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Cholesterol metabolism

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Biological Functions of Cholesterol

Because of the well established predictive association between plasma cholesterol concentration and ischaemic heart disease, we are apt to think of cholesterol as a harmful substance that we might be better without. It must be remembered, however, that cholesterol is essential for the normal functioning of the animal body. Herbivorous animals must satisfy all their requirements for cholesterol by endogenous synthesis, and for some insects that are not equipped with the enzymes necessary for cholesterol synthesis, cholesterol is an essential food factor.

STRUCTURAL

Perhaps the most important role of cholesterol is that of an essential structural element of the membranes of all animal cells and subcellular particles. According to current ideas, the typical animal membrane consists basically of a mixed double layer of cholesterol and phospholipid molecules, arranged at right angles to the plane of the membrane so that the polar groups face outwards from both surfaces, with various other constituents embedded in the membrane. Esterified cholesterol cannot readily participate in membrane formation. Hence, since the bulk of tissue cholesterol is present in membranes, the cholesterol in most tissues is predominantly in free or unesterified form.

As well as forming cell membranes, cholesterol is a constituent of plasma lipoproteins. The free cholesterol of plasma lipoproteins is probably associated with protein and phospholipid to form a membrane-like structure covering the surface of the lipoprotein molecule and thus enabling much of the non-polar lipid of the plasma to be transported in soluble form. The possible role of cholesteryl ester in plasma is discussed below (see 'Transport of cholesterol').

BIOCHEMICAL

In addition to its structural role, cholesterol is an obligatory precursor of bile acids and of all the steroid hormones, including the sex hormones and the hormones made in the adrenal cortex.

Physiology of Cholesterol

EXCHANGEABLE CHOLESTEROL

The normal human body contains about 2 g of cholesterol per kg total weight. Much of this is exchangeable with the plasma cholesterol. Equilibration between plasma and tissue cholesterol is complex. In some tissues, such as liver and intestine, equilibration approaches completion within hours; in others, such as skin and artery, equilibration may not be achieved for days or weeks. Moreover, the proportion of the total cholesterol that is exchangeable varies from one tissue to another. For these reasons it is difficult, if not impossible, to measure the total amount of exchangeable cholesterol in the whole body. It is sometimes assumed that the total mass of exchangeable cholesterol must always be closely correlated with the plasma cholesterol concentration. Although there may well be a general tendency for this to be so, it is known that cholesterol can accumulate in the tissues with little or no change in plasma cholesterol concentration. In rats, for instance, cholesterol feeding may lead to the accumulation of cholesteryl esters in the liver without perceptible increase in plasma cholesterol level. Cholesterol balance studies on human subjects have also shown that cholesterol may enter or leave the tissues without significant change in the amount present in the plasma. In view of this, the effect of a drug or dietary modification on cholesterol metabolism as a whole cannot be assessed from its effect on the plasma cholesterol alone.

TURNOVER OF EXCHANGEABLE CHOLESTEROL

The exchangeable pool of cholesterol participates in an incomplete enterohepatic circulation, in which cholesterol is excreted via the bile duct into the duodenum where it mixes with dietary cholesterol. It is then partially reabsorbed from the jejunum, the unabsorbed fraction passing down the intestine to be excreted in the faeces. Some cholesterol is converted into bile acids in the liver before excretion in the bile. The bile acids are largely reabsorbed from the ileum, the unabsorbed fraction leaving the body

in the faeces. There is thus continual loss of cholesterol from the exchangeable pool, partly as cholesterol itself and partly as bile acids derived from cholesterol. In the steady state, when the mass of the pool remains constant, this loss is replaced by endogenous synthesis in the tissues together with absorption of dietary cholesterol.

The fractional rate of turnover of the pool, due to loss and replacement, is such that about 2% is renewed each day. Since the main channel for outflow from the pool is via the intestinal tract, the absolute rate of turnover, in g/day, can be estimated by measuring the daily faecal output of bile acids plus that of neutral steroids derived from the cholesterol pool. There may be small losses of cholesterol by excretion through skin and by conversion into steroid hormones, some of which may be lost in the urine, but these losses are very small compared with the main outflow via the intestine. In some human subjects, loss of neutral steroids may take place in the lower intestine by bacterial action, but this can be corrected for. Measurements of faecal steroid output, when so corrected, give values of 1 to 2 g/day for turnover of cholesterol in man, excretion of bile acids accounting for about half the total turnover.

SYNTHESIS

Almost all animal tissues synthesize cholesterol from acetyl-CoA. In adult animals the most actively synthesizing organs are liver and intestinal wall, these two tissues probably supplying over 90% of the plasma cholesterol of endogenous origin. In most tissues the rate of synthesis of cholesterol is determined by the capacity of hydroxymethylglutaryl-CoA (HMG-CoA) reductase (EC 1.1.1.34), the enzyme catalyzing an early and rate-limiting step in the biosynthetic sequence from acetyl-CoA to cholesterol. Hepatic HMG-CoA reductase is subject to induction and repression by several hormonal and dietary factors (see 'Regulation').

ABSORPTION

Cholesterol is absorbed from the jejunum in the presence of bile salts. After entering the cells of the intestinal mucosa, it is incorporated into chylomicrons, which enter the blood circulation via the lymphatic system. The proportion of the cholesterol in the food that is absorbed depends upon the intake of cholesterol and the amount of triglyceride in the diet, triglyceride tending to promote cholesterol absorption. When the continuous intake of cholesterol is less than about 300 mg/day, 40-60% is absorbed. If the intake is increased to 2 to 3 g/day, as little as 10% may be absorbed.

REGULATION

Several mechanisms have been described which must tend to minimize fluctuations in the amount of cholesterol in the body. The relative importance of these mechanisms differs widely in different species.

When laboratory animals are fed diets rich in cholesterol, hepatic synthesis of cholesterol is almost completely inhibited, owing to repression of the synthesis of HMG-CoA reductase. This provides the animal with a regulatory system by which decreased hepatic synthesis compensates for increases in the amount of cholesterol absorbed from the food.

Whether or not a similar control by negative feedback is present in man is a debatable question. Cholesterol balance studies suggest that the extent to which cholesterol synthesis is suppressed by dietary cholesterol varies from one individual to another and, possibly, from one race to another. Some human subjects show no evidence of suppression of endogenous synthesis when cholesterol intake is increased, whereas others show quite marked suppression. It would be of interest to know whether individual differences in the efficiency of this regulatory mechanism contribute to individual differences in plasma cholesterol concentration within populations.

In man, increased absorption of cholesterol is followed by increased excretion of cholesterol from the exchangeable pool. In some species, not including man, increased absorption of dietary cholesterol leads to increased conversion of cholesterol into bile acids. Increased conversion of cholesterol into bile acids is also brought about by interruption of the enterohepatic circulation of bile salts, both in laboratory animals and in human subjects. Bile salts returning to the liver from the intestine repress the formation of an enzyme catalyzing the rate-limiting step in the conversion of cholesterol into bile acids. When bile salts are prevented from returning to the liver, the activity of this enzyme increases and degradation of cholesterol to bile acids is stimulated. This effect may be exploited therapeutically in the treatment of hypercholesterolaemia by the use of unabsorbable resins which bind bile acids in the lumen of the intestine and so prevent their return to the liver.

EFFECT OF DIET AND DRUGS

The association between hypercholesterolaemia and atherosclerosis has led to intensive search for effective methods of lowering the plasma cholesterol concentration. Among the most effective agents in current use are diets with a high ratio of polyunsaturated to saturated fat and drugs such as clofibrate, cholestyramine, nicotinic acid, and neomycin. The mechanisms by which some of these

agents bring about their effect on plasma cholesterol level are far from clear, nor is it certain that in lowering the plasma cholesterol level they diminish the mass of cholesterol in the exchangeable pool and, in particular, in the walls of atherosclerotic arteries. Polyunsaturated fats probably stimulate the conversion of cholesterol into bile acids, but they may also cause a shift of cholesterol from plasma to tissues. Clofibrate has several effects on cholesterol metabolism, including increased excretion of cholesterol from the exchangeable pool, decreased secretion of lipoprotein by the liver and diminished hepatic synthesis of cholesterol, but it is not known which, if any, of these effects is mainly responsible for lowering the plasma cholesterol level. Cholestyramine is a bile-acid-binding resin that acts by preventing the return of bile acids from the intestine to the liver and so promoting cholesterol degradation (see 'Regulation'). Nicotinic acid, like clofibrate, has more than one effect on cholesterol metabolism; it inhibits cholesterol synthesis in the liver, and it diminishes the flux of free fatty acids from adipose tissue. This last effect may decrease the amount of fatty acid available to the liver for triglyceride synthesis and this may, in turn, reduce the amount of very-low-density lipoprotein (VLDL) available for carrying cholesterol into the plasma. Neomycin, an unabsorbable antibiotic, interferes with the absorption of cholesterol from the jejunum, possibly by preventing the formation of mixed micelles in the lumen of the intestine.

In considering the possible mode of action of any agent that leads to a change in plasma cholesterol concentration it must be remembered that cholesterol circulates in the plasma in lipoproteins. Therefore any change in cholesterol concentration will reflect an underlying change in lipoprotein concentration if, as is likely, the metabolism of the lipid moiety of a lipoprotein molecule is intimately linked with that of the protein moiety. Hence, an effect of a drug or dietary modification on plasma cholesterol metabolism may be secondary to an effect on the metabolism of the protein component of one or other of the plasma lipoproteins.

Transport of Cholesterol

Cholesterol is present in all plasma lipoproteins, but more than 60% of the total in plasma from a fasting human subject is carried in low-density lipoproteins (LDL). About two-thirds of the total plasma cholesterol is esterified with long-chain fatty acids, linoleic acid being the predominant fatty acid in man. The cholesteryl esters of plasma are in a state of turnover owing to their continual hydrolysis and resynthesis. Hydrolysis takes place in the liver, but synthesis occurs mainly in the plasma by transfer of a fatty acid residue from lecithin to free cholesterol. This reaction is catalyzed by a plasma enzyme known as lecithin: cholesterol acyltransferase or LCAT. The preferred substrate for human LCAT is high-density lipoprotein (HDL), and it seems likely that the bulk of the esterified cholesterol in the plasma is formed on HDL, cholesteryl ester then being transferred from HDL to LDL and VLDL, partly in exchange for triglyceride. Some of the cholesteryl ester on LDL may be formed directly by the LCAT reaction, but LDL is not a good substrate for LCAT.

As suggested above, the role of free cholesterol in plasma is probably to act as a structural constituent of plasma lipoproteins, one of whose functions is to transport triglyceride through the circulation. The biological function of the large amounts of esterified cholesterol present in plasma is more difficult to explain. The answer, suggested in principle by Glomset (1968), may lie in the fact that cholesterol is synthesized in most tissues of the body but can only be disposed of to any significant extent by the liver. There must, therefore, be some means available to the body for transporting cholesterol in bulk from extrahepatic tissues to liver.

Although it is not yet possible to describe in detail the three-dimensional form of a plasma lipoprotein, there is reason to believe that an LDL molecule consists of a micellar core of non-polar lipid including triglyceride and cholesteryl ester, surrounded by a liquid crystalline membrane of protein and polar lipids (free cholesterol and phospholipid).

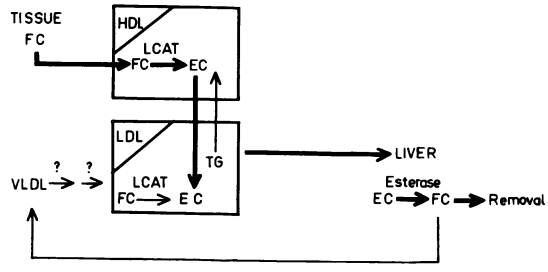

Fig. 1 *Scheme to suggest the role of LDL as a vehicle for transporting tissue cholesterol to the liver. Free cholesterol (FC) is transferred from tissues to HDL and is esterified to esterified cholesterol (EC) by the action of LCAT. EC formed on HDL is transferred to LDL. Some FC on LDL may also be esterified by LCAT. EC in LDL goes to the liver, where it is hydrolysed by an intracellular lipase, the FC so formed being removed by excretion in the bile, conversion into bile acids, or resecretion into the plasma after incorporation into lipoprotein. The path taken by cholesterol from tissues to liver is shown by the thick black line. Broken arrows show the probable conversion of VLDL into LDL.*

If this is so, it might be supposed that one of the functions of LDL is to transport cholesterol, in esterified form, from the tissues to the liver. On this view the function of LCAT is to generate esterified cholesterol from the free cholesterol formed in extrahepatic tissues. One might then envisage the sequence of events shown in figure 1. Tissue free cholesterol is transferred to high-density lipoprotein. It is then esterified by LCAT, enabling HDL to take up more free cholesterol. The esterified cholesterol formed in HDL is handed on to LDL, where it is incorporated into the non-polar core of the lipoprotein molecule. Low-density lipoprotein, carrying its load of cholesteryl ester, reaches the liver where the cholesteryl esters are hydrolysed. The free cholesterol so formed enters the pool of free cholesterol in the liver that is available for removal via the bile, conversion into bile acids, or re-incorporation into plasma lipoprotein.

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