LBL-32776
UC-000

Lawrence Berkeley Laboratory
UNIVERSITY OF CALIFORNIA

Presented at the Therapeutic Approaches to the Diabetic Hypertensive Patient: Role of Risk Factors on Morbidity and Mortality, New York, NY, May 9, 1992, and to be published in the Proceedings

Lipids: Part of the Tangled Web

R.M. Krauss

August 1992

Donner Laboratory

Biology & Medicine Division

Prepared for the U.S. Department of Energy under Contract Number DE-AC03-76SF00098

DISCLAIMER

LBL-32776

Lipids: Part of the Tangled Web

Ronald M. Krauss, M.D.

Life Sciences Division
Lawrence Berkeley Laboratory
University of California
Berkeley, California 94720

This work was supported by National Institute of Health Program Project Grant HL-18574 from the National Heart, Lung, Blood Institute and was conducted at the Lawrence Berkeley Laboratory through the U.S. Department of Energy under Contract No. DE-AC03-76SF00098.

Proofs and reprints should be sent to:

Ronald M. Krauss, M.D.
Lawrence Berkeley Laboratory
University of California
Donner Laboratory, Room 465
Berkeley, CA 94720

Phone Number:
(510) 486-4994

FAX Number:
(510) 486-4750

Lipids: Part of the Tangled Web

Ronald M. Krauss, M.D.
Life Sciences Division
Lawrence Berkeley Laboratory
University of California
Berkeley, California 94720

Involvement of lipoproteins in the development of atherosclerosis

Hypercholesterolemia, and particularly an increase in levels of plasma low density lipoproteins (LDL), is a well documented epidemiologic risk factor for the development of atherosclerotic cardiovascular disease. Further, it has been demonstrated that LDL accumulate in arterial lesions ranging from the preatherosclerotic fatty streak to the mature fibrous plaque. More recently, there has been an impressive body of evidence suggesting that a major factor leading to the atherogenicity of LDL is oxidative modification [1]. LDL lipid oxidation products have been found to be cytotoxic to endothelial cells, and to trigger a number of tissue inflammatory responses, including recruitment and retention of white blood cells, and stimulation of cellular growth factors. Furthermore, oxidation of LDL leads to modification of its principal protein component, apoB-100, such that it is no longer recognized by high-affinity LDL receptors, but instead becomes a ligand for scavenger receptors on tissue macrophages. Unlike LDL receptors, scavenger receptors are not regulated by cholesterol levels, and thus excessive tissue accumulation of LDL can lead to engorgement of macrophages and appearance of lipid droplets that give rise to the typical apearance of "foam cells" which are prominent components of the fatty streak. Oxidized LDL may also potentiate processes that lead to formation of the mature atherosclerotic plaque, including stimulation of smooth muscle cell proliferation, and alteration in platelet function.

More recently, reduced levels of high-density lipoproteins (HDL) have been shown to be yet another powerful independent predictor of risk for atherosclerosis [2]. While the precise mechanism for the inverse relationship of HDL to cardiovascular disease risk has not been determined, a number of possibilities have been suggested. The ability of HDL to mediate efflux of cellular cholesterol has led to the notion that HDL may promote "reverse cholesterol transport" from arterial tissues to the liver for uptake and excretion [3]. Other studies have indicated that HDL concentrations are inversely related to levels and rates of plasma clearance of triglyceride-rich lipoproteins [4], and thus low HDL levels may reflect transient accumulation of potentially atherogenic remnants of these lipoproteins. Finally, there has been recent evidence that HDL may reduce the accumulation of oxidized lipids in LDL. Whatever mechanisms may be involved, the potential for HDL to directly influence atherosclerosis risk has been demonstrated recently in animal models, most convincingly in transgenic mice in which high level expression of human apoA-I, the major HDL protein, leads to increased plasma HDL concentrations and marked reduction in the extent of aortic fatty streaks [5].

Interrelated coronary disease risk factors: the atherogenic lipoprotein phenotype

Until recently, a statistical lack of correlation between levels of LDL- and HDL-cholesterol in plasma has led to the conclusion that increased levels of LDL and reduced levels of HDL contribute independently to the risk of developing atherosclerotic cardiovascular disease . However, we and others have demonstrated that LDL are heterogeneous and comprise a number of discrete subclasses which differ in particle size and density [6]. Levels of the largest and most buoyant LDL (LDL-I), measured by ultracentrifugal techniques, are strongly positively correlated with HDL cholesterol concentrations, while levels of smaller, denser LDL (principally LDL-III) are inversely correlated with HDL. Furthermore, LDL-III levels are associated with relatively

increased concentrations of triglyceride-rich lipoproteins, including relatively atherogenic lipolytic "remnants" measured as intermediate density lipoproteins (IDL). Increased levels of small, dense LDL have been associated with increased risk of coronary artery disease, and changes in levels of these particles have been related to disease progression as assessed by coronary angiography [6,7]. There has been recent evidence that smaller LDL particles have properties which lead to reduced uptake by high-affinity LDL receptors, increased binding to arterial wall proteoglycans, and increased susceptibility to copper-induced oxidation in vitro [7]. Therefore, high levels of small, dense LDL may contribute to increased coronary disease risk directly, and also by virtue of highly intercorrelated changes in other lipoprotein classes.

Analysis of LDL subclasses by non-denaturing gradient gel electrophoresis has led to the identification of a subclass pattern characterized by predominance of small LDL, designated LDL subclass pattern B (Figure 1) [8]. The prevalence of pattern B in the general population is approximately 25%, but varies as a function of age and gender, being relatively uncommon in children and in premenopausal women. The remainder of the population has a predominance of larger LDL (pattern A) or an intermediate pattern (approximately 10-15% of subjects).

Two large family studies have provided evidence for heritability of pattern B. The results of complex segregation analysis in these families are consistent with a major gene underlying the expression of pattern B, with a prevalence of 0.25, and autosomal dominant or co-dominant inheritance [8]. As suggested by the prevalence data, penetrance of the genetic trait is maximal in men older than age 20 and in postmenopaual women, suggesting that hormonal or developmental factors have important influences on the production of small, dense LDL. Recently, pattern B has been linked to a genetic locus at or near the LDL receptor on the short arm of chromosome 19p, further strengthening the evidence for a genetic determinant of this phenotype [9].

Consistent with the relationships of small, dense LDL to levels of other lipoproteins, individuals with LDL subclass pattern B have been shown to have significantly higher levels of triglyceride-rich lipoproteins and apoprotein B, and lower levels of HDL, than subjects with pattern A [8](Table 1). The relationship of LDL size to triglyceride levels is particularly strong. Pattern B is highly prevalent at levels of triglyceride greater than 120-140 mg/dl, and reductions to below this range with nicotinic acid have been shown to result in suppression of the pattern B phenotype.

In a case-control study, LDL subclass pattern B has been associated with up to a three-fold increased risk of myocardial infarction [10]. Statistical analyses have indicated that the risk associated with small, dense LDL cannot be dissociated from concomitant changes in other lipids and lipoproteins, particularly plasma triglyceride. Thus, the lipoprotein profile associated with pattern B has been designated the "atherogenic lipoprotein phenotype" [8].

Small, dense LDL and the insulin resistance syndrome

The lipoprotein variations in this phenotype are similar to those which have been described for patients with both non-insulin dependent diabetes mellitus (NIDDM) and insulin resistance. Recent studies in a population of 691 women have indicated that approximately 1/3 of subjects with impaired glucose tolerance and 2/3 of subjects with NIDDM have either pattern B or the intermediate subclass pattern, and that in non-diabetic subjects, pattern B is strongly associated with elevated fasting and two-hour post-load insulin levels [11]. Furthermore, consistent with other studies of the insulin resistance syndrome, pattern B is associated with increased blood pressure [11]. While there is a significant tendency for small, dense LDL to be associated with increased relative body weight and upper body obesity [12], the relationships of LDL particle size to insulin resistance and elevations in blood pressure are independent of body mass index and waist to hip girth ratio [11].

These findings indicate that LDL subclass pattern B is an integral part of the "tangled web" of interrelated coronary disease risk factors associated with insulin resistance [13]. It may be that the pathologic features of this lipoprotein profile, including the relative atherogenicity of small, dense LDL and IDL, contribute importantly to the increased risk of cardiovascular disease in subjects with insulin resistance and hypertension. Furthermore, pattern B serves as a marker for a common genetic trait which may underlie a substantial portion of the familial predisposition to coronary artery disease in the general population. Studies of hormonal, dietary, and pharmacologic influences on expression of this atherogenic phenotype should lead to more effective identification and management of high-risk individuals, and improved approaches to disease prevention in high-risk families.

Acknowledgements

This work was supported by National Institute of Health Program Project Grant HL-18574 from the National Heart, Lung, Blood Institute and was conducted at the Lawrence Berkeley Laboratory through the U.S. Department of Energy under Contract No. DE-AC03-76SF00098.

REFERENCES

1 Witztum JL. The role of oxidized LDL in atherosclerosis. Adv Exp Med Biol 1991;285:353-365.

2 Miller NE. Associations of high-density lipoprotein subclasses and apolipoproteins with ischemic heart disease and coronary atherosclerosis. Am Heart J 1987;113:589-597.

3 Gwynne JT. High-density lipoprotein cholesterol levels as a marker of reverse cholesterol transport. Am J Cardiol 1989;64:10G-17G.

4 Patsch JR, Prasad S, Gotto AM Jr., Patsch W. High density lipoprotein2: Relationship of the plasma levels of this lipoprotein species to its composition, to the magnitude of postprandial lipemia, and to the activities of lipoprotein lipase and hepatic lipase. J Clin Invest 1987;80:341-347.

5 Rubin EM, Krauss RM, Spangler EA, Verstuyft JG, Clift SM. Inhibition of early atherogenesis in transgenic mice by human apolipoprotein AI. Nature 1991;353:265-267.

6 Krauss RM. Relationship of intermediate and low-density lipoprotein subspecies to risk of coronary artery disease. Am Heart J 1987;113:578-582.

7 Krauss RM. Low-density lipoprotein subclasses and risk of coronary artery disease. Curr Opin Lipidol 1991;2:248-252.

8 Austin MA, King MC, Vranizan KM, Krauss RM. Atherogenic lipoprotein phenotype: A proposed genetic marker for coronary heart disease risk. Circulation 1990;82:495-506.

9 Nishina PM, Johnson JP, Naggert JK, Krauss RM. Linkage of atherogenic lipoprotein phenotype to the low density lipoprotein receptor locus on the short arm of chromosome 19. Proc Natl Acad Sci 1992;89:708-712.

10 Austin MA, Breslow JL, Hennekens CH, Buring JE, Willett WC, Krauss RM. Low-density lipoprotein subclass patterns and risk of myocardial infarction. JAMA 1988;260:1917-1921.

11 Selby JV, Austin MA, Newman B, Mayer EJ, Krauss RM. LDL subclass phenotypes, insulin, hypertension and obesity in women. Circulation 1991;84:II-547.

12 Terry RB, Wood PD, Haskell WL, Stefanick ML, Krauss RM. Regional adiposity patterns in relation to lipids, lipoprotein cholesterol, and lipoprotein subfraction mass in men. J Clin Endocrinol Metab 1989;68:191-199.

13 Krauss RM. The tangled web of coronary risk factors. Am J Med 1991;90 (Suppl 2A):38S-41S.

Table 1. Adjusted Plasma Lipid, Apolipoprotein, Lipoprotein Mass Levels by Atherogenic Lipoprotein Phenotype

	ALP phenotype A		ALP phenotype B	
	n	Mean±SD	*n*	Mean±SD
Total cholesterol*	208	177±37	93	197±40
Triglyceride†*	208	69±26	93	141±79
VLDL cholesterol†*	208	14±5	93	28±16
LDL cholesterol‡	208	116±35	92	126±36
HDL cholesterol*	208	46±15	92	37±14
Apo A-I‡	206	131±29	92	122±31
Apo B*	206	76±34	93	98±36
VLDL mass†*	151	18±31	60	111±68
LDL mass				
Large*	151	119±38	60	87±34
Small*	151	164±55	60	221±64
IDL mass*	151	20±14	60	38±17
HDL_2 mass*	151	55±44	60	13±26
HDL_3 mass	151	189±47	60	180±59

ALP, atherogenic lipoprotein phenotype; VLDL, very low density lipoprotein; LDL, low density lipoprotein; HDL, high density lipoprotein; IDL, intermediate density lipoprotein

Values are give as mean±SD mg/dl.

Mean values are adjusted to 50-year-old men with analysis of covariance and adjusting for age, gender, and relative weight.

* $p<0.001$, ‡ $p<0.05$, for difference in means between phenotype A and phenotype B subjects based on analysis of covariance.

† Log_{10} transformation used in calculations; reported values based on antilogs.

Originally published in Circulation 82/2; 498 (1990).

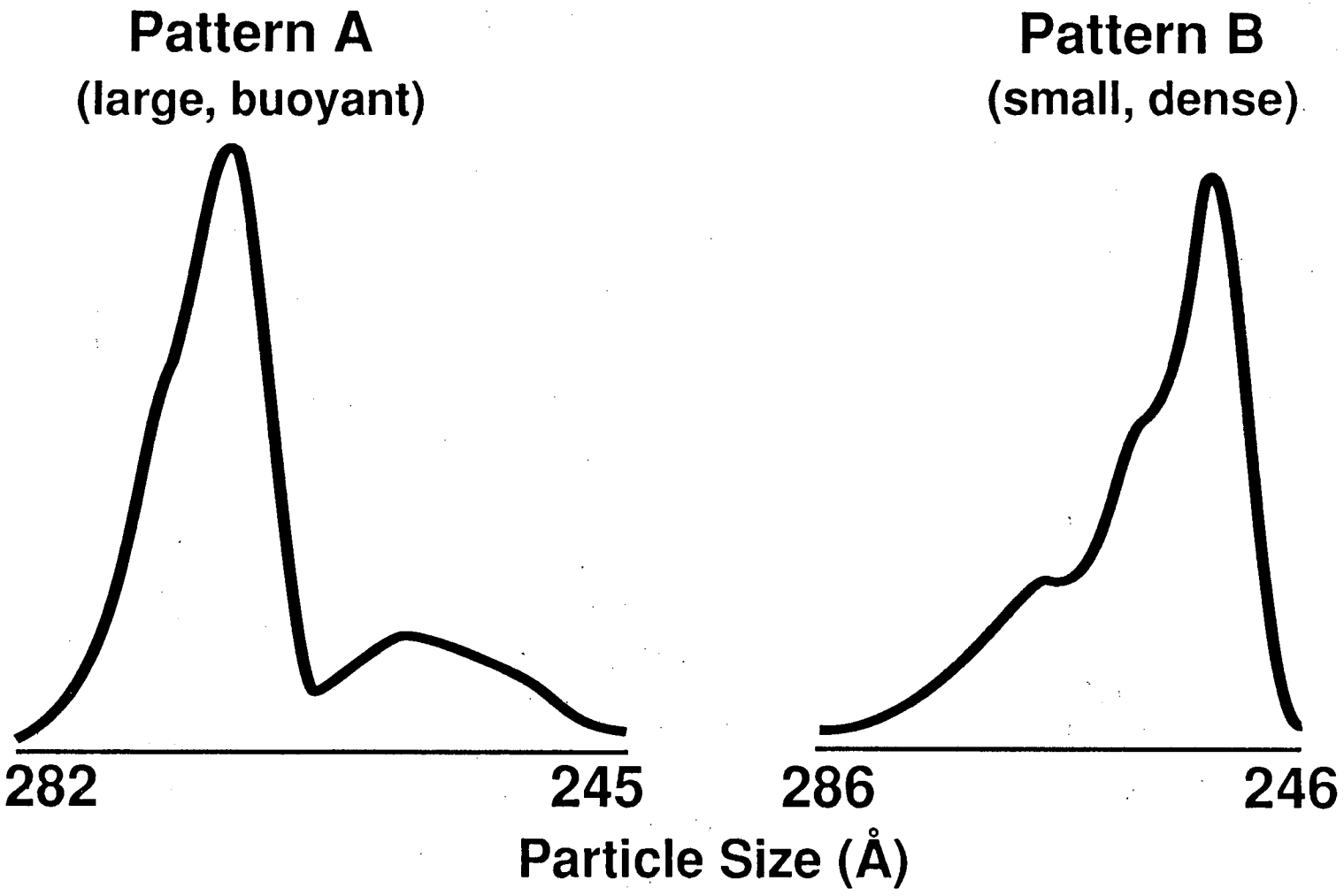
Pattern A
(large, buoyant)
Pattern B
(small, dense)
282
245
286
246
Particle Size (Å)

FIGURE LEGEND

Figure 1. Analysis of low density lipoprotein subclasses by non-denaturing 2-16% gradient gel electrophoresis.

LAWRENCE BERKELEY LABORATORY
UNIVERSITY OF CALIFORNIA
TECHNICAL INFORMATION DEPARTMENT
BERKELEY, CALIFORNIA 94720