fasting low density lipoprotein cholesterol	Chylomicron fraction	0·23	0·17	0·19
	Non-chylomicron fraction	0·18	−0·16	−0·01
	Triglycerides	0·13	0·15	0·14
Fasting high density lipoprotein cholesterol	Chylomicron fraction	−0·23*	−0·39*	−0·31*
	Non-chylomicron fraction	−0·09	−0·31	−0·32*
	Triglycerides	−0·16	−0·53	−0·43*

*P<0·001.

Key messages

● Postprandial lipoprotein, chylomicrons, and chylomicron remnants have atherogenic potential through enrichment in cholesterol

● The atherogenic effect of postprandial lipoproteins is hypothesised to be inversely related to their metabolic capacity

● Patients with coronary artery disease have decreased metabolic capacity of chylomicron remnants as compared with subjects with normal coronary arteries

● An ensuing postprandial dyslipidaemia may exist in patients with coronary artery disease who have normal fasting lipid values

● A vitamin A fat loading test may be useful in identifying these patients

tions and chylomicron as well as chylomicron remnant retinyl palmitate areas and areas below plasma triglyceride curves in all participants ($r=-0.31$, $r=-0.32$, and $r=-0.43$ respectively; $P<0.001$)

The significant difference in area below the non-chylomicron retinyl palmitate curve between the coronary artery disease patients and the controls ($P<0.0001$) was not explained by the different intake of nitrates ($P=0.078$) or aspirin ($P=0.075$).

Discussion

This study shows clearly that coronary artery disease patients with normal fasting lipid and low density lipoprotein cholesterol concentrations have significantly higher concentrations of chylomicron remnants in plasma than non-coronary artery disease patients with normal fasting lipid values. The findings therefore strongly support Zilversmit's hypothesis that atherogenesis is a postprandial phenomenon.[5]

Postprandial lipoprotein metabolism is believed to occur in two stages. Initially, the chylomicrons produced in the intestines which carry exogenous lipids interact with lipoprotein lipase in extrahepatic tissues. This results in triglyceride hydrolysis and the delivery of free fatty acids to the tissues.[11 34 35] After most of the triglycerides are hydrolysed remnant particles are formed,[36 37] which are removed from the circulation by hepatocyte receptors that recognise apolipoprotein E.[38 39] Though considerable postprandial exchange between lipoproteins of total retinyl esters has been reported in humans,[40] little exchange of retinyl palmitate has been shown between intestine derived lipoproteins and other lipoproteins.[23] Also, most of the exchange of retinyl esters was found in the low density lipoprotein fraction. Our study groups had similar low density lipoprotein concentrations. Hence this exchange would not affect the difference found in the non-chylomicron fractions of the two groups.

Karpe et al reported a delayed appearance of retinyl palmitate compared with the patterns of increase of plasma triglycerides and apolipoprotein B-48.[41] This finding questions the validity of retinyl palmitate labelling of chylomicrons. In hundreds of tests performed by us[16 18-20 29] and in many other studies[21-28] plasma triglyceride and retinyl palmitate concentrations peaked simultaneously. This disparity may be explained by the different methods used. Karpe et al used exclusively soybean oil in their fat load meal whereas we and others used dairy cream. Polyunsaturated fat delays the absorption of vitamin A.[42]

We think that the vitamin A fat loading method as used by us in the past eight years is an efficient tool for studying postprandial lipoprotein metabolism. Results with this method indicate that coronary artery disease patients clear chylomicron remnants at a significantly slower rate than do subjects with normal coronary vessels. Chylomicron remnants accumulated in the

circulation may be particularly atherogenic. In type III hyperlipoproteinaemia accumulation of these fat particles due to delayed clearance is associated with severe premature atherosclerotic disease.[68] Thus our findings may explain the mechanism underlying the susceptibility to atherosclerosis in the subgroup of coronary artery disease patients with normal fasting lipid values

EFFICIENCY OF CATABOLIC MECHANISM

Though fasting lipid and lipoprotein concentrations were normal in our two groups of patients, the coronary artery disease group had significantly higher concentrations of fasting triglycerides and lower concentrations of high density lipoprotein cholesterol. This disparity, however, had no influence on non-chylomicron metabolism. All participants showed significant positive correlations between fasting triglyceride concentrations and postprandial lipoprotein areas under the curve ($P<0.001$) and significant negative correlations between fasting high density lipoprotein cholesterol concentrations and postprandial lipoprotein concentrations. These observations were reported in most earlier studies on postprandial lipoprotein metabolism and accord with current concepts on the relations between the metabolic rates of these lipoproteins, the fasting concentrations of triglycerides, and the surface remnant transfer to the high density lipoprotein system.[43 44]

Could the difference in fasting triglyceride and high density lipoprotein concentrations rather than the difference in remnant clearance in the two groups be the determining factor in coronary artery disease? The catabolic pathways of all the triglyceride rich lipoproteins, endogenous very low density lipoproteins, intermediate density lipoproteins, and exogenous chylomicrons and chylomicron remnants are identical. They are lipolysed by the same enzyme—namely, lipoprotein lipase—and taken up by the same receptors. It seems that this catabolic mechanism may be less efficient in patients with coronary artery disease. Measuring only fasting lipid and lipoprotein values will not detect this "defect." However, in the postprandial state, when the catabolic pathways are confronted by an increased mass of exogenous triglycerides and lipoproteins, this defect is augmented and can be detected by the fat loading test.

Thus in patients with coronary artery disease high normal fasting plasma triglyceride concentrations, increased concentrations of chylomicron remnants, and low concentrations of high density lipoprotein cholesterol are due to the same inefficient catabolic mechanism. Borderline concentrations of fasting triglycerides and high density lipoprotein cholesterol should therefore raise suspicion of a possible accumulation of postprandial lipoproteins, especially in high risk patients. In these cases a vitamin A fat loading test can be done. Though the test is cumbersome and time consuming, it may be simplified. We find that testing one or two blood samples drawn seven or eight hours or both after a fat load may detect a defect in chylomicron remnant removal. Testing a single blood sample drawn at five or six hours will detect defects in chylomicron lipolysis.

In conclusion our study shows that there is a substantial accumulation of chylomicron remnants in patients with premature coronary heart disease and normal fasting lipid and low density lipoprotein cholesterol concentrations. It seems that the arteries of these patients are exposed to high concentrations of chylomicron remnants for prolonged periods. This may have a role in the aetiology of the disease, as first suggested by Zilversmit.[5] The accumulation of chylomicron remnants can be ameliorated dramatically by diet or fibric acid derivatives.[29] The vitamin A fat

loading test as described or a simplified version may be useful for identifying these patients.

We thank Professor Jan L Breslow, of the Rockefeller University, New York, for help in planning the study and support in developing the methods for studying postprandial lipoprotein metabolism.

1 Lipid Research Clinics. Coronary primary prevention trial results. II: The relationship of reduction in incidence of coronary heart disease to cholesterol lowering. *JAMA* 1984;251:365-74.
2 Gordon T, Castelli WP, Hjortland MD, Kannel WB, Dawber TR. High density lipoprotein as a protective factor against coronary heart disease. The Framingham study. *Am J Med* 1977;62:707-14.
3 Miller NE, Hammet F, Saltissi S. Relation of angiographically defined coronary artery disease to plasma lipoprotein subfractions and apolipoproteins. *BMJ* 1981;282:1741-4.
4 Lippel K, Tyroler H, Eder H. Meeting summary: relationship of hypertriglyceridemia to atherosclerosis. *Arteriosclerosis* 1981;1:406-17.
5 Zilversmit DB. Atherogenesis: a postprandial phenomenon. *Circulation* 1979;60:473-85.
6 Morganroth J, Levy RI, Fedrickson DS. The biochemical, clinical and genetic features of type III hyperlipoproteinemia. *Ann Intern Med* 1975;82:158-64.
7 Nestel PJ, Fidge NH, Tan NH. Increased lipoprotein remnant formation in chronic renal failure. *N Engl J Med* 1982;307:327-33.
8 Kane JP, Chen GC, Hamilton RL, Hardman DA, Malloy MJ, Havel RJ. Remnants of lipoproteins of intestinal and hepatic origin in familial dysbetalipoproteinemia. *Arteriosclerosis* 1983;3:47-56.
9 Thompson KH, Zilversmit DB. Plasma very low density lipoprotein (VLDL) in cholesterol-fed rabbits: chylomicron remnants or liver lipoprotein? *J Nutr* 1983;113:2002-9.
10 Floren CH, Albers JJ, Bierman EL. Uptake of chylomicron remnants causes cholesterol accumulation in cultured human arterial smooth muscle cells. *Biochim Biophys Acta* 1981;663:336-49.
11 Blanchett-Mackie EJ, Scow RO. Sites of lipoprotein lipase activity in adipose tissue perfused with chylomicrons. Electron microscopic, cytochemical study. *J Cell Biol* 1971;51:1-25.
12 Stender S, Zilversmit DB. Arterial influx of esterified cholesterol from two plasma lipoprotein fractions and its hydrolysis in vivo in hypercholesterolemic rabbits. *Atherosclerosis* 1981;39:97-109.
13 Stender S, Zilversmit DB. Comparison of cholesterol ester transfer from chylomicrons and other plasma lipoproteins to aorta intima-media of cholesterol-fed rabbit. *Arteriosclerosis* 1982;2:493-9.
14 Goldstein JL, Ho YK, Brown MS, Innerarity TL, Mahley RW. Cholesterol ester accumulation in macrophages resulting from receptor-mediated uptake and degradation of hypercholesterolemic canine beta very low density lipoproteins. *J Biol Chem* 1980;255:1839-48.
15 Gianturco SH, Lin AH, Hwang SL, Young J, Brown SA, Via DP. Distinct murine macrophage receptor pathway for human triglyceride-rich lipoproteins. *J Clin Invest* 1988;82:1633-43.
16 Weintraub MS, Zechner R, Brown A, Eisenberg S, Breslow JL. Dietary polyunsaturated fats of W-6 and W-3 series reduce postprandial lipoprotein levels. Chronic and acute effects of fat saturation on postprandial lipoprotein metabolism. *J Clin Invest* 1988;82:1884-93.
17 Demacker PNM, Reijnen IGM, Katan MB, Stuyt PMJ, Stalenhoef AFH. Increased removal of remnants of triglyceride-rich lipoproteins on a diet rich in polyunsaturated fatty acids. *Eur J Clin Invest* 1991;21:197-203.
18 Weintraub MS, Rosen Y, Otto R, Eisenberg S, Breslow JL. Physical exercise conditioning in the absence of weight loss reduces fasting and postprandial triglyceride-rich lipoprotein levels. *Circulation* 1989;79:1007-14.
19 Weintraub MS, Eisenberg S, Breslow JL. Dietary fat clearance in normal subjects is regulated by genetic variation in apolipoprotein E. *J Clin Invest* 1987;80:1571-7.
20 Weintraub MS, Ringel Y, Rassin T, Arad J, Bodner G, Liron M. Octogenarian subjects have low postprandial levels of chylomicron remnants: a possible explanation for protection against atherosclerosis. *Journal of Gerontology* 1992;47:B209-13.
21 Groot PHE, van Stiphout WAHJ, Krauss XH, Jansen H, van Tol A, van Ramshaorst E, et al. Postprandial lipoprotein metabolism in normolipidemic men with and without coronary artery disease. *Arterioscler Thromb* 1991;11:653-62.
22 Hazard WR, Bierman EL. Delayed clearance of chylomicron remnants following vitamin A-containing oral fat load in broad-B disease (type III hyperlipoproteinemia). *Metabolism* 1976;25:777-801.
23 Wilson DE, Chan I, Ball M. Plasma lipoprotein retinoids after vitamin A feeding in normal man. Minimal appearance of retinyl esters among low density lipoproteins. *Metabolism* 1983;32:514-7.
24 Berr F, Kern F Jr. Plasma clearance of chylomicrons labelled with retinyl palmitate in healthy human subjects. *J Lipid Res* 1984;25:805-12.
25 Wilson DE, Chan I, Buchi KN, Horton SC. Postchallenge plasma lipoprotein retinoids: chylomicron remnants in endogenous hypertriglyceridemia. *Metabolism* 1985;34:551-8.
26 Berr F, Eckel RH, Kern F. Plasma decay of chylomicron remnants is not affected by heparin-stimulated plasma lipolytic activity in normal fasting man. *J Lipid Res* 1985;26:852-9.
27 Berr F, Eckel RH, Kern F. Contraceptive steroids increase hepatic uptake of chylomicron remnants in healthy young women. *J Lipid Res* 1986;27:645-51.
28 Cortner JA, Coates PM, Lee NA, Cryer DD, Ragni MC, Faulkner A, et al. Kinetics of chylomicron remnant clearance in normal and hyperlipoproteinemic subjects. *J Lipid Res* 1987;28:195-206.
29 Weintraub MS, Eisenberg S, Breslow JL. Different patterns of postprandial metabolism in normals and type IIa, type III and type IV hyperlipoproteinemics: effects of treatment with cholestyramine and gemfibrozil. *J Clin Invest* 1987;79:1110-9.
30 Grundy SM, Mok HYI. Chylomicron clearance in normal and hyperlipidemic man. *Metabolism* 1976;25:1225-39.
31 Dale VP, Hamlin JI. Particulate fat in lymph and blood. *Physiol Rev* 1962;42:674-701.
32 Lindgren FT, Jensen LC, Hatch FT. The isolation and quantitative analysis of serum lipoproteins. In: Nelson GS, ed. *Blood lipids and lipoproteins: quantitation, composition and metabolism.* New York: Wiley Interscience, 1972:181-274.
33 Matthews JNS, Altman DG, Campbell MJ, Royston P. Analysis of serial measurements in medical research. *BMJ* 1990;300:230-5.
34 Korn ED. Clearing factor, a heparin activated lipoprotein lipase. I. Isolation and characterization of the enzyme from normal rat heart. *J Biol Chem* 1955;215:1-14.
35 Schoefe GI, French JE. Vascular permeability to particulate fate: morphological observation on levels of lactating mammary gland and of lung. *Proceedings of the Royal Society of London. Series B: Biological Sciences* 1968;169:153-65.
36 Redgrave TG. Formation of cholesterol ester-rich particulate lipid during metabolism of chylomicrons. *J Clin Invest* 1970;49:465-71.
37 Goodman DS. The metabolism of chylomicron cholesterol ester in the rat. *J Clin Invest* 1962;41:1886-96.
38 Hui DY, Innerarity TL, Mahley RW. Lipoprotein binding to canine hepatic membranes: metabolically distinct apo-E and apo-B,E receptors. *J Biol Chem* 1981;256:5646-55.
39 Hui DY, Brecht WJ, Hall EA, Friedman G, Innerarity TL, Mahley RW. Isolation and characterization of the apolipoprotein E receptor from canine and human liver. *J Biol Chem* 1986;261:4256-67.
40 Krainski SD, Cohn JS, Russel RM, Schaefer EJ. Postprandial plasma vitamin A metabolism in humans: a reassessment of the use of plasma retinyl esters as markers for intestinally derived chylomicrons and their remnants. *Metabolism* 1990;39:357-65.
41 Karpe F, Bell M, Bjorkegren J, Hamsten A. Quantification of postprandial triglyceride-rich lipoproteins in healthy men by retinyl ester labeling and simultaneous measurement of apoliporotein B-48 and B-100. *Arterioscler Thromb Vasc Biol* 1995;15:199-207.
42 Hollander D. Intestinal absorption of vitamins A, E, D, and K. *J Lab Clin Med* 1981;97:449-62.
43 Patsch JR, Karlin JB, Scott LW, Smith LC, Gotto AM Jr. Inverse relationship between blood levels of high density lipoprotein subfractions 2 and magnitude of postprandial lipemia. *Proc Natl Acad Sci USA* 1983;80:1449-53.
44 Eisenberg S. High density lipoprotein metabolism. *J Lipid Res* 1984;25:1017-58.

ONE HUNDRED YEARS AGO

ENGLISH DOCTORS ON THE CONTINENT.

The question of medical reciprocity between this country and the Continent is one of practical importance, not only to the members of our profession, but to the public at large. The number of English-speaking tourists, in France and Switzerland especially, is so large, and their presence in the Continental health resorts is such an undoubted advantage to these places from the pecuniary point of view, that it would seem at first sight quite natural to think that every facility and even encouragement should be given to English practitioners to establish themselves abroad. But foreigners think otherwise, and point out that there are two sides to this question.

First of all, there is no lack of medical men abroad, and the congested condition of the professional ranks is, perhaps, more marked in the favoured districts than anywhere in England; therefore any arrangement made for the purpose of facilitating the migration of English medical men to the Continent would have, they contend, to be based on reciprocity. But what inducement is there for a foreign medical man to settle in England? Comparatively very little; and for ten Englishmen who would succeed in earning their living on the Riviera or the Engadine, there is, perhaps, one foreign doctor who could do the same in England. The advantages of reciprocity would, they argue, be almost entirely on one side.

Then it was asked, how are the foreign Governments to distinguish between our many registrable qualifications? Some of them are equal to the Continental ones, but many are inferior. Until we have adopted a uniform system of examinations it is declared to be useless to ask for reciprocity, and even then it is not in the least likely that it would be granted at present, whatever may have been the case in the past.

The fact is that an excellent opportunity has been lost. Whilst we were patching up an anomalous system of registration the foreign doctors were learning English and our countrymen were picking up French, so that the question of languages is much less important now than formerly. Experience is said to show that Englishmen travelling or residing abroad generally go to the best known man in the place, who is often not an Englishman, but who, in nine cases out of ten, speaks English sufficiently well. (*BMJ* 1896;i:1465.)