PLASMA LIPOPROTEINS

COMPOSITION, STRUCTURE AND BIOCHEMISTRY

1. Composition and Structure

Lipoproteins are complex aggregates of lipids and proteins that render the lipids compatible with the aqueous environment of body fluids and enable their transport throughout the body of all vertebrates and insects to tissues where they are required. Because of their clinical importance, a very high proportion of research on lipoproteins deals with their functions in humans in relation to health, and the discussion that follows has a human bias. Lipoproteins are synthesised mainly in the liver and intestines. Within the circulation, these aggregates are in a state of constant flux, changing in composition and physical structure as the peripheral tissues take up the various components before the remnants return to the liver. The most abundant lipid constituents are triacylglycerols, free cholesterol, cholesterol esters and phospholipids (phosphatidylcholine and sphingomyelin especially), though fat-soluble vitamins and anti-oxidants are also transported in this way. Free (unesterified) fatty acids and lysophosphatidylcholine are bound to the protein albumin by hydrophobic forces in plasma and in effect are detoxified. The circulating lipoproteins are structurally and metabolically distinct from the **proteolipids** containing covalently linked fatty acids or other lipid moieties, which are described on another webpage.

Ideally, the lipoprotein aggregates should be described in terms of the different protein components or **apoproteins** (or 'apolipoproteins'), as these determine the overall structures and metabolism, and the interactions with receptor molecules in liver and peripheral tissues. However, the practical methods that have been used to segregate different lipoprotein classes have determined the nomenclature. Thus, the main groups are classified as chylomicrons (**CM**), very-low-density lipoproteins (**VLDL**), low-density lipoproteins (**LDL**) and high-density lipoproteins (**HDL**), based on the relative densities of the aggregates on ultracentrifugation. However, these classes can be further refined by improved separation procedures, and intermediate-density lipoproteins (**IDL**) and subdivisions of the HDL (e.g. HDL_1, HDL_2, HDL_3 and so forth) are often defined. Density is determined largely by the relative concentrations of triacylglycerols and proteins and by the diameters of the broadly spherical particles, which vary from about 6000Å in CM to 100Å or less in the smallest HDL. An alternative nomenclature is based on the relative mobilities on electrophoresis on agarose gels. Thus, α, pre-β and β lipoproteins correspond to HDL, VLDL and LDL, respectively. Some compositional details are listed in **Table 1**.

Table 1. Physical properties and lipid compositions of lipoprotein classes.

	CM	VLDL	LDL	HDL
Density (g/ml)	< 0.94	0.94-1.006	1.006-1.063	1.063-1.210
Diameter (Å)	6000-2000	600	250	70-120
Total lipid (wt%) *	99	91	80	44
Triacylglycerols	85	55	10	6
Cholesterol esters	3	18	50	40
Cholesterol	2	7	11	7
Phospholipids	8	20	29	46

* Most of the remaining material comprises the various apoproteins.

The data for the relative compositions of the various lipid components should not be considered as absolute, as they are in a state of constant flux, but in general the lower the density class, the higher the proportion of triacylglycerols and the lower the proportions of phospholipids and the other lipid classes. In fact, the VLDL and LDL exhibit a continuum of decreasing size and density.

The fatty acid compositions of the main lipid classes are listed in **Table 2**. As might be expected, the triacylglycerols tend to contain a high proportion of saturated and monoenoic fatty acids, while the phospholipids contain the highest proportion of polyunsaturated, specifically arachidonate. The cholesterol esters are enriched in linoleate, reflecting their biosynthetic origin (see below). Minor differences only occur for each lipid among the lipoprotein classes. The composition of the triacylglycerols of the chylomicrons (not listed) depends largely on that of the dietary fatty acids.

Table 2. Fatty acid compositions (wt% of the total) in the main lipids of human lipoprotein classes.*

	Triacylglycerols			Cholesterol esters			Phospholipids		
	VLDL	LDL	HDL	VLDL	LDL	HDL	VLDL	LDL	HDL
16:0	27	23	23	12	11	11	34	36	32
18:0	3	3	4	1	1	1	15	14	14
18:1	45	47	44	26	22	22	12	12	12
18:2	16	16	16	52	60	55	20	19	21
20:4(n-6)	2	5	8	6	7	6	14	13	16

* From Skipski, V.P. In: *Blood Lipids and Lipoproteins. Quantitation, Composition and Metabolism.* pp. 471-483 (ed. G.J. Nelson, Wiley-Interscience, New York) (1972).

The other important components of the lipoproteins are the apoproteins and the various types with their main (but not exclusive) lipoprotein associations, molecular weights and broad functions are listed in **Table 3**.

Table 3. The main properties of the apoproteins.*

Apoprotein	Molecular weight	Lipoprotein	Function
Apo A1	28,100	HDL	Lecithin:cholesterol acyltransferase (LCAT) activation. Main structural protein.
Apo A2	17,400	HDL	Enhances hepatic lipase activity
Apo A4	46,000	CM	
Apo AV(5)	39,000	HDL	Enhances triacylglycerol uptake
Apo B48	241,000	CM	Derived from Apo B100 – lacks the LDL receptor
Apo B100	512,000	LDL, VLDL	Binds to LDL receptor
Apo C1	7,600	VLDL, CM	Activates LCAT
Apo C2	8,900	VLDL, CM	Activates lipoprotein lipase
Apo C3	8,700	VLDL, CM	Inhibits lipoprotein lipase
Apo D	33,000	HDL	Associated with LCAT, progesterone binding
Apo E	34,000	HDL	At least 3 forms. Binds to LDL receptor
Apo(a)	300,000-800,000	LDL, Lp(a)	Linked by disulfide bond to apo B100 and similar to plasminogen
Apo H, J, L			Poorly defined functions
Apo M		HDL	Transports sphingosine-1-phosphate

* Roman numerals are often used to designate apoproteins (e.g. Apo AI, AII, AIII, etc)

In general, apoproteins consist of a single polypeptide chain often with relatively little tertiary structure, and they are required to solubilize the non-polar lipids in the circulation and in some instances to recognise specific receptors.

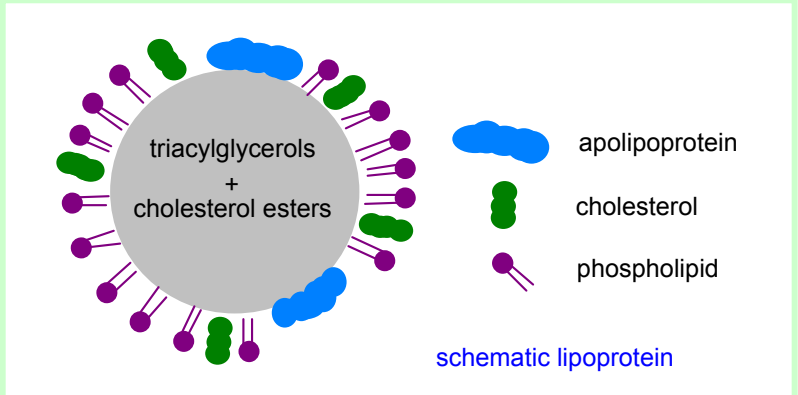
Apo B100 and apo B48 are large and water-insoluble and they are the only non-exchangeable apoproteins, which are assembled into triacylglycerol-rich lipoproteins with their lipid components in the intestines or liver. Cholesterol esters are required for proper folding of apo B. With 4536 amino acid residues, apo B100 is one of the largest monomeric proteins known; apo B48 represents the *N*-terminal 48% of apo B100. These stay with their lipid aggregates during their passage in plasma and the various metabolic changes that occur, until they are eventually removed via specific receptors. The remaining soluble or exchangeable apoproteins, such as apo E, apo A4, apo C3, apo A5, and apo A1, are much smaller in molecular weight and can exchange between lipoprotein classes and acquire lipids during circulation.

Apo A1 is the main protein component of HDL, and is synthesised within the liver (70%) and intestine (30%). It is a 28-kDa single polypeptide consisting of 243-amino acids, which has no disulfide linkages or glycosylation. Apart from the 44 amino acid *N*-terminal region, the protein is arranged as eight α-helical segments of 22 amino acids with two 11-mer repeats, and in some instances these are separated by proline residues. It is believed that the helices are amphipathic, each with a hydrophobic face that interacts with lipids and a polar face that interacts with the aqueous phase. The molecule probably exists in several conformational forms. Apo A2 is the second most important HDL apolipoprotein, and it exists as a homodimer with two polypeptide chains, each 77 amino acids in length and linked by a disulfide bond. Human apo A4 is the largest member of the exchangeable apolipoprotein family and is a 376-amino acid glycoprotein, which is synthesised in intestinal enterocytes and secreted as a constituent of chylomicrons

Apo E is an *O*-linked glycoprotein in three isoforms and is synthesised by many tissues including liver, brain, adipose tissue, and artery wall, but most is present in plasma lipoproteins derived primarily from the liver. It is involved in many aspects of lipid and lipoprotein homeostasis, both for the triacylglycerol-rich lipoproteins and HDL, but is also important for lipid metabolism in brain and other tissues. In addition, it is believed to have some non-lipid related functions, for example on immune response and inflammation.

Apo D is atypical in that it is very different in structure from other apolipoproteins, and it is expressed widely in mammalian tissues (most others are produced mainly in liver and intestine). In plasma, it is present mainly in HDL and to a lesser extent in LDL, where it may function as a multi-ligand binding protein capable of transporting small hydrophobic molecules such as arachidonic acid, steroid hormones, and cholesterol for metabolism or signalling.

Lipoproteins are spherical (VLDL, LDL, HDL) to discoidal (nascent HDL) in shape with a core of non-polar lipids, triacylglycerols and cholesterol esters, and a surface monolayer, ~20Å thick, consisting of apoproteins, phospholipids and non-esterified cholesterol, which serves to present a hydrophobic face to the aqueous phase, as illustrated schematically below.



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The physical properties of apoproteins enable them to bind readily at the interface between water and phospholipids, and specifically they bind to the phospholipids on the surface of the lipoprotein aggregates. In effect, this outer shell of amphipathic lipids and proteins solubilizes the hydrophobic lipid core in the aqueous environment. Each apolipoprotein, other than apo B100, tends to have a helical shape with a hydrophobic domain on one side that binds to the lipid core and a hydrophilic face that orientates to the aqueous phase. As the lipid compositions of the lipoproteins change during circulation throughout the body, the apoproteins are able to adapt to the altering affinities at the surface by changing conformation. For example, some have very little tertiary structure so are flexible, while apo A1 has a mobile or hinge domain. The polar nature of the surface monolayer prevents the lipoprotein particles from aggregating to form larger units. In addition, apoproteins have many different functions, some of which are listed in Table 3 (and are discussed further below). For example, some are ligands for receptors on cell surfaces and specify the tissues to which the lipid components are delivered, while others are cofactors for lipases or regulate lipid metabolism in the plasma in various ways.

LDL particles, for example, average 22 nm in diameter with roughly 3000 lipid molecules in total, and they contain a core of approximately 170 triacylglycerol, 1600 cholesterol ester and 200 unesterified cholesterol molecules. The surface monolayer has a single copy of apo B100 together with about 700 phospholipid and 400 free cholesterol molecules. Phosphatidylcholine, about 450 molecules, and sphingomyelin, about 185 molecules, are the main phospholipids, together with smaller numbers of lysophosphatidylcholine, phosphatidylethanolamine and other lipid molecules.

In contrast, the discoidal nascent HDL particles are believed to consist of a small unilamellar bilayer, containing approximately 160 molecules of phospholipid, which is surrounded by four apolipoprotein molecules, including at least two apo A1 monomers.

In addition to the apoproteins, lipoproteins contain a number of important enzymes, including lipases, acyl transferases, transport proteins and some with anti-oxidative or anti-inflammatory functions. Some are not concerned with lipid metabolism.

The lipoproteins can be categorised simplistically according to their two main metabolic functions. The principal role of the chylomicrons and VLDL is to transport triacylglycerols ‘forward’ as a source of fatty acids from the intestines or liver to the peripheral tissues. In contrast, the HDL remove excess cholesterol from peripheral tissues and deliver it to the liver for excretion in bile in the form of **bile acids** (‘reverse cholesterol transport’). While these functions are considered separately here for convenience, it should be recognised that the processes are highly complex and inter-related, and they involve transfer of apoproteins, enzymes and lipid constituents among the heterogeneous mix of all the lipoprotein fractions.

2. Lipoprotein and Triacylglycerol Metabolism

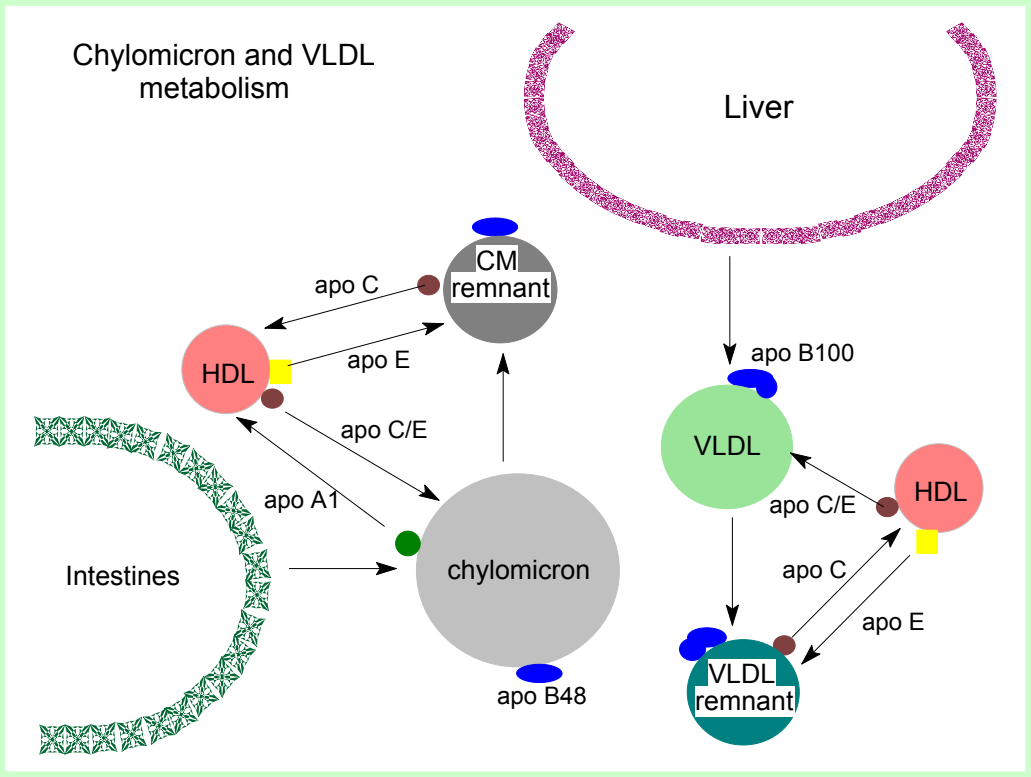
Triacylglycerols are the most energy-dense molecules available to the body as a source of fuel. For efficient transport from the intestine and the liver to other organs of the body, it is essential that they be packaged in a form compatible with the aqueous environment in plasma. Chylomicrons and VLDL are mainly involved, although some proteins that are shared with HDL are essential for the process to function normally. For example, exchangeable apolipoproteins protect triacylglycerol-rich particles from non-specific interactions in plasma and ensure that they have the correct configuration for action by lipases.

Dietary fatty acids and monoacylglycerols are absorbed by the enterocytes in the intestines, where they must cross the cytoplasm to the endoplasmic reticulum with the aid of fatty acid binding proteins. These are immediately utilized to form new triacylglycerols, and are thus detoxified (see our web page on **triacylglycerol biosynthesis**), mainly by the monoacylglycerol pathway. The triacylglycerols are incorporated together with dietary cholesterol and a small amount of,

cholesterol esters into chylomicrons particles with a surface layer of phospholipids to which is attached a single molecule of the truncated form of apo B, apo B48, which is diagnostic for triacylglycerol-rich lipoproteins of intestinal origin. The synthesis of apo B100 and its truncated form, and the accumulation of lipids to form chylomicrons or VLDL in intestinal cells and liver are complex processes that are still only partly understood. Simplistically, secretory proteins such as apo B are synthesised on ribosomes on the surface of the endoplasmic reticulum and translocated through the membrane to the lumen of the endoplasmic reticulum. VLDL are then assembled by accretion of lipids, for example with the aid of a microsomal triacylglycerol transfer protein, to the growing apo B core in three stages - pre-VLDL (pre-chylomicrons - nascent lipoproteins), VLDL2, a triacylglycerol-poor form of VLDL that is assembled in the Golgi and is transported to the basolateral membrane, where the final triacylglycerol-rich VLDL1 or chylomicrons are secreted by a process of reverse exocytosis into the intestinal lamina propria.

The chylomicrons are transported via the intestinal lymphatic system and enter the blood stream at the left subclavian vein. During circulation throughout the body, triacylglycerols are removed by the peripheral tissues by endothelial-bound lipoprotein lipase with entry of fatty acids into muscle for energy production and adipocytes for storage. However, the apo B48 remains with the residual particle. The chylomicrons also contain some apo A1, which is only synthesised in the intestines, but this is transferred spontaneously to the HDL as soon as the chylomicrons reach the circulation. Transfer of apo E and apo C in the reverse direction from the HDL to the surface of the chylomicrons occurs at the same time. The depleted or 'remnant' chylomicrons, containing the dietary cholesterol, apo E and apo B48 mainly, eventually reach the liver where they are cleared from the circulation by a receptor-mediated process that requires the presence of apo E.

The triacylglycerols of the remnant chylomicrons, together with cholesterol and cholesterol esters, are secreted by the liver into the circulation in the form of VLDL, which contain one molecule of the full-length form of apo B, apo B100. In addition, an appreciable amount of triacylglycerol in VLDL is synthesised in the liver from free fatty acids reaching it from adipose tissue via the plasma in the post-absorptive and fasted states. In effect, VLDL serve to buffer the plasma free fatty acids released following lipolysis in adipose tissue in excess of the requirements of muscle and liver.



Within the liver, the nascent VLDLs are synthesised in the endoplasmic reticulum and are transported to the Golgi in a complex multistep process, involving a specific VLDL transport vesicle. In the lumen of the *cis*-Golgi lumen, VLDLs undergo a number of essential modifications before they are transported to the plasma membrane and secreted in the circulatory system. The surface layer of the newly synthesised VLDL is enriched in phosphatidylethanolamine, which rapidly exchanges with the phosphatidylcholine of other lipoproteins. The newly synthesised VLDL contain a little apo C, but they rapidly take up more (10-20 molecules of apo C2) and apo E from HDL after a few minutes in the circulation. The small amount of apoA1 of intestinal origin is transferred to HDL.

Lipoprotein lipase, the key enzyme in the peripheral tissues that is responsible for the hydrolysis of triacylglycerols from the chylomicrons and VLDL, is bound to heparin sulfate-proteoglycans on the vascular surface of the endothelial cells of the capillaries of adipose tissue, muscle and lactating mammary gland, primarily. Apo C2 is an absolute requirement for activation of the enzyme. It is believed that apo C2 opens a lid-like region of the enzyme, permitting the active site to hydrolyse the fatty acid ester bonds of the triacylglycerols. Several molecules of the enzyme, each activated by one molecule of apo C2, become attached to the surface of the chylomicron/VLDL particles simultaneously during lipolysis. Apo AV is also stimulatory, probably via an interaction with a small glycosylphosphatidylinositol-anchored glycoprotein, designated GPIHBP1, which is essential for lipoprotein lipase activity. GPIHBP1 is located in capillary endothelial cells where it binds the chylomicrons and LDL.

Lipoprotein lipase hydrolyses the primary ester linkages (positions *sn*-1/3) in triacylglycerols to produce free fatty acids and 2-monoacylglycerols. Then, the latter isomerize spontaneously to form 1/3-monoacylglycerols, which can be hydrolysed also by the enzyme. However, monoacylglycerols can also be taken up directly by cells and are not found in the remnant lipoproteins or bound to circulating albumin. As the transport of VLDL particles progresses, the core of triacylglycerols is reduced and the proteins and phospholipids on the surface are transferred away to the HDL. However, sufficient apo C2 remains to ensure that most of the triacylglycerols are removed. As partially delipidated lipoproteins are detected in the circulation, it is believed that there is a process of dissociation and rebinding to the enzyme, during each step of which triacylglycerols are hydrolysed and apo C2 is gradually released with formation of remnant particles. Lipoprotein lipase is also involved in the non-hydrolytic uptake of esters of cholesterol and retinol, possibly by facilitating transport.

In contrast, apo C3 inhibits lipoprotein lipase and the binding of lipoproteins to receptors at the cell surface. It has a controlling influence on the turnover of triacylglycerols, and high levels have been correlated with elevated levels of blood lipids (hypertriglyceridemia).

Some only of the unesterified fatty acids resulting from the action of lipoprotein lipase are taken up immediately by the cells, where they can be used for energy purposes or for the synthesis of other lipids. The remainder is bound to circulating albumin from which it is slowly released to meet the cellular requirements. The glycerol produced is transported back to the liver and kidneys, where it can be converted to the glycolytic intermediate dihydroxyacetone phosphate. In muscle tissue, much of the fatty acids taken up are oxidized to two-carbon units, but in adipose tissue triacylglycerols are formed for storage purposes. During fasting, hormone-sensitive lipase releases fatty acids from the triacylglycerols stores and they are transported back into the circulation. Some lipoprotein lipase is present in the circulation, where it can continue to degrade the chylomicrons/VLDL, and during lactation the enzyme is very active in the mammary gland.

Eventually, a high proportion of the VLDL remnants (or 'IDL'), with apo B100 and apo E as the remaining proteins, are converted to LDL with further loss of triacylglycerols. Both apoproteins are required for recognition of the VLDL remnants and LDL by the LDL receptors in the liver. After forming a complex with the receptor, the LDL and some of the VLDL remnants, the latter

containing most of their original cholesterol ester and retinol ester contents, are taken up by the liver by a process of endocytosis and catabolized.

The main liver LDL receptor is a polypeptide of 839 amino acids that spans the plasma membrane and has an extracellular domain, which is responsible for binding to apo B100 and apo E. A domain within the cell is responsible for the clustering of these receptors into regions of the plasma membrane known as 'coated pits'. After binding of the LDL to the receptor, the complexes are internalized and then dissociated by means of an ATP-dependent proton pump, which lowers the pH in the endosomes, enabling the receptors to be recycled to the plasma membrane. The LDL-containing endosomes fuse with lysosomes, and lipolytic enzymes release free fatty acids and cholesterol, while acid hydrolases degrade the apoproteins. Much of the apo E is believed to escape this process and is returned to the circulation and the HDL.

3. VLDL, LDL and Cholesterol Metabolism

LDL are the main carriers of cholesterol to the adrenals and adipose tissue, where there are receptors requiring apo B100 that are able to take in the LDL by a similar process to that occurring in liver. Within these tissues, the cholesterol esters are hydrolysed to yield free cholesterol, which is incorporated into the plasma membranes as required. Any excess cholesterol is re-esterified by an acyl-CoA-cholesterol acyltransferase for intracellular storage. Other peripheral tissues have much lower requirements for cholesterol, but that delivered by the LDL may be helpful in suppressing synthesis of cholesterol *de novo* within cells. It may also inhibit the expression of lipoprotein receptors.

Cholesterol is essential to provide hydrophobicity to the VLDL so that they can carry triacylglycerols efficiently in the aqueous environment of plasma. However, once this has been accomplished the cholesterol-rich, triacylglycerol-depleted remnant LDL by-products are potentially toxic and must be safely removed from circulation. Some have argued that a significant part of the complexity of lipoprotein metabolism is concerned with the disposal of this LDL cholesterol before it can cause damage to the cardiovascular system. The liver is able to scavenge chylomicron remnants much more rapidly than LDL particles, and it seems likely that this specificity has evolved because the former are especially atherogenic. Therefore, further mechanisms such as that involving the HDL discussed next are required to return the excess LDL cholesterol to liver.

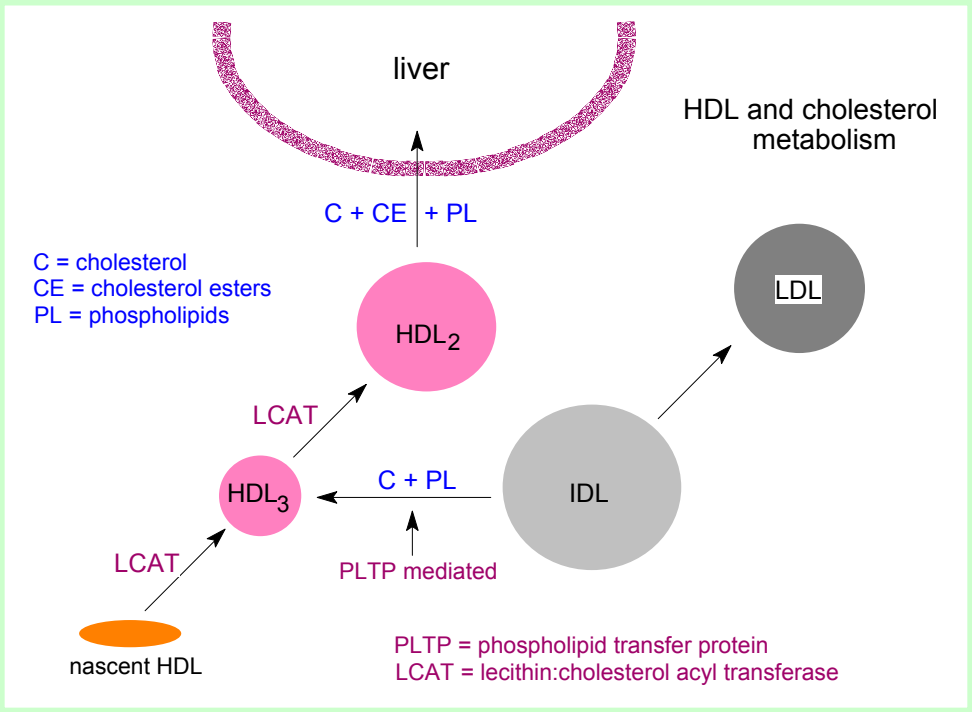
4. High-Density Lipoprotein and Cholesterol Metabolism

HDL are the most complicated and diverse of the lipoproteins, as they contain many different protein constituents, whose main purpose is to enable secretion of cholesterol from cells, esterification of cholesterol in plasma, transfer of cholesterol to other lipoproteins, and the return of cholesterol from peripheral tissues to the liver for excretion – a process that has been termed 'reverse cholesterol transport'. In addition, they have an important function in triacylglycerol transport by facilitating the activation of lipoprotein lipase, in the transfer of triacylglycerols between lipoprotein classes, and in the removal of chylomicron remnants and VLDL enriched in triacylglycerols. As well as the apolipoproteins, other key components of HDL include anti-oxidative enzymes and phospholipid transfer proteins. As many of these HDL lipid and protein constituents are exchangeable with other lipoproteins, many different types of HDL particle are generated with differing metabolic roles.

The nascent HDL are synthesised in the extracellular space of the liver and small intestine as protein-rich disc-shaped particles. Apo A1 synthesised in the liver together with that released spontaneously from chylomicrons is a key molecule that binds to phospholipids with a little cholesterol of cellular origin. It has been described as the scaffold for HDL assembly and is secreted as pro-apo A1, which is rapidly cleaved by a circulating metalloproteinase to generate the

mature polypeptide. The apoproteins apo C1, apo C2 and apo E also accumulate in HDL. It is believed that the origin of the cholesterol lies in **caveolae** (a type of membrane raft) in the plasma membrane. A specific transporter molecule 'ABCA1' facilitates the transfer of phospholipids and cholesterol to lipid-poor apoproteins, especially apo A1, in the nascent HDL particles.

The further development of mature HDL is dependent on the enzyme lecithin:cholesterol acyltransferase (LCAT), present mainly in the plasma compartment of the circulation. It requires apo A1 for activation. Our webpage on **cholesterol esters** contains a description of the mechanism of action of this enzyme, but in brief it transfers a fatty acid from position *sn*-2 of **phosphatidylcholine** ('lecithin') to the hydroxyl group of cholesterol, resulting in the formation of cholesterol esters and lysophosphatidylcholine. The cholesterol esters are highly hydrophobic and accumulate in the core of the HDL, while the lysophosphatidylcholine is removed by binding to albumin. By this means LCAT enables the nascent HDL to continue to draw free cholesterol and phospholipids from IDL and LDL, the latter mediated by a phospholipid transfer protein, until spherical HDL particles are formed, i.e. HDL_3 and HDL_2, with a surface coat of phospholipid, free cholesterol and apolipoproteins. Early in the formation of HDL, apo A2 is secreted from the liver and added to the surface. While its function is uncertain, it has been suggested that it may provide structural integrity to the particles. As the HDL grow, they acquire apo A4 and apo A5, the three apo C proteins and apo E from the VLDL and chylomicrons.



Cholesterol (but not cholesterol esters or phospholipids) is also obtained by extraction from cell surface membranes to the spherical HDL using the 'ABCG-1' transporter, a protein that is related to but distinct from the ABCA-1 transporter that is utilized by the nascent HDL (both are members of a large family of trans-membrane proteins). As a result, the levels of intracellular cholesterol are reduced, as cholesterol stored in cells in the form of cholesterol esters is mobilized to replace that removed from the plasma membrane. The removal of excess cholesterol from macrophage-derived foam cells present in atherosclerotic plaques is of particular importance.

The entire HDL particle can enter the hepatocytes through an apo A1 receptor interaction, where it undergoes a facilitated transfer of cholesterol and cholesterol esters to distinct pools within the cell. The modified HDL are secreted back into the circulation where they can acquire further cholesterol before returning to the liver. In a second less-efficient pathway, the added apo E in the HDL facilitates their uptake and catabolism by a process of endocytosis via a specific receptor, similar to

that described above for LDL, which results in the degradation of all the HDL constituents. This completes the process of 'reverse cholesterol transport'. Some of this cholesterol is converted to **bile acids** and exported into the intestines to aid digestion. As a proportion of these are eventually excreted, it is a means of reducing total amount of cholesterol in the body. The apo A1 recycles extracellularly between lipid-poor (pre-beta) and lipid-rich (spheroidal) lipoproteins.

In addition, cholesterol esters of HDL can be transferred to VLDL and LDL by the action of an enzyme associated with HDL, the cholesterol ester transfer protein (CETP). This means that excess cellular cholesterol can be returned to the liver by the LDL-receptor pathway as well as by the HDL-receptor pathway. Conversely, cholesterol esters can be exchanged for triacylglycerols by CETP from VLDL to HDL and vice versa. Once in the HDL, triacylglycerols are rapidly broken down by various lipolytic enzymes, though the physiological significance of this in health terms at least is a matter of debate.

In contradiction to the standard explanation for cholesterol elimination, experiments with animal models now suggest that a significant amount of cholesterol is secreted directly into the intestines by a process known as trans-intestinal cholesterol efflux. How this occurs and the relevance to human metabolism are under active investigation.

There are a number of biochemists who are now suggesting that too much weight may have been given to the role of HDL in cholesterol metabolism at the expense of other beneficial effects. While an important function of HDL is to act as a circulating store of apo C1, apo C2 and apo E, 60% of the proteins in HDL are not concerned with transport but have a protective role against cardiovascular disease, for example by acting as anti-inflammatory regulators to limit the activity of pro-inflammatory cytokines. HDL also carries an enzyme that hydrolyses platelet activating factor (PAF-acetyl hydrolase), which is a potent phospholipid mediator with pro-inflammatory properties. HDL prevents the oxidation of LDL and limits the concentrations of oxidized components, which might otherwise render them atherogenic. Thus, human serum paraoxonase is a calcium-dependent enzyme associated with HDL, which catalyses the hydrolysis of oxidized fatty acids from phospholipids and prevents the accumulation of oxidized lipids in lipoproteins, especially LDL. In addition, HDL transports small RNAs, hormones, carotenoids, vitamins and bioactive lipids. As it has the ability to interact with most cells and to deliver lipid-soluble cargo, HDL has the capacity to affect innumerable biological processes other than those concerned with cholesterol metabolism.

5. Lipoproteins and Disease

While only a few persons carry inherited defects in lipoprotein metabolism, such as hyper- or hypolipoproteinemias, abnormal lipoprotein metabolism is often observed as a secondary effect of diabetes, hypothyroidism and kidney disease. In Tangier disease, patients have mutations in both copies of the genes that code for the ABCA1 transporter protein (see previous section), so they have very little circulating HDL.

The role of lipoproteins in the metabolism of triacylglycerol and cholesterol in relation to cardiovascular disease is highly complex and contentious, and cannot be treated at length here. It is a subject best left to clinical specialists. Suffice it to say that there are a number of epidemiological studies that have demonstrated that low concentrations of HDL cholesterol are associated with a higher risk of atherosclerosis. This is the so-called 'good cholesterol', which is being transported back to the liver for catabolism. It is believed that HDL may protect against atherosclerosis via the promotion of reverse cholesterol transport. Conversely, higher concentrations of LDL cholesterol have been associated with increasing severity of cardiovascular disease, although the experimental correlations are not as good as for HDL. This is the 'bad cholesterol'. The apolipoproteins and associated enzymes of HDL are believed to be important for the maintenance of health in many further ways, including antioxidant, anti-inflammatory and anti-thrombotic effects.

It has also been argued that the levels of apo B and of apo C3 in plasma may be good predictors of the risk of coronary heart disease. For example, apo B may mediate the interaction between LDL and the arterial wall, and this may initiate the development of atherosclerosis. Indeed, virtually all of the apo B-containing lipoproteins can pass through the endothelial layer of arteries and initiate atherogenesis, but the smaller LDL are especially atherogenic because they enter the plaques with relative ease and have a high content of cholesterol. Thus, they provide substrates that trigger plaque initiation and growth.

Lipoproteins are not able to cross the blood-brain barrier, but various brain cells are able to express lipoprotein receptors, lipid transporters and apoproteins, which are required for cholesterol turnover and HDL-biogenesis. Apo E is especially important for cholesterol metabolism in the brain, but excess of one specific form (apo E4) is associated with Alzheimer's disease. While apo E does not appear to affect overall levels of cholesterol and phospholipids in the central nervous system, it does appear to modulate cholesterol and phospholipid homeostasis in particular subcellular membrane compartments. In addition, apo E mediates sulfatide trafficking and metabolism in the brain, and it is seen as a target for therapeutic intervention in diseases of the central nervous system. Apo E is also believed to influence susceptibility to parasitic, bacterial, and viral infections, and in HIV-positive patients apo E4 may hasten the progression to AIDS.

Lipoproteins and HDL especially play an important role also in host defense as part of the innate immune system. Infection and inflammation induce the acute-phase response, which leads to many changes in lipid and lipoprotein metabolism and initially protects the host from the harmful effects of bacteria, viruses, and parasites, provided that the infections are not prolonged. For example, an important defensive function is the ability of HDL and other lipoproteins to bind the endotoxin lipopolysaccharides, which are primary constituents of the outer surface membrane of Gram-negative bacteria, and so neutralize their toxic effects.

6. Insect Lipoproteins

Insects have a distinctive but relatively simple lipoprotein metabolism that serves as a useful model system for comparative studies. The hemolymph, the circulatory fluid in insects, contains a single multifunctional lipoprotein termed lipophorin that transports lipids to wherever they are required for energy and other purposes. Rather than triacylglycerols, the main lipid components are 1,2-diacyl-*sn*-glycerols, together with hydrocarbons, phospholipids and sterols. High-density lipophorin (HDLp) is the main lipoprotein class and it is composed of two integral and non-exchangeable apolipoproteins, apolipophorin I (240 kDa) and apolipophorin II (80 kDa) (the insect equivalents of apo B), with approximately 50% of the lipoprotein mass comprised of lipids. This transports dietary lipids from the insect gut to the fat body, an organ that simplistically can be considered to combine the roles of the mammalian liver and adipose tissue, and thence to peripheral tissues. Rather than being immediately internalized and the constituents recycled as with the mammalian lipoproteins, lipophorin functions as a reusable lipid shuttle, which delivers lipids to storage or peripheral tissues before returning for another cycle of loading and unloading.

In insects that have a high energy demand, for example for prolonged flight muscle activity, triacylglycerols in the fat bodies are mobilized by conversion to 1,2-diacyl-*sn*-glycerols (rather than to free acids as in vertebrates). These are loaded onto the HDLp particles, decreasing their density to produce low-density lipophorin (LDLp). The increase in hydrophobicity of the particles is countered by the uptake of up to 16 molecules of the exchangeable apolipophorin III (similar to vertebrate apolipoproteins in function) from the hemolymph. When this lipoprotein reaches the flight muscles, the diacylglycerols are hydrolysed to free acids by the action of lipoprotein lipase, and a fatty acid-binding protein then facilitates fatty acid transport within the cell.

Recommended Reading

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William W. Christie

James Hutton Institute (and Mylnefield Lipid Analysis), Invergowrie, Dundee (DD2 5DA), Scotland

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