The Concept of Apolipoprotein-defined Lipoprotein Families and Its Clinical Significance

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Classification of plasma lipoproteins on the basis of apolipoprotein (apo) composition recognizes two lipoprotein (Lp) classes, one of which is characterized by apoA-I and the other by apoB as major protein constituents. The former lipoprotein class consists of three major subclasses referred to (according to their apolipoprotein constituents) as Lp-A-I, Lp-A-I:A-II, and Lp-A-II, and the latter one of five subclasses called Lp-B, Lp-B:E, Lp-B:C, Lp-B:C:E, and Lp-A-II:B:C:D:E. As polydisperse systems of particles, the apoA-I–containing lipoproteins overlap in high-density segments and apoB-containing lipoproteins in low-density segments of the density gradient. Each subclass is characterized by a specific chemical composition and metabolic property. Normolipidemia and dyslipoproteinemias are characterized by quantitative rather than qualitative differences in the levels of apoA- and apoB-containing subclasses. Furthermore, apoA-containing subclasses seem to differ with respect to their relative antiatherogenic capacities, and apoB-containing subclasses regarding their relative atherogenic potentials. Whereas Lp-A-I may have a greater antiatherogenic capacity than other apoA-containing subclasses, the cholesterol-enriched Lp-B:C appears to be the most atherogenic subclass among apoB-containing lipoprotein families. The use of pharmacologic and/or dietary interventions to treat dyslipoproteinemias has already shown that these therapeutic modalities may affect selectively individual apolipoprotein-defined lipoproteins, and thus allow the selection of individualized treatments targeted at decreasing harmful and/or increasing beneficial lipoprotein subclasses.

Introduction

The traditional classification of plasma lipoproteins based on physical properties such as electric charge, size, and density has provided an indispensable conceptual framework for systematic studies of their physical, chemical, metabolic, and pathophysiologic characteristics [1-4]. However, the compositional and metabolic heterogeneity of major lipoprotein density classes necessitated the introduction of an alternative approach to definition and classification of plasma lipoprotein system [3,5–10]. Discovery and characterization of a number of specific lipid-binding proteins, the apolipoproteins, led to the recognition of their pivotal roles as determinants of the structural integrity and metabolic and functional properties of lipoproteins [9–12]. Furthermore, as chemically unique constituents, apolipoproteins have also been recognized as the most suitable markers for identifying, differentiating, and classifying lipoproteins [9–11]. The introduction and use of immunologic techniques for the quantification and distribution of apolipoproteins among various lipoproteins [10,11,13–16] indicated that subfractions of very low-density (VLDL), intermediate-density (IDL), low-density (LDL), and high-density (HDL) lipoproteins do not have the same apolipoprotein composition. This apolipoprotein heterogeneity was interpreted as an indication that each of these major lipoprotein density classes consists of several discrete lipoprotein subclasses characterized by similar density properties but different apolipoprotein composition [9,11]. Although the identification of discrete lipoprotein species added another dimension to the complexity of plasma lipoproteins, it also disclosed apolipoprotein-defined lipoprotein families as the fundamental chemical and functional entities of lipid transport system. The purpose of this review is to present the conceptual aspects of a classification system of plasma lipoproteins based on apolipoprotein composition as the criterion for identifying and differentiating discrete lipoprotein families, and its usefulness in selecting and monitoring pharmacologic and/or dietary interventions of dyslipoproteinemic states.

The Concept of Apolipoprotein-defined Lipoprotein Families

Chemistry and metabolism of lipoprotein families

According to the lipoprotein family concept, there are two major classes of lipoprotein families, one of which is characterized by apolipoprotein A (apoA-I and apoA-II) and the

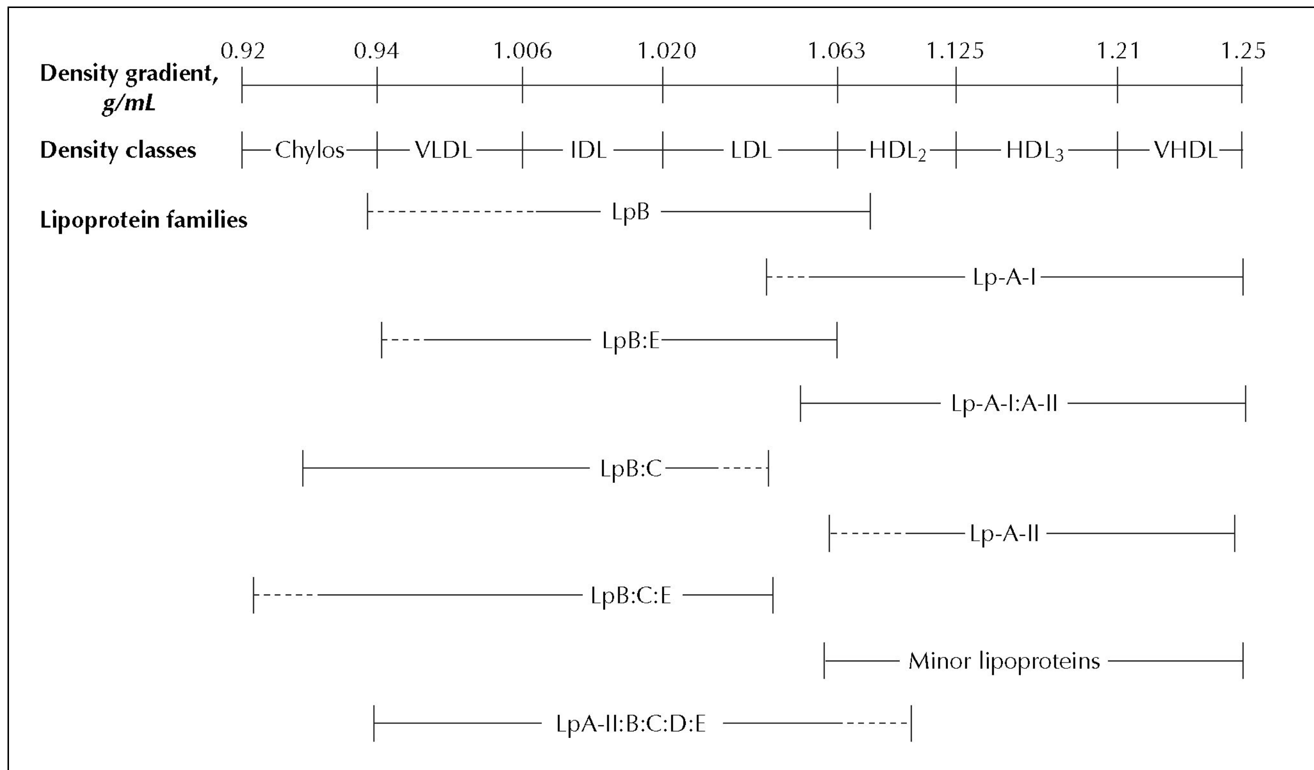

Figure 1. The relationship of individual apolipoprotein A (apoA)- and apoB-containing lipoprotein (Lp) families defined by their unique apolipoprotein composition to major lipoprotein density classes against the density (d) gradient background (d=0.92 to 1.25 g/mL). The lines under lipoprotein families designate the approximate density boundaries, with *solid lines* depicting the actual localization of each lipoprotein family and *dotted lines* the possible localization of each lipoprotein family. Lipoprotein families represent polydisperse systems of particles, each of which has a different lipid/protein ratio, but the same qualitative apolipoprotein composition. The polydisperse character of lipoprotein families is the main reason for their overlap within certain density segments. (Chylos—chylomicron; HDL—high-density lipoprotein; IDL—intermediate-density lipoprotein; LDL—low-density lipoprotein; VHDL—very high-density lipoprotein; VLDL—very low-density lipoprotein.)

other by apoB. The former class consists of three major lipoprotein families referred to (according to their apolipoprotein constituents) as Lp-A-I, Lp-A-I:A-II, and Lp-A-II, and the latter of five major lipoprotein families named Lp-B, Lp-B:C, Lp-B:E, Lp-B:C:E, and Lp-A-II:B:C:D:E (Fig. 1) [17]. The third class of lipoproteins of high- and very high-density properties encompasses lipid-protein complexes characterized by apolipoproteins A-IV, A-V, D, E, F, G, H, I, K, L, and M as sole apolipoprotein constituents or, frequently, in association with apoA-I; however, with a few exceptions, a direct role of these lipoproteins in lipid transport has not yet been defined.

Both the apoA- and apoB-containing lipoproteins represent polydisperse systems of particles heterogeneous with respect to physical properties (*ie*, size and density) and lipid/protein ratios, but homogeneous with respect to qualitative apolipoprotein composition. ApoA-containing lipoprotein families overlap within the HDL density range and apoB-containing lipoproteins within the VLDL, IDL, and LDL density ranges (Fig. 1). The magnitude of polydispersity and distribution of lipoprotein families along the density gradient of 0.92 to 1.21 g/mL depend on the concentrations of lipids to be transported and processes responsible for their degradation and removal.

The lipid composition of all three apoA-containing lipoproteins is characterized by high percentages of phospholipids (55% to 65%) and varying cholesterol ester/free cholesterol ratios and triglyceride levels [18,19]. Approximately 20% to 25% of lipoprotein-A-I (Lp-A-I) and Lp-A-I:A-II subclasses contain minor apolipoproteins as integral protein constituents [11].

The lipid composition of Lp-B and Lp-B:E is characterized by cholesterol esters as the main neutral lipid constituent regardless of their density properties; on the other hand, Lp-B:C, Lp-B:C:E, and Lp-A-II:B:C:D:E families contain triglycerides as the most characteristic neutral lipid [11]. However, with increasing densities of these particles, the relative content of triglycerides decreases and that of cholesterol esters increases. The apolipoprotein composition of triglyceride-rich lipoproteins also changes with increasing densities: the relative content of apoB increases and the relative contents of apoC-peptides and apoE decrease [11].

It has been established in a number of studies that apolipoprotein-defined apoA- and apoB-containing lipoprotein families are characterized not only by specific apolipoprotein composition, but also by specific metabolic and

functional properties [11,17]. The turnover rate of apoA-I in Lp-A-I has been found to be faster than that of apoA-I in Lp-A-I:A-II [20]. It has been shown recently that apoE is metabolized at a slower rate when associated with Lp-A-I:A-II than Lp-A-I or Lp-A-II subclasses [21•]. The Lp-A-I particles, but not Lp-A-I:A-II, seem to function as the acceptors of peripheral cholesterol [22,23], and, in association with lecithin:cholesterol acyltransferase (LCAT) and cholesterol ester transfer protein (CETP), as templates for esterifying and transferring cholesterol in plasma [24]. Lp-A-I have greater binding affinities for various cell membranes than Lp-A-I:A-II [25]. On the other hand, Lp-A-I:A-II seem to have a greater capacity than Lp-A-I for binding apoC peptides and apoE released during the lipolytic degradation of triglyceride-rich lipoproteins, and to provide these minor apolipoproteins for the completion of extracellular formation of triglyceride-rich lipoproteins in space of Disse [26,27].

It has been shown that HepG2 cells secrete mainly triglyceride-rich Lp-B and Lp-B:E, suggesting that these two families may be the precursors for the extracellular formation of Lp-B:C and Lp-B:C:E [28]. Alaupovic *et al.* [29] have shown that the triglyceride-rich Lp-B:C, Lp-B:C:E, and Lp-A-II:B:C:D:E families have different affinities for lipoprotein lipase (LPL) despite similar triglyceride and apoC-III contents. Thus, Lp-B:C:E particles seem to be the most efficient and Lp-A-II:B:C:D:E particles the least efficient substrate for LPL. ApoB-containing lipoprotein families differ also in their binding affinities for the LDL receptors. The Lp-B:E bind to LDL receptors on human fibroblasts, HepG2, and HeLa cells with greater affinity than Lp-B [30–32]. In contrast, the binding of Lp-B:C to LDL receptors of HeLa cells has been found to be negligible, suggesting that this lipoprotein family has very little effect on regulating 3-hydroxy-3-methylglutaryl coenzyme A (HMG CoA) reductase activity [31]. ApoB-containing lipoprotein families may also have different binding affinities and rates of uptake by human THP1 macrophages [33]; Lp-B:C were shown to have the greatest affinity followed by Lp-B:C:E, Lp-A-II:B:C:D:E, and Lp-B in descending order.

These findings have provided strong evidence for the crucial role of minor apolipoproteins in modifying and determining the metabolic properties of individual apoA- and apoB-containing lipoprotein families.

Clinical significance of lipoprotein families

Metabolic derangements of lipid transport processes are of great clinical significance because they are considered as one of the main factors responsible for the genesis and development of atherosclerotic disease [1,2,4,34]. Normolipidemia and a variety of dyslipoproteinemias are characterized by specific concentration profiles of apoA- and apoB-containing lipoprotein families. Due to the complexity of plasma lipoproteins, the only reliable methods for the quantification of apolipoprotein-defined lipoprotein families are based on highly specific and sensitive immunologic procedures, including immunoprecipitation [35,36], immunoaffinity chromatography [37], enzyme linked differential-antibody immunosorbent assays [38], and differential electroimmunoassays [39,40].

It is generally accepted that apoB is the marker of atherogenic lipoproteins and apoA-I the marker of antiatherogenic lipoproteins. The recognition that apoA- and apoB-containing lipoprotein families have distinct apolipoprotein composition and metabolic properties has raised the question as to whether or not lipoprotein families may also possess different antiatherogenic or atherogenic potentials. Although the entire HDL has been thought to be nonatherogenic, the finding that patients with documented coronary artery disease (CAD) have lower levels of LpA-I than LpA-II:A-II in comparison with control subjects has suggested that the former may possess a greater cardioprotective capacity than the latter [41]. In subsequent studies, some investigators found no difference in the levels of Lp-A-I and Lp-A-I:A-II between subjects with and without CAD [42–44], whereas others confirmed the original observation about the greater antiatherogenic potential of Lp-A-I in comparison with that of LpA-I-A-II [45–49]. In a recently reported prospective study of two populations of Northern Ireland and France [50], it was concluded that both Lp-A-I and LpA-I:A-II were significantly associated with the incidence of CAD, and that there was no difference between these two subclasses as predictors of atherosclerotic disease. However, due to its direct role in reverse cholesterol transport, Lp-A-I appears to be more compatible with a greater cardioprotective capacity than Lp-A-I:A-II. Thus, the possible differences in the relative antiatherogenicity between Lp-A-I and Lp-A-I:A-II still remain to be resolved in future studies.

The question of absolute and relative atherogenicity of apoB-containing lipoproteins is quite complicated due to differences in the lipid and apolipoprotein composition and physical properties of individual lipoprotein families. According to the classification of lipoproteins based on density properties, cholesterol-rich LDL are considered to have the highest atherogenic capacity followed in decreasing order of atherogenicity by intact or partially delipidized triglyceride-rich IDL and small VLDL; the large VLDL and chylomicrons are thought to have negligible atherogenic potential. Thus, the size or density of lipoprotein particles represents an important determinant of their atherogenicity [51]. Based on the distribution studies of discrete apoB-containing lipoproteins along the density gradient between 0.94 to 1.063 g/mL, it is possible to equate cholesterol-rich Lp-B and Lp-B:E particles with LDL and the triglyceride-rich Lp-B:C, Lp-B:C:E, and Lp-A-II:B:C:D:E particles with VLDL and IDL. On this basis, Lp-B and Lp-B:E particles may be considered to be the main atherogenic lipoproteins, whereas the atherogenicity of Lp-B:C, Lp-B:C:E, and Lp-A-II:B:C:D:E particles may be relatively low in comparison with that of cholesterol-rich apoB-containing lipoproteins. However, the atherogenicity of discrete apoB-containing lipoprotein families cannot be derived from or determined by comparison

with that of lipoprotein density classes because density classes are mixtures of varying concentrations of individual, polydisperse apoB-containing lipoprotein families. The problem of atherogenicity is further compounded by the controversial views regarding the contribution of hypertriglyceridemic and hypercholesterolemic lipoprotein profiles to the risk of CAD. Although, in general, hypertriglyceridemia with or without associated hypercholesterolemia occurs more frequently in patients with premature CAD than hypercholesterolemia, most of the major prospective studies have shown that lowering of LDL cholesterol, but not triglycerides, has a significant beneficial effect on the progression of atherosclerotic disease and reduction of coronary events [52]. Although these studies have provided little evidence that plasma triglyceride levels represent an independent predictor of CAD, it has been demonstrated in some subsets of patient populations (*eg*, women, elderly subjects, and patients with type 2 diabetes) that increased triglyceride concentrations are an independent risk factor for the progression of atherosclerotic disease [53]. These findings have been supported by results of a meta-analysis of population-based prospective studies [54] and clinical trials [55,56•] confirming that hypertriglyceridemia is an independent risk factor for coronary events even after adjustments for several lipid and clinical confounding factors, including HDL cholesterol. In addition, a number of clinical and metabolic studies have indicated that partially delipidized triglyceride-rich lipoproteins (remnant lipoproteins) may have atherogenic potentials similar to, if not greater than, that of cholesterol-rich LDL particles [56•,57]. One of the possible reasons for this LDL cholesterol/triglyceride controversy may be due to the selection of markers for the statistical evaluation of the atherogenicity of cholesterol-rich and triglyceride-rich lipoproteins. As a marker of cholesterol-rich lipoproteins, LDL cholesterol encompasses a relatively narrow density segment of apoB-containing lipoproteins and, when determined by the Friedewald formula, it also includes the IDL known to contain atherogenic, partially delipidized, triglyceride-rich lipoproteins (*ie*, remnant lipoproteins). In contrast, plasma triglyceride levels as the marker of triglyceride-rich lipoproteins encompass the entire lipoprotein density spectrum, including the non-atherogenic chylomicrons and large VLDL as well as anti-atherogenic HDL. Based on these considerations, it appears that the use of triglycerides as a marker of potentially atherogenic triglyceride-rich lipoproteins is not equivalent to the use of LDL cholesterol as a marker of cholesterol-rich lipoproteins. Thus, when estimating atherogenicity of apoB-containing lipoproteins, the use of triglycerides introduces an a priori bias in statistical evaluations.

The relationship of apoB-containing lipoproteins to atherosclerosis was explored in two prospective angiographic trials, including the Cholesterol Lowering Atherosclerosis Study (CLAS) [58], and Monitored Atherosclerosis Regression Study (MARS) [47]. The 2-year treatment with a niacin-colestipol combination of subjects with moderate hypercholesterolemia and previous bypass surgery in CLAS and the treatment with lovastatin of subjects with moderate hypercholesterolemia and angiographically documented CAD in MARS resulted in significantly reduced progression of atherosclerotic lesions due to the lowering effect of these drugs on the levels of apoB and LDL cholesterol or cholesterol-rich Lp-B particles. However, despite highly reduced levels of LDL cholesterol (97 ± 27 mg/dL in CLAS and 82 ± 17 mg/dL in MARS), the continuing progression of atherosclerotic lesions in 35% to 40% of patients was shown to be associated with increased levels of the constituents of triglyceride-rich lipoproteins (triglycerides and apoC-III) in CLAS or increased concentration of directly measured sum of triglyceride-rich Lp-B:C, Lp-B:C:E, and Lp-A-II:B:C:D:E (referred to as Lp-B complex) and individually measured Lp-A-II:B:C:D:E. It was shown in both trials that niacin-colestipol combination and lovastatin had significant lowering effects on LpB, but no effects on triglyceride-rich Lp-B:C and LpB:C:E [40,59].

In a search to replace plasma triglycerides with a more adequate marker for the atherogenicity of triglyceride-rich lipoproteins, it was shown in the ancillary Cholesterol and Recurrent Events (CARE) study [60] that statistically, the most powerful predictors of the recurrent coronary events in patients with previous myocardial infarction were VLDL-apoB and apoC-III bound to apoB-containing lipoproteins, even after adjustments for LDL and HDL cholesterol and total and VLDL triglycerides.

Results of CLAS and MARS have already suggested that minor apolipoproteins may not only modify the metabolic properties of lipoprotein families, but also affect to various degrees their atherogenic potentials by establishing that Lp-B:C, Lp-B:C:E, or Lp-A-II:B:C:D:E may have similar, if not greater, atherogenic capacity than Lp-B. This was confirmed in a clinical trial of men undergoing coronary angiography indicating that the severity of CAD was significantly correlated with Lp-B:C:E and Lp-B:C, but not with Lp-B particles [61]. In a study of subjects with type 2 diabetes, it was shown that diabetic patients with clinically verified vascular disease had similar levels of Lp-B, but a twofold higher level of Lp-B:C than diabetic patients without vascular disease [62]. This finding has been recently confirmed in a larger study of type 2 diabetic patients showing that the levels of Lp-B:C particles were independently associated with macrovascular complications of diabetes, with the highest odds ratio among independent variables [63]. In a recently reported ancillary CARE study [64••], type 2 diabetic patients with previous myocardial infarction were followed for 5 years, and 121 who had a recurrent coronary event were matched to 121 who did not. Measurement of Lp-B and Lp-B:C in VLDL and IDL plus LDL provided the first opportunity to compare the predictive power of these two atherogenic subclasses. Results showed that Lp-B:C particles in IDL plus LDL were a several-fold greater predictor of recurrent coronary events than Lp-B (relative risk [RR] of 6.6; $P<0.0001$ for Lp-B:C

Table 1. Lipoprotein A-I and lipoprotein A-I:A-II in plasma of normolipidemic and dyslipoproteinemic subjects

Dyslipoproteinemias	HDL-C, *mg/dL* (mean ± SD)	ApoA-I, *mg/dL* (mean ± SD)	Lp-A-I, *mg/dL* (mean ± SD)	Lp-A-I:A-II, *mg/dL* (mean ± SD)
Moderate hypercholesterolemia (*n*=253)	38 ± 10*	127 ± 20*	31.4 ± 4.8*	94.5 ± 18.4*
Hypertriglyceridemia (*n*=16)	36 ± 9*	123 ± 18†	33 ± 6	97 ± 17
Chronic renal failure (*n*=93)	38.7 ± 15*	114 ± 22*	34 ± 8	80 ± 21*
Type 2 diabetes (*n*=224)	44 ± 9*	114 ± 24*	30 ± 6*	84 ± 19*
Normolipidemia (*n*=238)	51 ± 14	139 ± 27	34 ± 7.5	104 ± 22
Men (*n*=125)	46 ± 11	131 ± 22	32 ± 6	98 ± 18
Women (*n*=113)	57 ± 17	150 ± 28	36 ± 8	112 ± 22

*$P<0.001$ in normolipidemic versus dyslipoproteinemic subjects.
†$P<0.05$ in normolipidemic versus dyslipoproteinemic subjects.
Apo—apolipoprotein; HDL-C—high-density lipoprotein cholesterol; Lp—lipoprotein.

versus RR of 2.2; $P<0.07$ for Lp-B). The relationship between Lp-B:C and coronary events was independent of standard lipid and apolipoprotein risk factors and other baseline characteristics of patients. The significant difference in the capacities of Lp-B and Lp-B:C particles to elicit recurrent coronary events in diabetic subjects commensurate with other presented findings indicates not only a greater atherogenic potential of Lp-B:C particles in comparison with that of Lp-B particles, but also supports the concept of relative atherogenicity of apoB-containing lipoproteins. The relative atherogenicity of Lp-B:E, Lp-B:C:E, and Lp-A-II:B:C:D:E particles in relation to those of Lp-B and Lp-B:C remains to be established in future studies.

The Effect of Pharmacologic and Dietary Interventions on ApoA- and ApoB-containing Lipoprotein Families

Apolipoprotein A- and apoB-containing lipoprotein families occur in varying concentrations in almost all normolipidemic and dyslipoproteinemic subjects. Lp-A-I accounts for approximately 20% to 25% and Lp-A-I:A-II for 70% to 75% of total apoA-I (Table 1). In general, dyslipoproteinemic subjects with or without CAD tend to have lower levels of apoA-I than normolipidemic, asymptomatic subjects. Due to higher levels of apoA-I, women have higher levels of both Lp-A-I and Lp-A-I:A-II than men. There are, however, significant differences in the levels of Lp-A-I and Lp-A-I:A-II among individuals of both genders depending on several genetic and environmental factors. The goal of intervention therapy is to raise levels of Lp-A-I and/or Lp-A-I:A-II in subjects with low levels of these subclasses.

In both the dyslipoproteinemic and normolipidemic subjects, Lp-B is the main lipoprotein subclass, accounting for 50% to 65% of total apoB (Table 2). However, the sum of complex apoB-containing lipoproteins, including Lp-B:C, Lp-B:E + Lp-B:C:E, and Lp-A-II:B:C:D:E, accounts for an almost equal percentage of total plasma apoB content, especially in subjects with primary hypercholesterolemia and hypertriglyceridemia. All complex apoB-containing lipoprotein families are significantly increased in both primary and secondary dyslipoproteinemias when compared with normolipidemic, asymptomatic control subjects.

Pharmacologic interventions

Atmeh *et al.* [65] first showed that lipid-lowering drugs may affect the levels of lipoprotein families in a specific manner by establishing that niacin increases and probucol decreases the concentration of Lp-A-I with little or no effect on the levels of Lp-A-I:A-II. All statins increase moderately the levels of Lp-A-I and Lp-A-I:A-II [40,66], with the exception of pravastatin, which seems to be the only statin that increases significantly the levels of Lp-A-I:A-II [67,68]. In contrast, both gemfibrozil [68] and fenofibrate [66] increase markedly the concentration of Lp-A-I:A-II, but show no effect on that of Lp-A-I. Cholestyramine [67] increases both the Lp-A-I and Lp-A-I:A-II, and, in combination with niacin, appears to be the most efficient pharmacologic agent in raising the levels of apoA-I and its corresponding lipoprotein subclasses.

Statins and fibrates, in addition to niacin and bile acid-sequestering resins, represent a wide choice of drugs primarily affecting atherogenic apoB-containing lipoproteins. It has been well documented that, in general, statins are more effective in lowering the cholesterol-rich [40,47,66-69] and fibrates the triglyceride-rich apoB-containing lipoproteins [66,68]. It is customary to determine the effect of lipid-lowering drugs by measuring the levels of LDL cholesterol as a marker of cholesterol-rich lipoproteins and plasma triglyceride as a marker of triglyceride-rich lipoproteins. However, because these two markers may encompass several discrete apoB-containing lipoprotein families, the actual effect of statins or fibrates cannot be assessed solely on the basis of LDL cholesterol or triglyceride levels. There is already evidence to illustrate this point. For example, in subjects with combined hyperlipidemia, atorvastatin (20 mg/d) lowered significantly cholesterol-rich Lp-B (26%) and triglyceride-rich Lp-B:C and Lp-B:C:E (44%), but had very little effect on the levels of Lp-A-II:B:C:D:E (10%) [69]. On the other hand, in subjects with combined hyperlipidemia, simvastatin

Table 2. Apolipoprotein B-containing lipoprotein families in plasma of normolipidemic and dyslipoproteinemic subjects

Dyslipoproteinemias	ApoB, *mg/dL* (mean ± SD)	Lp-B, *mg/dL* (mean ± SD)	Lp-B:C, *mg/dL* (mean ± SD)	Lp-B:E + Lp-B:C:E, *mg/dL* (mean ± SD)	Lp-A-II:B:C:D:E, *mg/dL* (mean ± SD)
Phenotype IIA (*n*=35)	142 ± 15*	73 ± 7.7*	16.4 ± 4*	18.3 ± 4.7*	20.8 ± 5.9*
Phenotype IIB (*n*=26)	171 ± 15*	91 ± 10.7*	21.7 ± 7.1*	21.7 ± 8.2*	31.5 ± 10.7*
Chronic renal failure before dialysis (*n*=15)	137 ± 409*	82 ± 30*	20.3 ± 10*	22.3 ± 7.5*	12 ± 5.6
Type 2 diabetes (*n*=7)	119 ± 17†	69 ± 18‡	14 ± 7.2	21 ± 18§	15 ± 5.3
Normolipidemia (*n*=75)	93 ± 19	56 ± 12	10.6 ± 5.3	12 ± 5.3	13.4 ± 8
Men (*n*=34)	95 ± 20	54 ± 13	13 ± 6.1	13 ± 6.1	14.8 ± 8
Women (*n*=39)	90 ± 17	58 ± 11	8.5 ± 3.7	11 ± 4.4	11.8 ± 6.7

*$P<0.0001$ in normolipidemic versus dyslipoproteinemic subjects.
†$P<0.001$ in normolipidemic versus dyslipoproteinemic subjects.
‡$P<0.01$ in normolipidemic versus dyslipoproteinemic subjects.
§$P<0.005$ in normolipidemic versus dyslipoproteinemic subjects.
Apo—apolipoprotein; Lp—lipoprotein.

(80 mg/d) reduced significantly Lp-B (18%), Lp-B:E + Lp-B:C:E (30%), and Lp-A-II:B:C:D:E (45%), but had only a slight lowering effect on Lp-B:C (10%) [70]. This outcome could not have been predicted from almost identical lowering effect of these two statins on LDL cholesterol (34% vs 33%), triglycerides (30% vs 32%), apoB (32% vs 31%), or apoC-III (27% vs 26%). As inhibitors of HMG CoA reductase, all statins share the ability to lower plasma cholesterol by inhibiting hepatic cholesterol synthesis and upregulating LDL receptors. Consequently, the most characteristic lipoprotein-lowering effect of statins is to decrease plasma levels of cholesterol-rich Lp-B and Lp-B:E particles; statins differ, however, in the capacity to lower equally and consistently the levels of Lp-B:C and Lp-A-II:B:C:D:E particles.

In contrast to statins, fibrates lower markedly plasma triglycerides accompanied by moderate to insignificant reduction of plasma cholesterol or LDL cholesterol. Surprisingly, the pronounced lowering of plasma triglycerides is not always reflected in an equal reduction of the number of triglyceride-rich lipoprotein particles. For example, lowering of triglycerides by gemfibrozil was threefold greater than lowering of apoC-III or corresponding triglyceride-rich particles [68]. On the other hand, fenofibrate treatment resulted in a similar reduction of triglycerides, apoC-III, and triglyceride-rich lipoprotein families [66]. Because the lowering effect of fibrates is mediated through the activation of peroxisome proliferator activated receptors causing, among others, the repression of apoC-III and induction of LPL genes, it may be that gemfibrozil and fenofibrate have a similar effect on the expression of LPL gene, but a differential effect on the expression of apoC-III gene. Aside from statins and fibrates, it was shown in CLAS that the combination of niacin-colestipol had a highly significant effect, similar to that of statins, in reducing Lp-B, but no effect in lowering Lp-B:C and Lp-B:C:E particles [59].

These rather sketchy findings should provide the basis for further systematic studies on differential effects of hypolipidemic drugs on apoA- and apoB-containing lipoprotein families.

Dietary interventions

Diet plays an important, if not essential, role in affecting lipid transport processes as manifested by recommendations that dietary interventions should always be undertaken as the first therapeutic step in normalizing or ameliorating abnormalities of plasma lipoproteins. In innumerable studies, nutritional interventions have been aimed mainly, but not exclusively, at the content of cholesterol and the quantity and fatty acid composition of dietary fat, because of well-documented influence of these nutrients on the levels of atherogenic apoB-containing and antiatherogenic apoA-containing lipoproteins. The intervention studies have been monitored and evaluated almost exclusively by measurements of plasma lipids and lipoprotein cholesterol and, more recently, by apoA-I and B. In contrast to pharmacologic interventions, there are only a few studies on the effects of diets on individual apoA-containing lipoproteins and practically no information on apoB-containing lipoproteins.

Delplanque *et al.* [71] and Fumeron *et al.* [72] were first to establish that diets with high ratio of polyunsaturated to saturated fatty acids (1.1 to 1.2) lowered the levels of antiatherogenic Lp-A-I, but had no effect on the levels of Lp-A-I:A-II. The lowering of Lp-A-I levels counteracted to some extent the beneficial effect of these diets in reducing the concentration of cholesterol-rich apoB-containing lipoproteins. However, it was shown by Delplanque *et al.* [71] that the negative effect of polyunsaturated fatty acids may be corrected by the use of an equal ratio of monounsaturated to polyunsaturated fatty acids; although this still decreased the levels of LDL cholesterol, this diet had no effect on Lp-A-I. It appears that increasing ratios of monounsaturated to polyunsaturated fatty acids, with no change in saturated fatty acids, tend to increase the levels of apoA-I, Lp-A-I, and Lp-A-I:A-II, although monounsaturated oleic acid alone

increases mainly Lp-A-I:A-II [71]. The effect of high polyunsaturated to saturated fatty acid ratio and that of equal monounsaturated to polyunsaturated fatty acid ratios on apoA-containing lipoprotein subclasses have been confirmed in subsequent studies [73,74].

There is only one study evaluating the effect of a high-fat diet (40% of calories from fat, polyunsaturated to saturated fat ratio of 0.11) on the postprandial levels of lipids, apolipoproteins, and apoB-containing lipoprotein families in normolipidemic (triglyceride level of 84 ± 36 mg/dL) and hypertriglyceridemic (triglyceride level of 237 ± 85 mg/dL) subjects [75]. The major changes in postprandial state only occurred in lipoproteins with density gradient less than 1.020 g/mL characterized by slight increases in the levels of triglycerides, apoB, and apoC-III in both groups of subjects. However, there were significant differences in the postprandial levels of apoB-containing lipoprotein subclasses between normolipidemic and hypertriglyceridemic subjects. The levels of atherogenic Lp-B:C were increased twofold in the latter and slightly decreased in the former, whereas the levels of Lp-A-II:B:C:D:E were increased in the former and decreased in the latter, a finding that could not have been predicted from the changes in the levels of plasma apoB. In a recent study (unpublished data) in collaboration with Kris-Etherton and associates, we have compared the effects of three experimental diets, including high-oleic acid fat blend, high-oleic acid fat blend with the addition of omega-3 marine fatty acids, and a high-oleic acid fat blend with the addition of α-linolenic acid, on the postprandial concentrations of apoB-containing lipoprotein families in type 2 diabetic subjects. The results have shown that all three diets had similar postprandial effects in increasing the levels of total apoB, but different effects on the concentration of individual apoB-containing lipoproteins. The postprandial increase in the levels of atherogenic Lp-B:C particles was significantly greater on the oleic acid diet compared with a moderate effect of α-linolenic and a negligible effect of omega-3 marine fatty acids; this increasing effect of oleic acid on the levels of Lp-B:C was further exacerbated in a subpopulation of diabetic subjects with plasma apoB levels greater than 100 mg/dL.

These findings and tentative conclusions deserve to be investigated and confirmed in larger population studies of normolipidemic and dyslipoproteinemic subjects, because they show that diets, especially in postprandial state, may have significant effects on individual apolipoprotein-defined apoA- and apoB-containing lipoprotein families similar to those of pharmacologic agents.

Conclusions

By emphasizing apolipoproteins as chemically unique constituents and metabolic and functional determinants of plasma lipoproteins, the concept of lipoprotein families provides a framework capable of incorporating any number of apolipoproteins and corresponding lipoprotein families into an integrated system of lipid transport. Moreover, this concept maintains that normal and defective lipid transport processes ought to be regarded, measured, and evaluated in terms of macromolecular lipoprotein families of particles rather than their individual lipid or apolipoprotein constituents. Because the ubiquitous lipoprotein constituents such as cholesterol or triglyceride are not specific or unique markers for discrete lipoprotein families, the measurement of lipids either in plasma or in major lipoprotein density classes cannot describe adequately the concentration profiles of these chemically and metabolically unique entities of plasma lipoprotein system. There is already substantial evidence to indicate that individual apoA-containing lipoprotein families may differ in their antiatherogenic and apoB-containing lipoprotein families in atherogenic potentials. Although the quantification of plasma apoA-I and apoB provides an estimate of the number of corresponding antiatherogenic and atherogenic lipoproteins, the distribution of these two major apolipoproteins among discrete metabolically and pathophysiologically different lipoprotein families can only be determined by direct measurement of these apolipoprotein-defined lipoproteins. Because, in general, the same distinct apoA- and apoB-containing lipoprotein families occur in subjects with normal or aberrant lipoprotein profiles, the difference between normal and dyslipoproteinemic states results from quantitative rather than qualitative changes in individual lipoprotein families.

The use of a variety of pharmacologic agents for treating dyslipoproteinemias has already shown that these drugs alone or in combination may affect selectively individual apolipoprotein-defined lipoproteins. Although it is necessary and important to continue with systematic studies of pharmacologic therapeutic interventions, there is an urgent need to extend such studies to the effect of diets and dietary interventions on individual apoA- and apoB-containing lipoprotein families in normal and dyslipoproteinemic states. The ultimate goal of both the pharmacologic and dietary interventions is to develop individualized treatments targeted at decreasing undesirable and/or increasing the desirable concentrations of apolipoprotein-defined lipoprotein families.

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References and Recommended Reading

Papers of particular interest, published recently, have been highlighted as:
- • Of importance
- •• Of major importance

1. Gofman JW, De Lalla O, Glazier F, *et al.*: **The serum lipoprotein transport system in health, metabolic disorders, atherosclerosis, and coronary heart disease.** *Plasma* 1954, **2**:413–484.
2. Jones HB, Gofman JW, Lindgren FT, *et al.*: **Lipoproteins in atherosclerosis.** *Am J Med* 1951, **11**:358–380.

3. Nichols AV: **Human serum lipoproteins and their interrelationships.** *Adv Biol Med Phys* 1967, **11**:109–158.
4. Fredrickson DS, Levy RI, Lees RS: **Fat transport in lipoproteins: an integrated approach to mechanism and disorders.** *N Engl J Med* 1967, **276**:32–281.
5. Ewing AM, Freeman NK, Lindgren FT: **The analysis of human serum lipoprotein distributions.** *Adv Lipid Res* 1965, **3**:25–61.
6. Nestel P, Billington T, Tada N, *et al.*: **Heterogeneity of very-low-density lipoprotein metabolism in hyperlipidemic subjects.** *Metabolism* 1983, **32**:810–817.
7. Nestel P: **High-density lipoprotein turnover.** *Am Heart J* 1987, **113**:518–521.
8. Packard CJ, Shepherd J: **Lipoprotein heterogeneity and apolipoprotein B metabolism.** *Arterioscler Thromb Vasc Biol* 1997, **17**:3542–3556.
9. Alaupovic P: **Conceptual development of the classification systems of plasma lipoproteins.** *Protides Biol Fluids Proc Colloq* 1972, **19**:9–19.
10. Osborne JC Jr, Brewer HB Jr: **The plasma lipoproteins.** *Adv Protein Chem* 1977, **31**:253–337.
11. Alaupovic P: **Apolipoprotein composition as the basis for classifying plasma lipoproteins. Characterization of ApoA- and ApoB-containing lipoprotein families.** *Prog Lipid Res* 1991, **30**:105–138.
12. Fredrickson DS: **Phenotyping. On reaching base camp (1950–1975).** *Circulation* 1993, **87(suppl II)**:1–15.
13. Alaupovic P, Lee DM, McConathy WJ: **Studies on the composition and structure of plasma lipoproteins. Distribution of lipoprotein families in major density classes of normal human plasma lipoproteins.** *Biochim Biophys Acta* 1972, **260**:689–707.
14. Lee DM, Alaupovic P: **Composition and concentration of apolipoproteins in very-low- and low-density lipoproteins of normal human plasma.** *Atherosclerosis* 1974, **19**:501–520.
15. Lee DM, Alaupovic P: **Apolipoproteins B, C-III and E in two major subpopulations of low-density lipoproteins.** *Biochim Biophys Acta* 1986, **879**:126–133.
16. Cheung MC, Albers JJ: **Distribution of cholesterol and apolipoprotein A-I and A-II in human high density lipoprotein subfractions separated by NaCl equilibrium gradient centrifugation: evidence for HDL subpopulations with differing A–I/A–II molar ratios.** *J Lipid Res* 1979, **20**:200–207.
17. Alaupovic P: **Significance of apolipoproteins for structure, function and classification of plasma lipoproteins.** In *Methods in Enzymology. Plasma Lipoproteins, Part C, Quantitation.* Edited by Bradley WA, Gianturco SH, Segrest JP. San Diego: Academic Press; 1996.
18. Bekaert ED, Alaupovic P, Knight-Gibson C, *et al.*: **Composition of plasma ApoA-I–containing lipoprotein particles in children and adults.** *Pediatr Res* 1991, **29**:315–321.
19. Ohta T, Ikeda Y, Nakamura R, *et al.*: **Lipoprotein-containing apolipoprotein A-I–sex-related quantitative and qualitative changes in this lipoprotein subspecies after ingestion of fat.** *Am J Clin Nutr* 1992, **56**:404–409.
20. Rader DJ, Castro G, Zech LA, *et al.*: **In vivo metabolism of apolipoprotein A-I on high density lipoprotein particles LpA-I and LpA-I:A-II.** *J Lipid Res* 1991, **32**:1849–1859.
21.• Hannuksela ML, Brousseau ME, Meyn SM, *et al.*: **In vivo metabolism of apolipoprotein E within the HDL subpopulations LpE, LpE:A-I, LpE:A-II and LpE:A-I:A-II.** *Atherosclerosis* 2002, **165**:205–220.

This paper provides the most recent evidence for the metabolic specificity of apoA-containing lipoprotein subclasses in HDL

22. Fielding CJ, Fielding PE: **Evidence for a lipoprotein carrier in human plasma catalyzing sterol efflux from cultured fibroblasts and its relationship to lecithin:cholesterol acyltransferase.** *Proc Natl Acad Sci USA* 1981, **78**:3911–3914.
23. Barkia A, Puchois P, Ghalim N, *et al.*: **Differential role of apolipoprotein A1-containing particles in cholesterol efflux from adipose cells.** *Atherosclerosis* 1991, **87**:135–146.
24. Cheung MC, Wolf AC, Lum KD, *et al.*: **Distribution and localization of lecithin:cholesterol acyltransferase and cholesteryl ester transfer activity in A-I–containing lipoproteins.** *J Lipid Res* 1986, **27**:1135–1144.
25. Rinninger F, Kaiser T, Windler E, *et al.*: **Selective uptake of cholesteryl esters from high-density lipoprotein-derived LpA-I and LpA-I:A-II particles by hepatic cells in culture.** *Biochim Biophys Acta* 1998, **1393**:277–291.
26. Alaupovic P: **David Rubenstein Memorial Lecture: the biochemical and clinical significance of the interrelationship between very low density and high density lipoproteins.** *Can J Biochem* 1981, **59**:565–579.
27. James RW, Pometta D: **Postprandial lipemia differentially influences high density lipoprotein subpopulations LpAI and LpAI,AII.** *J Lipid Res* 1994, **35**:1583–1591.
28. Dashti N, Alaupovic P, Knight-Gibson C, *et al.*: **Identification and partial characterization of discrete apolipoprotein B-containing lipoprotein particles produced by human hepatoma cell line HepG2.** *Biochemistry* 1987, **26**:4837–4846.
29. Alaupovic P, Knight-Gibson C, Wang CS, *et al.*: **Isolation and characterization of an apoA-II–containing lipoprotein (LP-A-II:B complex) from plasma very low density lipoproteins of patients with Tangier disease and type V hyperlipoproteinemia.** *J Lipid Res* 1991, **32**:9–19.
30. Koren E, Alaupovic P, Lee DM, *et al.*: **Selective isolation of human plasma low-density lipoprotein particles containing apolipoproteins B and E by use of a monoclonal antibody to apolipoprotein B.** *Biochemistry* 1987, **26**:2734–2740.
31. Agnani G, Bard JM, Candelier L, *et al.*: **Interaction of LpB, LpB:E, LpB:C-III, and LpB:C-III:E lipoproteins with the low density lipoprotein receptor of HeLa cells.** *Arterioscler Thromb* 1991, **11**:1021–1029.
32. Clavey V, Lestavel-Delattre S, Copin C, *et al.*: **Modulation of lipoprotein B binding to the LDL receptor by exogenous lipids and apolipoproteins CI, CII, CIII, and E.** *Arterioscler Thromb Vasc Biol* 1995, **15**:963–971.
33. Koren E, Koscec M, Corder C, *et al.*: **Differential atherogenicity of complex apoB-containing lipoprotein particles.** *Atherosclerosis* 1994, **109**:217–218.
34. Wallace RB, Anderson RA: **Blood lipids, lipid-related measures and the risk of atherosclerotic cardiovascular disease.** *Epidemiol Rev* 1987, **9**:95–119.
35. März W, Trommlitz M, Gross W: **Differential turbidimetric assay for subpopulations of lipoproteins containing apolipoprotein A-I.** *J Clin Chem Clin Biochem* 1988, **26**:573–578.
36. Alaupovic P, Tavella M, Fesmire J: **Separation and identification of apoB-containing lipoprotein particles in normolipidemic subjects and patients with hyperlipoproteinemias.** *Adv Exp Med Biol* 1987, **210**:7–14.
37. Alaupovic P, Koren E: **Immunoaffinity chromatography of plasma lipoprotein particles.** In *Analyses of Fats, Oils and Lipoproteins.* Edited by Perkins EG. Champaign, IL: American Oil Chemists' Society; 1991.
38. Koren E, Puchois P, Alaupovic P, *et al.*: **Quantification of two different types of apolipoprotein A-I containing lipoprotein particles in plasma by enzyme-linked differential-antibody immunosorbent assay.** *Clin Chem* 1987, **33**:38–43.
39. Parra HJ, Mezdour H, Ghalim N, *et al.*: **Differential electroimmunoassay of human LpA-I lipoprotein particles on ready-to-use plates.** *Clin Chem* 1990, **36**:1431–1435.
40. Alaupovic P, Fesmire JD, Hunninghake D, *et al.*: **The effect of aggressive and moderate lowering of LDL-cholesterol and low dose anticoagulation on plasma lipids, apolipoproteins and lipoprotein families in post coronary artery bypass graft trial.** *Atherosclerosis* 1999, **146**:369–379.
41. Puchois P, Kandoussi A, Fievet P, *et al.*: **Apolipoprotein A-I containing lipoproteins in coronary artery disease.** *Atherosclerosis* 1987, **68**:35–40.
42. Coste-Burel M, Mainard F, Chivot L, *et al.*: **Study of lipoprotein particles LpAI and LpAI:AII in patients before coronary bypass surgery.** *Clin Chem* 1990, **36**:1889–1891.

43. Montali A, Vega GL, Grundy SM: **Concentrations of apolipoprotein A-I–containing particles in patients with hypoalphalipoproteinemia.** *Arterioscler Thromb* 1994, **14:**511–517.
44. Cheung MC, Brown BG, Wolf AC, *et al.*: **Altered particle size distribution of apolipoprotein A-I–containing lipoproteins in subjects with coronary artery disease.** *J Lipid Res* 1991, **32:**383–394.
45. O'Brien T, Nguyen TT, Hallaway BJ, *et al.*: **The role of lipoprotein A-I and lipoprotein A-I/A-II in predicting coronary artery disease.** *Arterioscler Thromb Vasc Biol* 1995, **15:**228–231.
46. Duverger N, Rader D, Brewer HB Jr: **Distribution of subclasses of HDL containing ApoA-I without ApoA-II (LpA-I) in normolipidemic men and women.** *Arterioscler Thromb* 1994, **14:**1594–1599.
47. Alaupovic P, Mack WJ, Knight-Gibson C, *et al.*: **The role of triglyceride-rich lipoprotein families in the progression of atherosclerotic lesions as determined by sequential coronary angiography from a controlled clinical trial.** *Arterioscler Thromb Vasc Biol* 1997, **17:**715–722.
48. Brown BG, Zhao XQ, Chait A, *et al.*: **Simvastatin and niacin, antioxidant vitamins, or the combination for the prevention of coronary disease.** *N Engl J Med* 2001, **345:**1583–1592.
49. Asztalos BF, Batista M, Horvath KV, *et al.*: **Change in alpha1 HDL concentration predicts progression in coronary artery stenosis.** *Arterioscler Thromb Vasc Biol* 2003, **23:**847–852.
50. Luc G, Bard JM, Ferrières J, *et al.*: **Value of HDL cholesterol, apolipoprotein A-I, lipoprotein A-I, and lipoprotein A-I/A-II in prediction of coronary heart disease—The PRIME Study.** *Arterioscler Thromb Vasc Biol* 2002, **22:**1155–1161.
51. Nordestgaard BG, Tybaerg-Hansen A: **IDL, VLDL, chylomicrons and atherosclerosis.** *Eur J Epidemiol* 1992, **8:**92–98.
52. Gould AL, Rossouw JE, Santanello NC, *et al.*: **Cholesterol reduction yields clinical benefit: impact of statin trials.** *Circulation* 1998, **97:**946–952.
53. Alaupovic P: **On the atherogenicity of triglyceride-rich lipoproteins and a novel marker for the assessment of their atherogenic potentials.** *OCL* 2002, **9:**220–226.
54. Hokanson JE, Austin MA: **Plasma triglyceride levels is an independent risk factor for cardiovascular disease: a meta–analysis of population–based prospective studies.** *J Cardiovasc Risk* 1996, **3:**213–219.
55. Cullen P: **Evidence that triglycerides are an independent coronary heart disease risk factor.** *Am J Cardiol* 2000, **86:**943–949.
56.• Ginsberg HN: **New perspectives in atherogenesis. Role of abnormal triglyceride-rich lipoprotein metabolism.** *Circulation* 2002, **106:**2137–2142.

This paper is an excellent review and critical evaluation of recent studies on the atherogenicity of triglyceride-rich lipoproteins.

57. Zilversmit DB: **Atherogenic nature of triglycerides, post-prandial lipidemia, and triglyceride-rich remnant lipoproteins.** *Clin Chem* 1995, **41:**153–158.
58. Blankenhorn DH, Alaupovic P, Wickham E, *et al.*: **Prediction of angiographic change in native human coronary arteries and aortocoronary bypass grafts—lipid and nonlipid factors.** *Circulation* 1990, **81:**470–476.
59. Alaupovic P, Blankenhorn DH: **Identification of potentially atherogenic lipoprotein particles.** In *Molecular Biology of Atherosclerosis, Proceedings of the 57th European Atherosclerosis Meeting.* Edited by Halpern MJ. London: John Libbey & Company; 1992.
60. Sacks FM, Alaupovic P, Moye LA, *et al.*: **VLDL, apolipoproteins B, CIII, and E, and risk of recurrent coronary events in the cholesterol and recurrent events (CARE) Trial.** *Circulation* 2000, **102:**1886–1892.
61. Koren E, Corder C, Mueller G, *et al.*: **Triglyceride enriched lipoprotein particles correlate with the severity of coronary artery disease.** *Atherosclerosis* 1996, **122:**105–115.
62. Alaupovic P, Bard JM, Tavella M, *et al.*: **Identification of apoB-containing lipoprotein families in NIDDM.** *Diabetes* 1992, **41(suppl 2):**18–25.
63. Gervaise N, Garrigue MA, Lasfargues G, *et al.*: **Triglycerides, apoC3 and LpB:C3 and cardiovascular risk in type II diabetes.** *Diabetologia* 2000, **43:**703–708.
64.•• Lee SJ, Campos H, Moye LA, *et al.*: **LDL containing apolipoprotein CIII is an independent risk factor for coronary events in diabetic patients.** *Arterioscler Thromb Vasc Biol* 2003, **23:**853–858.

This paper is of pivotal importance for the recognition of relative atherogenicity of apoB-containing lipoproteins in general and a marked atherogenic potential of Lp-B:C particles in particular.

65. Atmeh RG, Shepherd J, Packard CJ: **Subpopulations of apolipoprotein A-I in human high-density lipoproteins. Their metabolic profiles and response to drug therapy.** *Biochim Biophys Acta* 1983, **751:**175–188.
66. Bard JM, Parra HJ, Camare R, *et al.*: **A multicenter comparison of the effects of simvastatin and fenofibrate therapy in severe primary hypercholesterolemia, with particular emphasis on lipoproteins defined by their apolipoprotein composition.** *Metabolism* 1992, **41:**498–503.
67. Bard JM, Parra HJ, Douste-Blazy P, *et al.*: **Effect of pravastatin, an HMG CoA reductase inhibitor, and cholestyramine, a bile acid sequestrant, on lipoprotein particles defined by their apolipoprotein composition.** *Metabolism* 1990, **39:**269–273.
68. Schweitzer M, Tessier D, Vlahos WD, *et al.*: **A comparison of pravastatin and gemfibrozil in the treatment of dyslipoproteinemia in patients with non–insulin-dependent diabetes mellitus.** *Atherosclerosis* 2002, **162:**201–210.
69. Alaupovic P, Heinonen T, Shurzinske L, *et al.*: **Effect of a new HMG-CoA reductase inhibitor, atorvastatin, on lipids, apolipoproteins and lipoprotein particles in patients with elevated serum cholesterol and triglyceride levels.** *Atherosclerosis* 1997, **133:**123–133.
70. Alaupovic P, Knight-Gibson C, Plotkin D, *et al.*: **Effect of simvastatin on apoB-containing lipoproteins in patients with hypertriglyceridemia.** *Abstract Book, XIV International Symposium on Drugs Affecting Lipid Metabolism.* New York: NY. 2001.
71. Delplanque B, Richard JL, Jacotot B: **Influence of diet on the plasma levels and distribution of apoA-I–containing lipoprotein particles.** *Prog Lipid Res* 1991, **30:**159–170.
72. Fumeron F, Brigant L, Parra HJ, *et al.*: **Lowering of HDL2-cholesterol and lipoprotein A-I particle levels by increasing the ratio of polyunsaturated to saturated fatty acids.** *Am J Clin Nutr* 1991, **53:**655–659.
73. Cheung MC, Lichtenstein AH, Schaefer EJ: **Effects of a diet restricted in saturated fatty acids and cholesterol on the composition of apolipoprotein A-I–containing lipoprotein particles in the fasting and fed states.** *Am J Clin Nutr* 1994, **60:**911–918.
74. Montoya MT, Porres A, Serrano S, *et al.*: **Fatty acid saturation of the diet and plasma lipid concentrations, lipoprotein particle concentrations, and cholesterol efflux capacity.** *Am J Clin Nutr* 2002, **75:**484–491.
75. Lee DM, Alaupovic P, Gibson C, *et al.*: **Effect of postprandial state on lipids, apolipoproteins and lipoprotein particles in normal and hypertriglyceridemic subjects.** *Atherosclerosis* 1994, **109:**217.