Regular Review

The lipoproteins: predictors, protectors, and pathogens

BARRY LEWIS

Low density lipoproteins (LDL) and high density lipoproteins (HDL) together carry more than 90% of the cholesterol in plasma. Independent but interacting, antagonistic but closely associated, they are the "Odd Couple" of plasma. The epidemiological aspects of their associations with coronary heart disease have been studied in depth; these associations are strong, predictive, and independent of other risk factors, as we expect of causal relations.¹

Concentrations of LDL cholesterol are directly related to and are predictive of the risk of coronary heart disease over a wide age range²; this underlies the association between coronary heart disease and serum cholesterol,³⁻⁶ for the latter reflects LDL concentrations. Mortality rates from coronary heart disease in different communities are directly and linearly correlated with serum concentrations of cholesterol and LDL cholesterol.^{6,7} HDL cholesterol concentrations are even more strongly predictive of the risk of coronary heart disease in most,^{2,8,9} but not all,¹⁰ studies, the relation being inverse; but, unlike LDL, HDL cholesterol concentrations do not correlate inversely with mortality rates from coronary heart disease in different countries.⁷

These associations have prompted extensive research on ways in which lipoproteins might influence the atherosclerotic and thrombotic processes. HDL and LDL particles are heterogeneous, and recent findings suggest that specific subclasses are selectively related to coronary heart disease. Of the three metabolically distinct subclasses of HDL,¹¹ most cholesterol is carried by HDL₃; concentrations of the small HDL₂ fraction are particularly low in coronary heart disease.^{12,13} HDL_c is characterised by the functionally important polypeptide apolipoprotein E.¹⁴ Earlier descriptions of LDL included several lipoproteins: these comprised intermediate density lipoprotein (IDL) and true LDL, the latter including up to four fractions¹⁵ often grouped into "light" and "heavy" subclasses. Very low density lipoprotein, rich in triglyceride, is partially converted, via IDL, to LDL within plasma¹⁶⁻¹⁹; the cascade is associated with loss of triglyceride and other components including apolipoprotein E. Heavy LDL may be the final product.²⁰ Concentrations of light LDL and IDL appear to be selectively increased in coronary heart disease.²¹⁻²³ The composition and size of light LDL are closer to those of the plasma LDL of monkeys fed an atherogenic diet²⁴; the latter causes greater accumulation of cholesterol in cultured cells than unmodified LDL.²⁵

LDL and IDL enter the arterial intima from plasma in man at rates directly related to their plasma concentrations^{26,27} and accumulate particularly in regions already atheromatous.²⁷ Endothelial injury greatly enhances this process, and may itself be caused experimentally by raised concentrations of plasma lipids.²⁸ The cholesterol of atheromatous lesions is

principally derived from plasma.²⁹ The interactions of LDL with cells of atheromatous plaques have been studied in some detail. Smooth muscle cells and fibroblasts have receptors that mediate uptake of LDL³⁰⁻³²; its cholesterol is released by lysosomal degradation. Macrophages lack these receptors but acquire lipoprotein cholesterol by other processes, including receptor mediated uptake of altered LDL. In contact with cultured endothelial cells, LDL is modified (acquiring increased negative charge), permitting macrophages to degrade it.³³ Much of the LDL extractable from atherosclerotic lesions is charge modified.³⁴

At the higher tissue concentrations resulting from pronounced hyperlipidaemia substantial amounts of LDL enter macrophages and other cells independently of receptors—by fluid phase endocytosis³⁵ and other processes. The foam cells of atheromatous plaques and xanthomas result from accumulation of lipoprotein lipids by these processes, and their degeneration leads to the extracellular lipid deposits of atherosclerosis.³⁶

Other mechanisms

These, then, are some of the mechanisms underlying the association between high concentrations of LDL, coronary heart disease, and atherosclerosis.³⁷ LDL may also contribute to other aspects of atherosclerosis by further actions. LDL, particularly light LDL in man and in animals fed cholesterol, is selectively mitogenic to arterial smooth muscle³⁸; studies on cultured cells suggest that LDL and other mitogens derived from platelets and endothelium might lead to the smooth muscle hyperplasia of atheromatous plaques. The response of platelets to aggregating agents is enhanced by their pre-incubation in media rich in cholesterol,³⁹ and platelets incubated with plasma from patients with high concentrations of LDL show increased aggregability.⁴⁰ Hence several mechanisms are known by which raised concentrations of LDL may promote the atherosclerotic and thrombotic processes underlying coronary heart disease—and biological plausibility is one of the criteria for explaining epidemiological associations in causal terms.¹

Most cells meet their requirements for cholesterol (for membrane synthesis) chiefly by uptake of plasma LDL^{41,42} and other lipoproteins; local synthesis of cholesterol is a further source in some tissues.^{43,44} Except for cells that synthesise lipoproteins (liver, small intestine) and those using cholesterol for steroid hormone synthesis cells in the steady state must possess other mechanisms for releasing cholesterol at rates equal to their uptake and synthesis. Cultured cells show a net efflux of cholesterol.^{45,46} As the liver is the only

organ that excretes cholesterol (and bile acids derived from it), mechanisms must exist for reverse, centripetal transport of cholesterol from peripheral cells to the liver; it was suggested in 1968 that HDL (in concert with the enzyme lecithin cholesterol acyltransferase that esterifies cholesterol in plasma) participates in centripetal transport.⁴⁷ Soon after, Miller and Miller⁴⁸ advanced the concept that this function of HDL, by favouring mobilisation of cholesterol from the arterial wall, might explain the inverse relation between HDL and risk of coronary heart disease. Many studies in vitro⁴⁹ (see references 49 and 54 for references) and some kinetic data⁵⁰ establish that HDL is an effective acceptor of cholesterol from extrahepatic cells; this is a function of HDL₃.⁵¹

which the enzyme hepatic lipase in hepatic sinusoids takes part,⁶⁰ HDL₃ could be regenerated. HDL₃ binds to receptors on arterial smooth muscle, a process that may play an important part in the "efflux" of cholesterol⁶¹; studies in vitro suggest that availability of HDL₃ and of HDL receptors influences the rate of efflux.⁴⁵ Cholesterol must also be returned to the liver via other lipoproteins; even if all HDL were cleared from plasma as intact particles by the liver, which is unlikely, the amount of cholesterol so delivered (up to 300 mg a day calculated from the known rates of HDL protein turnover) would be less than the turnover of cholesterol in extrahepatic tissues (some 500 mg a day derived from catabolism of LDL plus cholesterol synthesised locally). Of

TABLE I—Variables that influence HDL cholesterol concentrations in man

Sex	F > M	HDL ₂ lower, HDL ₃ higher in M
Age	Slight ↑ in F	
Diet:		
carbohydrates	↓	
vegetarian	↓	
high P:S* ratio	No change or ↓	No change AHA Prudent Diet, Oslo Heart Study diet, low fat high fibre diet; decrease on some experimental diets
fish protein	↑	
alcohol	↑	Even moderate intake; HDL ₂ and/or HDL ₃
soy lecithin	↑	Slight effect
Obesity	↓	Increases after weight reduction completed
Exercise	↑	HDL ₂ ↑, HDL ₃ ↓ or no change
Oral contraceptives	↑ no change or ↓	Depends on oestrogen dose (no effect with some low dose formulations) and progestogen type
Exog progestogen	↓	↓ with 19-nortestosterones (androgenic); not with medroxyprogesterone
Exog oestrogen	↓	
Exog androgen	↓	
Endog testosterone	Positive association	Positive or negative for free testosterone; negative for HDL ₃
Renal failure, nephrosis	↓	
Hypothyroidism	↓	
Diabetes	↓	↑ with treatment of IDDM; poor correlation with HbA _{1c}
Microsomal enzyme inducers	↓	
Smoking	↓	Reverses on stopping
Beta agonists	↑	
Beta blockers	↓ or no change	

*Polyunsaturated fatty acid:saturated fatty acid.

TABLE II—Variables that influence LDL cholesterol in man

Sex	M > F F > M	Premenstrual Postmenstrual
Age, children	↑	Rise from birth to age 6, then plateau
Adult age	↑	Rise to 6th decade, then decline (cross sectional data)
Saturated fat	↑	Palmitic, myristic acids; increased LDL synthesis
Polyunsaturated fat	↓	Especially linoleic acid; (ω-6); (eicosapentaenoic acid (ω-3) chiefly reduces VLDL)
Cholesterol	↑	Moderate effect; wide individual variation
Fibre	↓	Fruit, vegetable-derived fibre, oats; not wheat bran
Obesity	↑	Moderate ↑ with weight gain in adults; decrease with weight loss
Hypothyroidism	↑	↓ Catabolism via LDL receptors
Nephrosis	↑	
Cholestasis	↑	Abnormal LDL ("LPX")
Anorexia nervosa	↑	
Renal transplantation	↑	
Pregnancy	↑	
Familial hypercholesterolaemia	↑	Several mutant alleles causing reduced LDL receptor function (lack of receptor protein, absence of LDL recognition site, defects of post-translational processing)
Familial combined hyperlipidaemia	↑	Overproduction of LDL
β sitosterolaemia	↑ or N	
Hypertriglyceridaemia	↓	
Abeta and hypobeta lipoproteinaemia	Absent or ↓	

Moving cholesterol to the liver

Many pathways (not mutually exclusive) have been suggested for delivery of peripherally derived cholesterol to the liver. One proposes that the small cholesteryl ester poor HDL₃ particles,⁵² the initial acceptors, are in turn converted to HDL₂ and HDL_c as they acquire cholesterol and, in the latter step, apolipoprotein E⁵³⁻⁵⁵; HDL_c is then rapidly taken up by hepatocytes after binding to two types of receptors that recognise its apolipoprotein E.^{54 56 57} Alternatively, the HDL₂ might release its unesterified cholesterol in the liver (suggested by some kinetic findings)^{58 59} and, perhaps by a process in

the 750 mg cholesterol synthesised daily by adult man,⁶² an unknown but probably small proportion is produced in tissues other than the liver and small intestine. Tissue cholesterol entering HDL is esterified by lecithin cholesterol acyltransferase⁴⁷ and transfers to very low density lipoprotein (VLDL) in plasma, a step requiring an exchange protein⁶³; VLDL is converted within plasma to IDL, which is cleared protein A-I, is also synthesised in many tissues.⁶⁵

A fourth hypothesis is based on the finding that the efflux of cholesterol from macrophages is accompanied by secretion of apolipoprotein E⁶⁴; the increased efflux from cells pre-loaded with cholesterol is associated with enhanced production

of apolipoprotein E. Hence HDL_c may be assembled from components synthesised peripherally, at a rate dependent on efflux of cholesterol. Another component of HDL, apolipoprotein A-I, is also synthesised in many tissues.⁶⁵

All of these possibilities implicate HDL but all are more complex than the original fertile concept that HDL is the mediator of reverse transport of cholesterol; the epidemiology of HDL in coronary heart disease has been illuminated but not explained by this profusion of cell biology. If the last hypothesis is true it follows that high concentrations of HDL can be a consequence rather than a precondition of enhanced reverse transport.

HDL has other effects potentially relevant to atherosclerosis. HDL_c reduces uptake of LDL by cells, by competing for the LDL receptor,⁶⁶ and HDL stimulates release of prostacyclin by porcine endothelium.⁶⁷

Clinical observations have illuminated the roles played by lipoproteins in coronary heart disease. The life long elevation of LDL concentrations in familial hypercholesterolaemia is associated with a severalfold excess risk of premature coronary heart disease.⁶⁸ Serial quantitative arteriographic observations of the atherosclerotic femoral arteries of patients with hyperlipidaemia have shown that progression of the disease is directly related to mean concentrations of LDL cholesterol.⁶⁹ Conversely coronary heart disease has been reported to be rare in a familial syndrome characterised by high HDL and low LDL concentrations.⁷⁰ The risk of coronary heart disease varies in the uncommon disorders in which HDL is low. In Tangier disease, in which normal HDL is absent (and LDL relatively low), the risk of coronary heart disease is only modestly raised⁷¹; and the low concentrations of HDL in fish eye disease,⁷² in familial deficiency of lecithin cholesterol acyltransferase,⁷³ in a family with the apolipoprotein A-I-Milano mutant,⁷⁴ and in familial hypertriglyceridaemia and lipoprotein lipase deficiency⁷⁵ seem not to be associated with excess risk of coronary heart disease. Severe coronary heart disease did, however, develop in a woman with deficiency of the HDL protein apolipoprotein A-I.⁷⁶ In four European populations with differing mortality rates from coronary heart disease concentrations of HDL cholesterol were closely similar while LDL and VLDL showed congruent differences.⁷ In several disparate communities strong positive correlations were found between total cholesterol and HDL cholesterol concentrations⁷⁷⁻⁷⁸; in many countries with little coronary heart disease, mean concentrations of LDL and HDL are probably low. This is compatible with the inverse relation between carbohydrate intake and concentrations of both LDL and HDL⁷⁹⁻⁸⁰ and suggests that when concentrations of LDL are low the relation between HDL and risk of coronary heart disease may be attenuated.

Raised concentrations of LDL are readily decreased by diet and drugs.⁷⁹ Substantial reduction of the serum cholesterol by diet clearly decreased the incidence of coronary heart disease in the Oslo trial⁸¹; similar results in early trials are impressive for their consistency,⁸² though their designs were in various ways inadequate. Trials of selective increase in HDL may not be feasible. In the Coronary Drug Project, however, men with coronary heart disease were treated with oestrogen to reduce the serum cholesterol concentrations.⁸³ The treatment is also likely to have increased their HDL cholesterol—yet recurrences of coronary heart disease were commoner than in the controls. As oestrogens might influence risk, for example, by enhancing thrombotic tendency, this does not exclude a protective role for HDL. One further intervention

is worthy of discussion. The drug probucol, which decreases both LDL cholesterol and HDL cholesterol, may lead to rapid regression of xanthomas in tendons⁸⁴ and skin⁸⁵—impressive examples of mobilisation of tissue cholesterol.

There is, in summary, evidence that raised concentrations of LDL play an important causal part in coronary heart disease, with consistent findings from epidemiological and clinical observations, trials, research on animals, and cellular biology. But the powerful inverse relation between HDL and coronary heart disease should not be interpreted in causal terms until there is better understanding of the parts played by HDL in processes pertinent to atherosclerosis.

Genetic factors

The interacting environmental and genetic factors determining plasma concentrations of these lipoproteins have been studied extensively.⁷⁵⁻⁸⁶ Apart from major monogenic disorders,⁷⁰⁻⁷⁵ genetic control of concentrations of HDL cholesterol and apoproteins in normal people has been shown in some (but not all) studies⁸⁶⁻⁸⁸; the low HDL cholesterol concentration of patients with coronary heart disease appears to be familial.⁸⁹⁻⁹⁰ HDL cholesterol is substantially higher in blacks than in whites.⁹¹ This may also be genetic and it is associated with lower mortality from coronary heart disease. Table I shows some of the many variables influencing HDL.⁴⁹⁻⁹²⁻⁹⁵ An increased HDL cholesterol concentration might reflect several metabolic events, of which enhanced reverse transport is only one. It might also result from increased hepatic secretion of HDL—the probable basis of the increase in HDL found with microsomal enzyme inducing drugs and with alcohol. A third basis is redistribution of cholesterol within plasma. During catabolism of VLDL and chylomicrons cholesterol transfers to HDL; the activity of the rate limiting enzyme for this process, lipoprotein lipase, is increased by exercise, insulin, and clofibrate and related drugs. Thus a rise in HDL cannot be equated with mobilisation of cholesterol from tissues; and measures that raise the HDL level, or diseases that lower it, have not been shown to alter the risk of coronary heart disease.

Polygenic inheritance may explain around one third of the variance of concentrations of LDL and total cholesterol in normal people; but estimates range from 0 to 60%.⁹⁶⁻⁹⁹ Mean LDL cholesterol is lower in blacks than whites.⁹¹ Table II shows some of the influences determining plasma concentrations of LDL.¹⁰⁰⁻¹⁰²

A main interest of twentieth century philosophy is the study of evidence.¹⁰³ In medical science this has clinical as well as intellectual rewards. The former explains some of the current concern with cholesterol metabolism and coronary heart disease; but the convergence of research from numerous disciplines has been a further satisfaction. A theoretical basis now exists for treating hyperlipidaemia due to high concentrations of LDL, and testable hypotheses may be formulated concerning HDL. Three decades of lipoprotein research have taken us some way towards a goal epitomised by the Rubens painting "Triumph of reason over inharmonious discord."

BARRY LEWIS

Professor of Chemical Pathology,
Honorary Consultant in Medicine, and
Director of Lipid Clinic,
St Thomas's Hospital Medical School,
London SE1 7EH

- 1 Hill AB. *Principles of medical statistics*. 8th ed. London: The Lancet, 1967:306-13.
- 2 Gordon T, Kannel WB, Castelli WB, Dawber TR. Lipoproteins, cardiovascular disease and death. The Framingham study. *Arch Intern Med* 1981;141:1128-31.
- 3 Kannel WB, Castelli WP, Gordon T. Cholesterol in the prediction of atherosclerotic disease. *Ann Intern Med* 1979;90:85-91.
- 4 Reid DM, Hamilton PJS, McCartney P, Rose G, Jarrett RJ, Keen H. Smoking and other risk factors for coronary heart-disease in British civil servants. *Lancet* 1976;ii:979-84.
- 5 Pooling Project Research Group. Relationship of blood pressure, serum cholesterol, smoking habit, relative weight and ECG abnormalities to incidence of major coronary events. *J Chronic Dis* 1978;31:201-306.
- 6 Keys A. *Seven countries*. Cambridge: Harvard University Press, 1980.
- 7 Lewis B, Chait A, Sigurdsson G, et al. Serum lipoproteins in four European communities. *Eur J Clin Invest* 1978;8:165-73.
- 8 Miller NE, Forde OH, Thelle DS, Mjos OD. The Tromso heart study. High-density lipoprotein and coronary heart disease: a prospective case-control study. *Lancet* 1977;ii:965-8.
- 9 Goldbourt U, Medalie JH. High density lipoprotein cholesterol and incidence of coronary heart disease. *Am J Epidemiol* 1979;109:296-308.
- 10 Wiklund O, Wilhelmsson L, Ehnfeldt D, Wedel H, Valek J, Gustafson A. Alpha-lipoprotein cholesterol concentration in relation to subsequent myocardial infarction in hypercholesterolemic men. *Atherosclerosis* 1980;37:47-53.
- 11 Anderson DW, Nichols AV, Pan SS, Lindgren FT. High density lipoprotein distribution. *Atherosclerosis* 1978;29:161-79.
- 12 Gofman JW, Young W. Ischemic heart disease, atherosclerosis and longevity. *Circulation* 1966;34:679-97.
- 13 Miller NE, Hammett F, Rao S, Coltart DJ, van Zeller H, Lewis B. Relationship of angiographically defined coronary artery disease to plasma lipoprotein subfractions and apolipoproteins. *Br Med J* 1981;282:1741-4.
- 14 Mahley RW. Apolipoprotein E and cholesterol metabolism. *Klin Wochenschr* 1983;61:225-32.
- 15 Krauss RM, Burke DJ. Identification of multiple subclasses of plasma low density lipoproteins in normal humans. *J Lipid Res* 1982;23:97-104.
- 16 Eisenberg S, Bilheimer DW, Levy RI, Lindgren FT. On the metabolic conversion of human very low density lipoprotein to low density lipoprotein. *Biochim Biophys Acta* 1973;326:361-77.
- 17 Sigurdsson G, Nicoll A, Lewis B. Conversion of very low density lipoprotein to low density lipoprotein. *J Clin Invest* 1975;56:1481-90.
- 18 Janus ED, Nicoll A, Wootton R, Turner PR, Magill PJ, Lewis B. Quantitative studies of very low density lipoprotein conversion to low density lipoprotein in normal controls and primary hyperlipidaemic states. *Eur J Clin Invest* 1980;10:149-59.
- 19 Turner PR, Miller NE, Cortese C, Hazzard W, Coltart J, Lewis B. Splanchnic metabolism of plasma apolipoprotein B. *J Clin Invest* 1981;67:1678-86.
- 20 Thompson GR, Teng B, Sniderman A. Metabolic conversion of "light" to "heavy" low density lipoprotein (LDL) in subjects with normal and increased plasma LDL-APO-B levels. [Abstract.] *Circulation* 1982;66, suppl 11:158.
- 21 Gofman JW, Lindgren FT, Jones HB, Lyon TP, Strisower B. Lipoproteins and atherosclerosis. *J Gerontol* 1951;6:105-19.
- 22 Rao SN, Williams T, Coltart J, Cortese C, Miller NE, Lewis B. Plasma low density lipoprotein subclasses in coronary and peripheral atherosclerosis. [Abstract.] *Eur J Clin Invest* 1983;13:II:18a.
- 23 Tatami R, Mabuchi H, Ueda K, et al. Intermediate-density lipoprotein and cholesterol-rich very low density lipoprotein in angiographically determined coronary artery disease. *Circulation* 1981;64:1174-84.
- 24 Rudel LL, Shah R, Greene DG. Study of the atherogenic dyslipoproteinemia induced by dietary cholesterol in rhesus monkeys (Macaca mulatta). *J Lipid Res* 1979;20:55-65.
- 25 St Clair RW, Greenspan P, Leigh M. Enhanced cholesterol delivery to cells by low density lipoproteins from hypercholesterolemic monkeys. *Arteriosclerosis* 1983;3:77-86.
- 26 Niehaus CE, Nicoll A, Wootton R, et al. Influence of lipid concentrations and age on transfer of plasma lipoprotein into human arterial intima. *Lancet* 1977;ii:469-71.
- 27 Nicoll A, Duffield RGM, Lewis B. Flux of lipoproteins into human arterial intima. *Atherosclerosis* 1981;39:229-42.
- 28 Ross R, Harker L. Hyperlipidemia and atherosclerosis. *Science* 1976;193:1094-8.
- 29 Zilvermit DB. Cholesterol flux in the atherosclerotic plaque. *Ann NY Acad Sci* 1968;149:710-24.
- 30 Goldstein JL, Brown MS. Binding and degradation of low density lipoprotein by cultured human fibroblasts. *J Biol Chem* 1974;249:5153-62.
- 31 Bierman EL, Albers JJ. Lipoprotein uptake by cultured human arterial smooth muscle cells. *Biochim Biophys Res Commun* 1975;68:198-202.
- 32 Brown MS, Kovanen PT, Goldstein JL. Regulation of plasma cholesterol by lipoprotein receptors. *Science* 1981;212:628-35.
- 33 Henriksen T, Mahoney EM, Steinberg D. Enhanced macrophage degradation of low density lipoprotein previously incubated with cultured endothelial cells: recognition by receptors for acetylated low density lipoproteins. *Proc Natl Acad Sci USA* 1981;78:6499-503.
- 34 Hoff HF, Gaubatz JW. Isolation, purification, and characterization of a lipoprotein containing apo B from the human aorta. *Atherosclerosis* 1982;42:273-97.
- 35 Steinberg D. Metabolism of lipoproteins at the cellular level in relation to atherogenesis. In: Miller NE, Lewis B, eds. *Lipoproteins, atherosclerosis and coronary heart disease*. Amsterdam: Elsevier/North Holland, 1981.
- 36 Small DM. Cellular mechanisms for lipid deposition in atherosclerosis. *N Engl J Med* 1977;297:1st of 2 parts 873-7, 2nd of 2 parts 924-9.
- 37 Solberg LA, Strong JP. Risk factors and atherosclerotic lesions. A review of autopsy studies. *Arteriosclerosis* 1983;3:187-98.
- 38 Fless GM, Kirchhausen T, Fischer-Dzoga K, Wissler RW, Scanu AM. Serum low density lipoproteins with mitogenic effect on cultured aortic smooth muscle cells. *Atherosclerosis* 1982;41:171-83.
- 39 Shattil SL, Anayo-Galindo R, Bennett J, Colman RW, Cooper RA. Platelet hypersensitivity induced by cholesterol incorporation. *J Clin Invest* 1975;55:636-44.
- 40 Aviram M, Brook GJ. The effect of human plasma on platelet function in familial hypercholesterolemia. *Thromb Res* 1982;26:101-9.
- 41 Brown MS, Faust JR, Goldstein JL. Role of the low density lipoprotein receptor in regulating the content of free and esterified cholesterol in human fibroblasts. *J Clin Invest* 1975;55:783-93.
- 42 Brown MS, Goldstein JL. Multivalent feedback regulation of HMGCoA reductase, a control mechanism regulating isoprenoid synthesis and cell growth. *J Lipid Res* 1980;21:505-17.
- 43 Turley SD, Andersen JM, Dietschy JM. Rates of sterol synthesis and uptake in the major organs of the rat in vivo. *J Lipid Res* 1981;22:551-69.
- 44 Knight BL, Patel DD, Soutar AK. The regulation of 3-hydroxy-3-methylglutaryl-CoA reductase activity, cholesterol esterification and the expression of low-density lipoprotein receptors in cultured monocyte-derived macrophages. *Biochem J* 1983;210:523-32.
- 45 Fielding CJ, Fielding PE. Cholesterol transport between cells and body fluids. *Med Clin North Am* 1982;66:363-73.
- 46 Fielding PE, Fielding CJ, Havel RJ, Kane JP, Tun P. Cholesterol net transport, esterification and transfer in human hyperlipidemic plasma. *J Clin Invest* 1983;71:449-60.
- 47 Glimst J. The plasma lecithin-cholesterol acyltransferase reaction. *J Lipid Res* 1968;9:155-67.
- 48 Miller GJ, Miller NE. Plasma high density lipoprotein concentration and development of ischaemic heart disease. *Lancet* 1975;i:16-9.
- 49 Nicoll A, Miller NE, Lewis B. High-density lipoprotein metabolism. *Adv Lipid Res* 1980;17:53-106.
- 50 Nestel PJ, Miller NE. Mobilization of adipose tissue cholesterol in high density lipoprotein during weight reduction in man. In: Gotto AM, Miller NE, Oliver MF, eds. *High density lipoproteins and atherosclerosis*. Amsterdam: Elsevier/North Holland, 1978:51-4.
- 51 Oram JF, Albers JJ, Cheung MC, Bierman EL. The effects of subfractions of high density lipoprotein on cholesterol efflux from cultured fibroblasts. *J Biol Chem* 1981;256:8348-56.
- 52 Rothblat GH, Phillips MC. Mechanism of cholesterol efflux from cells. *J Biol Chem* 1982;257:375-402.
- 53 Schmitz G, Assman G. Isolation of human serum HDL₁ by zonal ultracentrifugation. *J Lipid Res* 1982;23:93-100.
- 54 Mahley RW. Atherogenic hyperlipoproteinemia. *Med Clin North Am* 1982;66:375-402.
- 55 Gordon V, Innerarity TL, Mahley RW. Formation of cholesterol- and apoprotein E-enriched high density lipoprotein in vitro. *J Biol Chem* 1983;258:6202-12.
- 56 Quarfordt S, Hanks J, Jones RS, Shellenberg T. The uptake of high density lipoprotein cholesteryl ester in the perfused rat liver. *J Biol Chem* 1980;255:2934-7.
- 57 Hui DY, Innerarity TL, Mahley RW. Lipoprotein binding to canine hepatic membranes. *J Biol Chem* 1981;256:5646-55.
- 58 Halloran LG, Schwartz CC, Vlahevic ZR, Nisman RM, Swell L. Evidence for high-density lipoprotein-free cholesterol as the primary precursor for bile-acid synthesis in man. *Surgery* 1978;84:1-6.
- 59 Price S, Cortese C, Miller NE, Lewis B. In preparation.
- 60 Jansen H, van Tol A, Hulsmann WC. On the metabolic function of heparin-releasable liver lipase. *Biochem Biophys Res Commun* 1980;92:53-9.
- 61 Biesbroeck R, Oram JF, Albers JJ, Bierman EL. Specific high-affinity binding of high density lipoproteins to cultured human skin fibroblasts and arterial smooth muscle cells. *J Clin Invest* 1983;71:525-39.
- 62 Myant NB. *The biology of cholesterol and related steroids*. London: Heinemann, 1981:464.
- 63 Barter PJ, Jones ME. Kinetic studies of the transfer of esterified cholesterol between human plasma low and high density lipoproteins. *J Lipid Res* 1980;21:238-49.
- 64 Basu SK, Brown MS, Ho YK, Havel RJ, Goldstein JL. Mouse macrophages synthesize and secrete a protein resembling apolipoprotein E. *Proc Natl Acad Sci USA* 1981;78:7545-9.
- 65 Blue M-L, Ostapchuk P, Gordon JS, Williams DL. Synthesis of apolipoprotein AI by peripheral tissues of the rooster. *J Biol Chem* 1982;257:1115-9.
- 66 Miller NE, Weinstein DB, Carew TE, Koschinsky T, Steinberg D. Interaction between high density and low density lipoproteins during uptake and degradation by cultured human fibroblasts. *J Clin Invest* 1977;60:78-89.
- 67 Fleischer LN, Tall AR, Witte LD, Miller RW, Cannon PJ. Stimulation of arterial endothelial cell prostaglandin synthesis by high density lipoproteins. [Abstract.] *Circulation* 1981;64, suppl IV:216.
- 68 Stone NJ, Levy RI, Fredrickson DS, Verter J. Coronary artery disease in 116 kindred with familial type II hyperlipoproteinemia. *Circulation* 1974;49:476-88.
- 69 Duffield RGM, Lewis B, Miller NE, Jamieson CW, Brunt JNH, Colchester ACF. Treatment of hyperlipidaemia retards progression of symptomatic femoral atherosclerosis. *Lancet* 1983;ii:639.
- 70 Glueck CJ, Gartside P, Fallat RW, Sielski J, Steiner PM. Longevity syndromes. Familial hypobeta- and familial hyperalpha-lipoproteinemia. *J Lab Clin Med* 1976;88:941-57.
- 71 Schaefer EJ, Zech LA, Schwartz DE, Brewer JB. Coronary heart disease prevalence and other clinical features in familial high-density lipoprotein deficiency (Tangier disease). *Ann Intern Med* 1980;93:261-6.
- 72 Carlson LA. Fish eye disease: a new familial condition with massive corneal opacities and dyslipoproteinemia. *Eur J Clin Invest* 1982;12:41-53.
- 73 Gione E. Familial lecithin: cholesterol acyltransferase deficiency—a clinical survey. *Scand J Clin Lab Invest* 1974;33, suppl 137:73-82.
- 74 Franchescini G, Sirtori CR, Capurso A, Weisgraber KH, Mahley RW. A-I-Milano apoprotein. Decreased high density lipoprotein cholesterol level with significant lipoprotein modifications and without clinical atherosclerosis in an Italian family. *J Clin Invest* 1980;66:892-900.
- 75 Lewis B. *The hyperlipidaemias: clinical and laboratory practice*. Oxford: Blackwell Scientific, 1976.
- 76 Schaefer EJ, Heaton WH, Wetzel MG, Brewer HB. Plasma apolipoprotein A-I absence associated with marked reduction of high density lipoproteins and premature coronary artery disease. *Arteriosclerosis* 1982;2:16-26.
- 77 Knudsen JT, West CE. HDL cholesterol in men from thirteen centres. *Lancet* 1981;ii:367-8.
- 78 Knudsen JT, Hermus RJ, Hautvast JGAJ. Serum total and high density lipoprotein cholesterol concentrations in rural and urban boys from 16 countries. *Arteriosclerosis* 1980;36:529-37.
- 79 Schonfeld G, Weidman SW, Witztum JL, Bowen RM. Alterations in levels and interrelations of plasma apolipoproteins induced by diet. *Metabolism* 1976;25:261-75.
- 80 Tyroler HA, ed. Epidemiology of plasma high-density lipoprotein cholesterol levels. *Circulation* 1980;62:1-136. (American Heart Association monograph, No 73.)
- 81 Hjermstad I, Velve Byre K, Holme I, Leren P. Effect of diet and smoking intervention on the incidence of coronary heart disease. *Lancet* 1981;ii:1303-10.
- 82 Mann JI, Marr JW. Coronary heart disease prevention. Trials of diets to control hyperlipidaemia. In: Miller NE, Lewis B, eds. *Lipoproteins, atherosclerosis and coronary heart disease*. Amsterdam: Elsevier North Holland, 1981:197-210.
- 83 Coronary Drug Project Group. The coronary drug project—findings leading to discontinuation of the 2-5 mg/d estrogen group. *JAMA* 1973;226:652-7.
- 84 Yamamoto A, Matsuzawa Y, Kishino B-I, Hayashi R, Hirobe K, Kikkawa T. Effect of probucol on homozygous cases of familial hypercholesterolemia. *Atherosclerosis* 1983;48:157-66.
- 85 Beaumont JL, Jacotot B, Buxtorf JC, Silvestre M, Beaumont V. Effects of probucol on the cholesterol content of skin in type II hyperlipoproteinemias. *Artery* 1982;10:71-87.
- 86 Segal P, Rifkind BM, Schull WJ. Genetic factors in lipoprotein variation. *Epidemiol Rev* 1982;4:137-60.
- 87 Berg K. Genetic influence on variation in serum high density lipoproteins. In: Gotto AM, Miller NE, Oliver MF, eds. *High density lipoproteins and atherosclerosis*. Amsterdam: Elsevier North Holland, 1978:207-11.
- 88 Heiberg A. The heritability of serum lipoprotein and lipid concentrations. A twin study. *Clin Genet* 1974;6:307-16.
- 89 Micheli H, Pometta D, Jornot C, Scherrer J-R. High density lipoprotein cholesterol in male relatives of patients with coronary heart disease. *Atherosclerosis* 1979;32:269-76.
- 90 Nupuf MS, Sutherland WHF. High density lipoprotein levels in children of young men with ischaemic heart disease. *Atherosclerosis* 1979;33:365-70.
- 91 Tyroler HA, Heiss G, Schonfeld G, Cooper G, Heyden S, Hames CG. Apolipoprotein A-I, A-II, and C-II in black and white residents of Evans County. *Circulation* 1980;62:249-54.
- 92 Krauss RM. Regulation of high density lipoprotein levels. *Med Clin North Am* 1982;66:403-30.
- 93 Goldberg AP, Hardier HR, Patsch W, et al. Racial differences in plasma high-density lipoproteins in patients receiving hemodialysis. *N Engl J Med* 1983;308:1245-52.
- 94 Heller RF, Miller NE, Lewis B, Wheeler M. Some hormonal determinants of lipoprotein levels in men. [Abstract.] *Eur Heart J* 1982;3B:3.
- 95 Semmens J, Rouse I, Beilin LJ, Masera JRL. Relationship of plasma HDL-cholesterol to testosterone, estradiol and sex-hormone-binding globulin levels in men and women. *Metabolism* 1983;32:428-32.
- 96 Osborne RH, Adlersberg D, DeGeorge FV, Wang C. Serum lipids hereditary and environment: a study of adult twins. *Am J Med* 1959;26:54-9.
- 97 Feinleib M, Garrison RJ, Fabritz R. The NHLBI twin study of cardiovascular disease risk factors: methodology and summary of results. *Am J Epidemiol* 1977;106:284-95.
- 98 Christian JC, Cheung SW, Kang K, Harmath FP, Huntzinger DJ, Powell RC. Variance of plasma free and esterified cholesterol in adult twins. *Am J Hum Genet* 1976;28:174-8.
- 99 Robertson FW, Cumming AM. Genetic and environmental variation in serum lipoproteins in relation to coronary heart disease. *J Med Genet* 1979;16:85-100.
- 100 Goldstein JL, Hazzard WR, Schrott HG, Bierman EL, Motulsky AG. Genetics of hyperlipidemia in coronary heart disease. *Trans Assoc Am Physicians* 1972;85:120-38.
- 101 Tollehaug H, Goldstein JL, Schneider WJ, Brown MS. Post-translational processing of the LDL receptor and its genetic disruption in familial hypercholesterolemia. *Cell* 1982;30:715-24.
- 102 Levy RI, Rifkind BM, Dennis BH, Ernst ND, eds. *Nutrition, lipids, and coronary heart disease*. New York: Raven Press, 1979:89-246.
- 103 Ayer A. *Twentieth century philosophy*. London: Weidenfeld and Nicolson, 1982.