APOLIPOPROTEINS AND METABOLISM IN ATHEROSCLEROSIS*

ANTONIO M. GOTTO, JR.**

HOUSTON

The term "apolipoproteins" was initially coined by John Oncley in the 1940s to refer to the protein constituents of the plasma lipoproteins. Since then, we have learned a great deal about the structures, functions, and in some cases, the mechanisms of apolipoproteins.

One of the more important functions of apolipoproteins is to help bind and emulsify the water-insoluble lipids in lipoprotein packages. However, the apolipoproteins are not limited to structural functions. They also contribute to the regulation of lipoprotein metabolism.

With the exception of apolipoprotein B, or apo B, the apolipoproteins are located on the surface of the lipoproteins. The apolipoproteins contain unique structures called amphipathic helices. These helices were first described by Segrest, et al. in 1974 (1). Amphipathic helices allow the apolipoproteins to reside on the surface of the lipoprotein particle. Table 1 summarizes the apolipoproteins and some of their important properties.

Apo A's

Apo A is secreted by the liver and intestine. It is found in HDL and is also present in chylomicrons. Apo A-I is an activator of LCAT and is believed to play an important role in reverse cholesterol transport. Like Apo A-I, Apo A-II is secreted by the liver, but its intestinal origin is unclear. In addition to binding lipid, it has been suggested that apo A-II serves as an activator of the enzyme hepatic triglyceride lipase. Apo A-IV is secreted by the intestine and is present in chylomicrons and HDL. Apo A-IV can activate LCAT and may affect dietary fat absorption. Some investigators have suggested that apo's A-I, A-IV and possibly A-II are involved in reverse cholesterol transport since these apolipoproteins are able to bind to 'HDL receptors' on the surface of cells and subsequently to promote cholesterol removal.

Apo B's

Apo B is the major protein constituent of LDL, but it is also found in chylomicrons and VLDL. Human apo B is a glycoprotein which occurs

* From the Department of Medicine, The Methodist Hospital, Houston, Texas.

** Address reprint requests to The Author: The Methodist Hospital, 6565 Fannin, Mail Station A-601, Houston, Texas 77030.

TABLE 1
Apolipoproteins and their association with human diseases

Apoprotein	Plasma concentration (mg/ml)	Isoelectric point (pI)	M.W.	Function	Association with clinical disorders
A-I	1.0-1.2	5.85-5.40 ^a	28K	Activates LCAT	Tangier disease; apoA-I—apoCIII deficiency; atherosclerosis
A-II	0.3-0.5	5.0	8.5K	—	—
A-IV	0.16	5.5	46K	—	—
B-100	0.7-1.0	—	550K	Receptor-mediated catabolism of LDL	Abetalipoproteinemia; normotriglyceridemic abetalipoproteinemia (B-100 deficiency); atherosclerosis
B-48	—	—	275K	Chylomicron production	—
CI	0.04-0.06	7.5	6.5K	Activates (moderately) LCAT	—
CII	0.03-0.05	4.9	9K	Activates lipoprotein lipase	Familial Type I hyperlipoproteinemia
CIII	0.12-0.14	4.7-5.0 ^b	9K	Inhibits catabolism of tri-glyceride-rich lipoproteins	ApoA-I-apoCIII deficiency
E	0.025-0.050	6.0-5.7 ^c	34.2K	Receptor-mediated catabolism of apoE-containing lipoproteins	Familial Type III hyperlipoproteinemia

^a The isoelectric points of apoA-I isoproteins are: apoA-I₂ = 5.85; apoA-I₃ = 5.74; apoA-I₄ = 5.65; apoA-I₅ = 5.52; apoA-I₆ = 5.40. The major plasma isoprotein is apoA-I₄.

^b The isoelectric points of individual apo CIII isoproteins are: apoCIII-0 = 5.0; apoCIII-1 = 4.85; apoCIII-2 = 4.65.

^c The isoelectric points of individual apoE3 isoproteins are: apoE3 = 6.02; apo E3_{s-1} = 5.89; apoE3_{s-2} = 5.78; apoE3_{s-3} = 5.68. The isoelectric points of the common apoE variants are apoE2 = 5.89; apoE4 = 6.18.

Source: Breslow JL: Lipoprotein genetics and molecular biology. A.M. Gotto, Jr., Ed. *Plasma Lipoproteins*. Elsevier. Science Publishers B.V. Amsterdam, 1987:360 (Reprinted with permission)

in two forms, apo B and apo B-100. In man, Apo B-48 is synthesized by the intestine, while apo B-100 is synthesized by the liver. The human gene for apo B contains 43 kilobases with 9.8 introns and is located on chromosome 2. Apo B-48 occurs because of the insertion of a stop codon in the editing of the DNA (2). Most of the mRNA of the intestine is about 15 kilobases in size. This is the same as that for the mRNA for apo B-100. However, the stop codon is triggered by a CAA substitution for an UAA sequence. This substitution results in an aborted synthesis of the apolipoprotein and in the absence of the LDL receptor binding region.

Apo C's

Apo C-I, C-II and C-III are secreted by the liver. They are found in HDL and may also be transferred to VLDL and chylomicrons. Apo C-I can serve as an activator of LCAT; its other functions are unknown. Apo C-II is required to activate lipoprotein lipase. In certain kindreds, apo C-II is defective or deficient and is associated with a severe form of fasting chylomicronemia called Type I hyperlipoproteinemia. The clinical and lipoprotein abnormalities are very similar to those seen in familial lipoprotein lipase deficiency. Although the function of apo C-III is unknown, some investigators have suggested that apo C-III prevents a premature uptake of remnant particles by the liver.

Apo D's

Apo D is a minor protein which appears to be able to form complexes with HDL and the cholesteryl ester transfer protein (CETP), also called the lipid transfer protein (LTP). CETP catalyzes the exchange of cholesteryl ester and triglyceride between HDL and the lower-density lipoproteins.

Apo E's

Apo E is mainly synthesized in the liver, although a number of other tissues, including the brain, kidney and adrenal gland, are capable of making it. Synthesis and secretion of apo E has been shown in macrophages. Apo E serves as a ligand for the LDL or apo B/E receptor present on hepatic and extrahepatic tissues. Apo E also serves as a ligand recognized by liver receptors for chylomicron remnants.

Apo E exists in three primary isoproteins which differ in size and/or charge. Apo E-2 has two cysteines replacing two arginines from apo E-4 and one arginine from apo E-3. Apo E has the lowest affinity for the

hepatic receptors. The closely-related apo E-3 and apo E-4 proteins have increasingly greater affinity with hepatic receptors.

Evolutionary Family of Apolipoproteins

Chan, et al. suggested that the apolipoproteins represent a gene family, with Apo A-I the primordial apolipoprotein (3). According to this concept, the apolipoprotein represents inter-related families that have evolved through a series of complete or incomplete gene duplications, primarily involving the amphipathic helical regions of the protein. In 1977, Barker and Dayhoff predicted that the amphipathic helical regions were repeated throughout the various apolipoproteins (4). Their prediction was based on the structural knowledge of apoproteins A-I, A-II and C-III.

Apo A-I is the prototypic protein with amphipathic regions which are represented in the mRNA or cDNA by 66 bp tandem DNA repeats (5). This translates into amphipathic helical regions of 22 amino acid residues, which may be subdivided into regions of 11 amino acids. The presence of a glutamic acid at residue 13 of the 22 unit amphipathic helix may be necessary for LCAT activation. The separate amphipathic helical regions tend to begin or end with a protein which serves as an alpha-helix breaker.

Apos A-I and C-III, on chromosome 11, are on opposite strands of DNA. Apo A-IV is downstream from apo A-I. The gene for Apo A-II is on chromosome 1. Genes for apo E and apo C-I are found on chromosome 19. Furthermore, there is extensive homology between the amphipathic helical regions of apoproteins A-I, A-IV and E. Homology has also been shown between the 5' region of apo A-I and a segment of apo C-II. If one lines up the mRNAs of these various apolipoproteins so that there is an approximate alignment between the regions coding for the protein, the introns have a remarkable degree of similarity in their location.

Apo B is quite different in its structure and whether it is part of the same apolipoprotein gene family is not yet determined.

HDL Function: Reverse Cholesterol Transport

The postulated role of apo A-I in reverse cholesterol transport as follows: apo A-I and its HDL particle are taken up by receptors on the cell's surface, but they do not participate in intracellular degradation. Instead, cholesterol is removed from the cell and transferred to particles. Since apo A-I is an activator of LCAT, it can also promote the conversion of cholesterol to cholesteryl ester, a necessary step in the transformation of nascent to mature HDL.

Some HDL particles contain only apo A-I. Fielding (6) has recently

described a particle with only apo A-I and pre-beta electrophoretic mobility which he believes may be involved in reverse cholesterol transport. Some HDL particles contain apo A-I and apo A-II, while others also contain apo E. HDLs with apo E may transfer cholesteryl ester directly to the liver. The cholesteryl ester in the HDL particles without apo E is transferred to VLDL, IDL, or LDL by the CETP.

As mentioned earlier, Apo A-I is synthesized by the intestine and liver, while Apo A-IV is synthesized by the intestine, and apolipoproteins A-II and E by the liver. These apolipoproteins are secreted with phospholipid, and form the nascent HDL particle. The nascent HDL particle is then converted to a mature particle as cholesterol is removed from tissues and by converted cholesteryl ester.

In one study, measurements of apo A-I were reported to be superior to those of lipoprotein cholesterol in predicting coronary disease, but this generally has not been the case.

HDL deficiency, Apo A-1 mutants and polymorphisms

A fascinating disorder, in which a portion of the chromosome 11 coding region for Apo C-III was displaced into the coding region for apo A-I, was reported in two sisters. This mutation severely disrupted the synthesis of both apolipoproteins A-I and C-III. As a result, the two sisters had essentially no HDL and overwhelming atherosclerosis.

There are other diseases associated with deficiencies of HDL, not all of which produce severe overwhelming atherosclerosis. The classical disease in which HDL deficiency occurs is Tangiers' Disease (7). It has recently been suggested that the defect in Tangiers' Disease is due to a defect in HDL. Coronary disease certainly occurs in patients with Tangiers' Disease but generally not until middle age. The deposition of cholesteryl esters occur mainly in the reticular endothelial cells of Tangier's patients resulting in orange tonsils, hepatosplenomegaly, and the deposition in the Schwann cells. The latter may contribute to the neuropathy of this disease. While levels of apo A-I and HDL are extremely low in Tangiers' Disease, it's possible that the HDL that is present can function in reverse cholesterol transport.

An HDL deficiency called apo A-I Milano is linked to a substitution of cysteine at residue 173. This HDL deficiency is associated with low levels of HDL, but not with accelerated coronary disease. In fact, in one Italian town where the apo A-I Milano mutant is common, many affected individuals have been reported to live into their 80's and 90's. It may be that with apo A-I Milano, HDL's are more rapidly catabolized. Thus, a low concentration of HDL in the blood does not necessarily mean a deficiency in reverse cholesterol transport.

HDL deficiency has been linked to corneal opacities as in Fish Eye Disease. Also, a primary deficiency of HDL has been described in other kindreds.

The variety of HDL mutants has been extensively reviewed by the Munster Laboratory in West Germany. Most of the HDL mutants are the result of amino acid substitutions which increase the negative charge and acidity or the positive charge and basicity of the protein.

The most frequent mutant is associated with a proline substitution causing depressed levels of HDL concentrations. Another mutation associated with a proline substitution involving residue 4 increases levels of pro apo A-I. In general, most of the mutations do not result in serious pathological conditions or in disruptions of HDL or apo A-I function.

Three prominent forms of apo A-I polymorphism have been described. In one form, the polymorphism occurs approximately 2.7 kilobases downstream from the A-I gene. In a second, the polymorphism is about 0.7 kilobases downstream. In a third, the polymorphism is approximately 8 kilobases upstream. Using the SST1 restriction enzyme, a group of investigators reported that the P2 allele was associated with a higher incidence of CHD and with HDL levels in the lowest 10th percentile of the population.

These findings have not been repeated in other populations. The inability to reproduce these results from one population to another raises the question of whether the mutations are really associated with CHD or dyslipidemia, or whether they indicate a genetic disequilibrium between genes. The development of a series of genetic markers to identify individuals who are susceptible to early atherosclerosis, and particularly to dietary-induced lipidemia, would be extremely valuable in targeting therapy and in identifying high-risk individuals early in life for intervention.

Apo B and CHD

The lipoprotein and apolipoprotein which have most clearly been associated with atherosclerosis are LDL and apo B-100. VLDL is thought to be secreted with one apo B-100 molecule and that this apo B-100 molecule is passed on down to LDL. Apo B-100 differs from the other apolipoproteins because it contains a greater quantity of beta or pleated sheet structure. The amphipathic helical structure is also present, but its relatively smaller content sets it apart from the other apolipoproteins. The high beta sheet content of apo B-100 contributes to the relative insolubility of the protein once its lipid constituents are stripped away. In this regard, the protein resembles amyloid which also has a high content of beta structure. Apo B also has a higher hydrophobicity than

the other apolipoproteins. The hydrophobicity of apo B and its beta-structure permit more of this apolipoprotein to be submerged below the surface of the LDL particle.

Yang, et al., (8) had proposed a model for apo B-100 on the surface of LDL which differs from the classical one (Figure 1). In their model, apo B exists as an extended structure so that portions of the apo B are on the surface while other portions are buried. Yang's structure is consistent with the relative distribution of peptides released with LDL is treated with trypsin. This finding would also explain the antigenicity of apo B with the carbohydrates on the surface of the apolipoprotein.

A major function of apo B-100 is to bind the LDL to the LDL receptors on the cell surfaces. Apo B-100 contains a region between approximately residues 3300 and 3800 that is very positively charged. This region of

FIG. 1

Source: Kodama T, Freeman M, Rohrer L, et al. Type I macrophage scavenger receptor contains a-helical and collagen-like coiled coils. *Nature*. 1990; 343: 535.

positive charge is thought to interact with a region of negative charge on the surface of the LDL receptor.

The structure of apo B-100 was solved simultaneously in three laboratories. Chen, et al., reported the structure of over one-half of the amino acids residues by direct sequencing which agreed with the C-DNA structure (9). These investigators also produced synthetic peptides of a putative lipid-binding region, which was able to bind to the B/E receptor (10). Synthetic peptides from other regions of apo B did not bind to the receptor. Scott, et al., had earlier pointed out that a homology between the putative binding regions between residues 140 and 150 of apo B-100 and apo E (11). Soria, et al., have identified a mutant at amino acid 3500 in the binding region, which reduces binding to the LDL receptor, and results in moderate elevations of plasma LDL (12).

This finding of a defective apo B-100 illustrates that a patient may have an elevation of LDL either due to a defective or abnormal LDL receptor, as in familial hypercholesterolemia, or due to a defect in the binding region of the apolipoprotein.

However, apo B-48 appears to play a role in lipid transport in normotriglyceridemic patients. In one variant of abetalipoproteinemia, apo B-48 is present while apo B-100 is absent. Individuals with this disorder do not have the degree of malabsorption of dietary fat that is typically seen in patients with a classical form of abetalipoproteinemia.

Apo B-100 is required for the secretion of VLDL. In the absence of apo B-100, the liver fails to produce VLDL, and VLDL, IDL, and LDL are all missing from the blood. Individuals with classical abetalipoproteinemia or with homozygous hypobetalipoproteinemia have a total deficiency of apo B-100 or apo B-48, and subsequently of chylomicrons as well as VLDL, IDL and LDL.

In abetalipoprotein, the gene for apo B appears to be normal and in some cases apo B-48 may be shown in intestinal cells. The defect appears to be in assembling the intact chylomicron particle or VLDL particle in the liver.

Apo B-100 is also bound by a disulfide linkage to apo Lp(a) in Lp(a). The structure of Lp(a) has a high degree of homology with plasminogen. Concentrations of Lp(a) greater than 30 mg/dl have been associated with a much higher incidence of coronary disease. Lp(a) promotes atherogenesis by inhibiting fibrinolysis. Lp(a) may interact with the receptors on the surface of the endothelial cells and prevent their binding to plasmin. Lp(a) appears to contain two apo B proteins which are linked together in a disulfide linkage. Thus, combined with vitamins, Lp(a) has the potential for inhibiting thrombolysis and for carrying cholesterol. Interesting possibilities are raised for linking thrombosis and apolipoprotein metabolism in the pathogenesis of coronary heart disease.

Sniderman, et al., have described a condition common in CHD patients which they called hyperapo B (13). This trait appears to run in families and is characterized by an increased proportion of apo B-100 in VLDL, IDL and LDL. Hyperapo B is particularly common in patients who have normal or only slightly elevated levels of LDL and total cholesterol, but who suffer from premature coronary disease. Figure 2 shows that the levels of apo B are just as high in hyperapo B as in familial hypercholesterolemia. However, the concentration of LDL is much higher in patients with familial hypercholesterolemia.

There are several conditions which may be related to hyperapo B. One is a genetically-determined hyperlipidemia first described by Goldstein, et al., called familial combined hyperlipidemia or familial multiple lipoprotein-type hyperlipidemia (14). This appears to be a dominantly-inherited trait that may be caused by an overproduction of apo B and subsequently of VLDL. Early coronary disease and atherosclerosis are characteristics of familial combined hyperlipidemia. The lipoprotein pattern in this condition may be one in which LDL or VLDL is increased, or in which both LDL and VLDL are increased. Any one pattern may predominate in a given individual. Related conditions include dyslipidemic hyperlipidemia, recently described in Utah kindreds by Williams, et al. (15). They may also include syndrome x, in which hyperinsulinemia, insulin resistance and small, dense LDL particles are present.

FIG. 2. Data for plasma LDL cholesterol and B protein are compared between patients with coronary disease (○, group II) and those with type II hyperlipoproteinemia (●, group D). Although the LDL cholesterol values are lower in the coronary group, many of the patients with coronary disease have LDL B protein levels similar to those in patients with type II hyperlipoproteinemia.

Source: Sniderman A, Shapiro S, Marpole D, et al. Association of coronary atherosclerosis with hyperapobetalipoproteinemia [increased but normal cholesterol levels in human plasma low density (B) lipoproteins]. *Proc Natl Acad Sci USA* 1980; 77: 606.

Krause recently described the LDL particles in hypertriglyceridemic subjects as small, dense proteins enriched with apo B (16). It is speculated that these particles may be more atherogenic. The presumed mechanism for the formation of these dense LDL particles is analogous to that by which the HDL particle size and concentration is reduced in hypertriglyceridemia. When the concentration of lipoprotein lipase is diminished, CETP or LTP causes an increased transfer of triglyceride from the remnant particles into LDL and HDL in exchange for cholesteryl ester. In this exchange, the remnant particles become enriched in cholesteryl ester while the HDL or LDL particles become enriched in triglyceride. The triglyceride-enriched HDL or LDL particles are thought to be enhanced substrates for hepatic lipase. The triglyceride is hydrolyzed, producing small, dense LDL and HDL particles and decreased concentration of HDL. Prolonged postprandial lipemia would favor this process. In fact, it is well known that the concentration of HDL is inversely related to those of triglycerides and postprandial lipemia.

In theory, hyperapo B could result from an increased proportion of apo B in VLDL, as in familial combined hyperlipidemia, or from the exchange of triglyceride into LDL, which ultimately results in smaller, apo B-enriched LDL particles. The latter scenario is presumed to be the case in hypertriglyceridemia. Data are not available for determining whether the small, dense LDL particles have an impaired interaction with the LDL receptor or an increased interaction with the macrophage receptors which recognize oxidized LDL. Fruchart showed that in dysbetalipoproteinemia, some LDL particles contain apo C-III or apo E in addition to apo B, and that the apo C-III particles have diminished binding to the LDL receptor (17).

While the LDL binds readily to the LDL receptor through the interaction of apo B, it binds poorly to the scavenger or macrophage receptor. (Figure 3) However, LDL that has been modified in various ways readily binds to scavenger receptors. Such is the case when LDL is oxidized on reaction with malondialdehyde. This uptake may stimulate the conversion of macrophages to foam cells. The scavenger receptor may play a role in the development of the atherosclerotic plaque by promoting cholesterol uptake in an unregulated way. Its structure has been recently described by Kodama, et al., and contains helical and collagen-like coiled coils and exists as a trimeric membrane glycoprotein (18). Interestingly, probucol appears to block this uptake. In the watanabe rabbit, probucol retards the progression of atherosclerosis apart from any LDL-lowering factor. This may be due to an anti-oxidant effect of the drug, although the mechanism for this effect is not clear. What does seem clear is that the oxidation of LDL is caused by oxygen radicals. Steinberg suggested that the initial oxidation of LDL may be the effect of oxidized double-bonds in the polyunsaturated fatty acids, but that there may also be a

FIG. 3

breakdown of apo B and of phospholipid (19). Oxysterols are very potent inhibitors of cholesterol synthesis, and may also be toxic to endothelial cells and smooth muscle cells.

In summary, apo B may be related to atherosclerosis in a number of ways, most of which are only poorly understood. As a carrier of cholesterol, the apolipoprotein directly contributes to atherogenesis. Alterations in the structure of apo B which affect its binding to the LDL receptor may also cause atherogenesis by increasing the concentration of LDL. A change in the concentration of apo B, itself, or its content within IDL and LDL as in hyperapo B, may increase its atherogenicity as well. In addition, the binding of apo B-100 to apo Lp(a) in Lp(a) may link together cholesterol transport thrombosis or an anti-thrombolytic effect. Finally, changes in the LDL structure through oxidation or other modification may result in an enhanced uptake of LDL by scavenger receptors or macrophages present in the arterial wall.

In conclusion, over the past 20 years we have learned a great deal about the structure and the function of the apolipoproteins. This knowledge has permitted us to test new hypotheses concerning their relationship to atherosclerosis. We believe that this knowledge will be of benefit in devising more rational approaches for the treatment and prevention of coronary heart disease, and its complications.

REFERENCES

1. Segrest JP, Jackson RL, Morrisett JD, Gotto AM, Jr. A molecular theory of lipid-protein interactions in the plasma lipoproteins. *FEBS Lett* 1974; 38: 247.

2. Chen SH, Habib G, Yang CY, et al. Apolipoprotein B-48 is the product of a messenger RNA with an organ-specific inframe stop codon. *Science* 1987; 238: 363.
3. Chan L, VanTuinen P, Ledbetter DH, et al. The human apolipoprotein B-100 gene: A highly polymorphic gene that maps to the short arm of chromosome 2. *Biochem Biophys Res Commun* 1985; 133: 248.
4. Baker WC, Dayhoff MO. Evolution of lipoproteins deduced from protein sequence data. *Comp Biochem Physiol* 1977; 57: 309.
5. Karathanasis SK, Zannis VI, Breslow JL. Isolation and characterization of the human apolipoprotein A-I gene. *Proc Natl Acad Sci USA* 1983; 80: 6147.
6. Fielding CJ. Monoglyceride hydrolase activities of rat plasma and platelets. Their properties and roles in the activity of lipoprotein lipase. *J. Biol. Chem.* 1981; 256: 876
7. Schmitz G, Assmann G, Robenek H, Brennhausen B. Tangier disease: A disorder of intracellular membrane traffic. *Proc Natl Acad Sci USA* 1985; 82: 6305.
8. Yang CY, Lee FS, Chan L, et al. Determination of the molecular mass of apolipoprotein B-100. A chemical approach. *Biochem J* 1986; 239: 777.
9. Chen SH, Yang CY, Chen PF, et al. The complete cDNA and amino acid sequence of human apolipoprotein B-100. *J. Biol. Chem.* 1986; 261:12918.
10. Sparrow JT, Sparrow DA, Culwell AR, Gotto AM, Jr. Apolipoprotein E: Phospholipid binding studies with synthetic peptides containing the putative receptor binding region. *Biochemistry* 1985; 24:6984.
11. Forgez P, Gregory H, Young JA, et al. Identification of surface-exposed segments of apolipoprotein B-100 in the LDL particle. *Biochem Biophys Res Commun* 1986; 140: 250.
12. Soria LF, Ludwig EH, Clarke HR, et al. Association between a specific apolipoprotein B mutation and familial defective apolipoprotein B-100. *Proc Natl Acad Sci USA* 1989; 86: 587.
13. Teng B, Sniderman AD, Soutar AK, Thompson GR. Metabolic basis of hyperapobetalipoproteinemia. Turnover of apolipoprotein B in low density lipoprotein and its precursors and subfractions compared with normal and familial hypercholesterolemia. *J Clin Invest* 1986; 77: 663.
14. Kita T, Brown MS, Bilheimer DW, Goldstein JL. Delayed clearance of very low density and intermediate density of lipoproteins with enhanced conversion to low density lipoprotein in WHHL rabbits. *Proc Natl Acad Sci USA* 1982; 79: 5693.
15. Williams RR, Hunt SC, Hopkins PN, Stults BM, Wu LL, et al. Familial dyslipidemic hypertension. Evidence from 58 Utah families for a syndrome present in approximately 12% of patients with essential hypertension. *JAMA* 1988; 259: 3579.
16. Krause BR, Newton RS. Apolipoprotein changes associated with the plasma lipid-regulating activity of gemfibrozil in cholesterol-fed rats. *J Lipid Res* 1985; 26: 940.
17. Lussier Cacan S, Bard JM, Boulet L, Nestruck AC, Grothe AM, Fruchart JC, et al. Lipoprotein composition changes induced by fenofibrate in dysbetalipoproteinemia type III. *Atherosclerosis* 1989; 78: 167.
18. Kodama T, Freeman M, Rohrer L, et al: Type I macrophage scavenger receptor contains a-helical and collagen-like coiled coils. *Nature* 1990; 343: 531.
19. Steinbrecher UP, Witztum JL, Parthasarathy S, Steinberg D. Decrease in reactive amino groups during oxidation or endothelial cell modification of LDL. Correlation with changes in receptor-mediate catabolism. *Arteriosclerosis* 1987; 7: 135.

DISCUSSION

Thorup: (Charlottesville) That was a beautiful paper. I'd like to ask a real world question. You referred to Moore's article in the *Atlantic Monthly*. That article infers that

there might be some sort of a conspiracy in this field. Since you are recognized as a national leader in this field, I wonder whether you have been plagued with requests for interviews, discussions, or articles about this paper and how you've responded.

Gotto: I suppose with a name like Antonio Gotto I could be accused of being a member of a mafia. Moore accuses a group of academic scientists of forming an alliance with the Heart Institute, the American Heart Association, the American Medical Association and various pharmaceutical companies, particularly Merck. He compares this to the military and industrial alliance. I have been inundated with calls from patients who have asked if they can forget about diet and have said that Moore says the medicine which we're giving them is liable to cause a heart attack. I think that Moore, perhaps unwittingly or wittingly, has caused a great deal of harm. The National Cholesterol Education Program has been a very vigorous one led by the Heartland Blood Institute along with the American Heart Association, the AMA and thirty some other organizations. Moore takes the position that there is no evidence that changing cholesterol is of any benefit, that you can't do anything about changing it with diet and that the drugs that change it may cause heart attacks or may cause you to die. My answer to this is that in carrying out this work, Moore, who was an investigative reporter and had previously worked on Gary Hart's senatorial staff as an investigator, didn't carry out much of an investigation. He didn't talk to anybody in the field who had viewpoints different from his. You will note that there are very few quotations in the article. There is a kernel of truth in most pieces like this, but every basic claim he makes in the article is erroneous.

Ingram (Minneapolis): I enjoyed your paper. One point of clarification: when you were talking about apo protein little a, you mentioned that it had some homology with plasminogen. Was that structural homology and if so, has there been any functional significance of that structural homology demonstrated?

Gotto: Plasminogen has a structure in it called Kringle because of its similarity to a Danish pastry. The first three Kringles are not present in apo little a and then Kringle 4 is reproduced 37 times and kringle 5 is reproduced once. There is some variability. At the present time the significance of this isn't known. There are some recent studies by others that show that plasminogen or that apo little a can compete with plasminogen for binding to endothelial receptors. Hypothetically, it could be an anti-thrombolytic agent, but it is interesting that it does bring together the thrombotic process or the anti-thrombolytic process with the cholesterol carrying process and we believe that they are intimately related in some way in the pathogenesis of atherosclerotic lesions.