Lipid Metabolism & Transport

Learning Objectives.

At the end of this course, you should be able to :

1. understand the basic methods of lipid transport;
2. appreciate the different sources of fatty acids for human function;
3. understand the basic structures of fatty acids, TAGs, and lipoproteins;
4. describe the roles of each of the classes of lipoprotein;
5. give an account of the endogenous and exogenous pathways of lipoprotein metabolism.

Lipids are the major and immediate source of energy in liver, muscle, kidney and other tissues. The term 'lipids' includes triacylglycerols (TAGs, or triglycerides), phospholipids and glycolipids.

TAG is constructed from fatty acids and glycerol by a process known as 'esterification'. Non-esterified fats are known as 'free' fatty acids (FFA) and are the major fuel for metabolism. They are an immediate source of energy, and their importance as metabolic fuels arises because their oxidation provides greater amounts of ATP than the oxidation of either carbohydrates or proteins.

Lipids also contribute to the make-up of cell membranes and intracellular membranes. They are also essential for maintaining lung alveoli integrity and cellular and metabolic regulation via the production of prostaglandins and steroid hormones.

In humans approximately 90% of fatty acids are supplied as dietary TAG, whilst the rest is made up of cholesterol, cholesteryl esters, non-esterified

fatty acids and phospholipids. TAGs are broken down to fatty acids via lipolysis.

Humans are unable to synthesise or desaturate polyunsaturated fatty acids (PUFA) with particular complex structures (e.g. linoleic and linolenic acids), and these are known as essential fatty acids. They are specifically required for the synthesis of eicosanoids (biologically active derivatives of arachidonic acid, found in cell membranes, with hormone-like functions) and also to act as second messengers (e.g. diacylglycerol). Essential fatty acids such as n-3PUFA and n-6PUFA, must be present in the diet (i.e. from plant or fish oils) or synthesised from other dietary fatty acids.

Dietary fats are digested by pancreatic enzymes (lipases) in the intestine to form FFAs, monoacylglycerols and glycerol, which are absorbed into the epithelial lining cells of the small intestine (enterocytes). The enterocyte re-esterifies the FFAs and monoacylglycerols to synthesise TAG. Chylomicrons are then assembled from the TAG, together with cholesterol, apolipoproteins and phospholipids.

Humans can synthesise fatty acids from intermediates products of the breakdown of sugars, some amino acids and other fatty acids, although this usually occurs only when there has been excessive intake of carbohydrate. Carbohydrate is converted to fatty acids in the liver and stored as TAG in adipose tissue.

Key Learning Points :

1. Lipids provide the major source of immediate energy for the body.
2. Lipids also are major contributors to cell membranes.
3. The diet is the main source of materials from which lipids are synthesised.

Fatty Acids

Lipids contain a mixture of fatty acids of different chain length, degree of saturation and branching, and must be transported by processes that are specific for varying chain lengths. In humans, lipids are catabolised only through oxidation, usually occurring in the mitochondria and peroxisomes.

FFAs are not water soluble. Their detergent effect would dissolve lipid cellular and intracellular membranes, so to avoid this they are transported wither bound to a plasma protein such as albumin, or esterified as TAG for transport between tissues and organs, and for storage. Each albumin molecule can bind up to eight fatty acid molecules.

In the small intestine, protein-bound short- and medium-chain fatty acids can diffuse directly into cells and from there into the hepatic portal circulation. Long-chain fatty acids are bound to fatty acid binding proteins to diffuse down a concentration gradient into cells. The presence of fatty acid-binding proteins within the cell facilitates cellular fatty acid uptake by reducing intracellular FFA concentration.

Triacylglycerols (Triglycerides, TAGs)

Storage of fatty acids in the form of TAG is more efficient for energy production than the storage of glycogen. TAGs constitute the major transport form of fat. They are formed by the esterification of one molecule of glycerol with three molecules of fatty acids. In both liver and adipose tissue, TAGs are produced by a pathway that involves the 3-carbon molecule glycerol 3-phosphate (gl-3-P).

The breakdown of adipose tissue TAG yields glycerol. This is transported to the liver for TAG synthesis. As a result of this adipo-hepatic axis, TAG stored in adipose tissue during times of dietary plenty can be transferred to the liver for metabolic remodelling to form VLDLs during times of starvation.

Triacylglycerols deliver fatty acids from adipose tissue stores to peripheral cells for catabolism. The hydrolysis of stored TAG is regulated by an enzyme called hormone-sensitive lipase. Lipolysis is activated by glucagon and adrenaline, but inhibited in the presence of insulin. Hormone-sensitive lipase removes the three fatty acids from TAG, and these liberated fatty acids and glycerol are then released to circulating blood.

Lipoproteins

Lipoproteins refer to complexes of cholesterol, TAG, and proteins that transport lipids in the aqueous environment of the blood stream.
There are 4 major classes of lipoproteins that are defined by density and size:

- CM (chylomicron)
- VLDL (very low density lipoprotein)
- LDL (low density lipoprotein)
- HDL (high density lipoprotein)
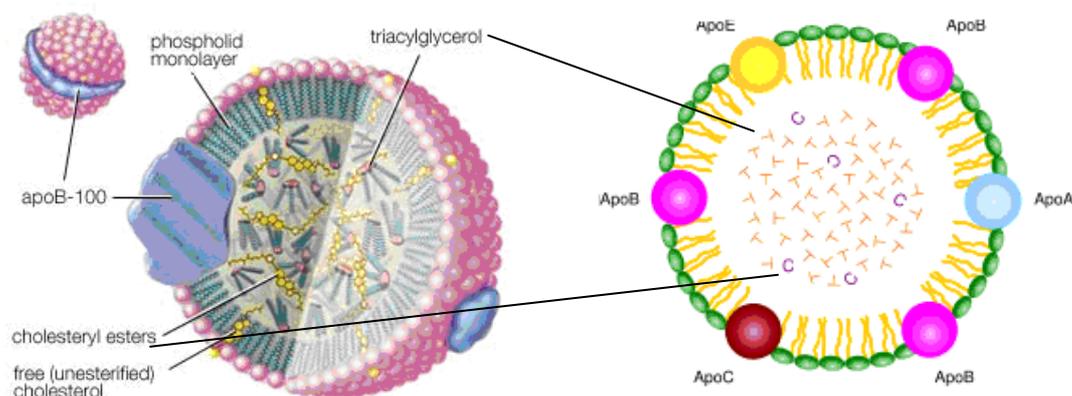
Lipoprotein particles are produced by hepatocytes (liver cells) and enterocytes from TAG, cholesterol, apolipoproteins, and phospholipids. The hydrophobic TAG and cholesteryl esters make up the central part, whilst apolipoprotein strands, cholesterol molecules and phospholipids are located in the outer shells. Hepatocytes assemble VLDLs and HDLs, and they are

distinguishable from each other by their differences in size, density, lipid composition and apolipoprotein content.

Apolipoproteins are protein components found on the outside of the lipoprotein, which emulsify the lipoprotein particle to make it more stable in aqueous solution for carriage in plasma. They also interact with cellular receptors that determine how and where lipoprotein particles are metabolised. Each lipoprotein particle has a specific set of apolipoproteins.

The important apolipoproteins are:

- ApoA - present in HDLs. The binding of ApoA-I to cellular receptors mediates the efflux of cholesterol from peripheral cells, and the influx of cholesterol into hepatocytes.
- ApoB - encourages cellular uptake of LDL. ApoB-100 is derived from liver and forms part of LDL. ApoB-48 is derived from the gut and is found in chylomicrons.
- ApoC - synthesised in the liver and is a peripheral activator of lipoprotein lipase (LPL). It is transferred between lipoproteins
- ApoE - stabilises VLDL for cellular uptake.
- Apo(a) - links with aAoB-100 to oxidise LDL, giving Lp(a) lipoprotein particles.

Apolipoproteins are important because they control lipoprotein metabolism. Apolipoprotein and LDL receptor genes have been identified, sequenced and mapped to chromosomes. Apolipoprotein disorders are known to lead to defects of lipid metabolism.

Lipoproteins vary in size and density, the denser being smaller. A high TAG content makes the lipoprotein particle less dense, and therefore larger in size. Increasing or reducing TAG content changes the protein : lipid ratio within the particle, and therefore it is possible to change density via enzyme activity.

Chylomicrons - The least dense and largest lipoproteins are chylomicrons, which are globules assembled by enterocytes following lipid digestion and composed principally of TAG, apoB-48, apoA-I and apoA-II, a smaller amount of cholesterol and cholesteryl esters. They transport products from dietary fat digestion to peripheral tissues. Progressive removal of TAGs by LPL leads to the formation of remnant particles, which are taken up and catabolised by the liver.

Very-low-density lipoprotein - VLDL is synthesised continuously in the liver. It is the main source of TAGs exported from liver to muscle and adipose tissues. TAGs make up around half the content of the particle, and apoB-100 is an essential component. ApoC-II and apoE are incorporated by transfer from HDL.

Intermediate-density lipoprotein - IDL is a particle remnant derived from VLDL by the removal of TAG by LPL, together with loss of apolipoprotein. IDLs are precursors of LDLs. IDL contains apoB-100 and apoE, but loses apoC-II.

Low-density lipoprotein - LDL is also a particle remnant, resulting from further removal of TAG in liver and peripheral tissues when the molecule is depleted of almost all of its TAG, and mainly consists of cholesterol. It contains a single apoB-100 molecule and some apoE. LDL is the main cholesterol carrier, delivering cholesterol to liver and peripheral tissues.

High-density lipoprotein - HDL particles are synthesised in both the liver and intestines. They carry cholesterol from adipose tissue directly to liver, and to tissues that synthesise steroid hormones. HDL also takes part in the metabolism of other lipoproteins by exchanging apolipoproteins, cholesteryl ester, TAG and phospholipids. HDL is rich in cholesteryl esters, and only around 5% of the particle consists of TAG. It contains apoA (A-I and A-II) and also apoC and apoE.

Lipoprotein metabolism

The handling of lipoproteins in the body is referred to as lipoprotein metabolism. It is divided mainly into exogenous and endogenous pathways, depending mostly on whether the lipoproteins are composed chiefly of dietary (exogenous) lipids or whether they originated in the liver (endogenous). There is also a third pathway, the reverse cholesterol transport pathway.
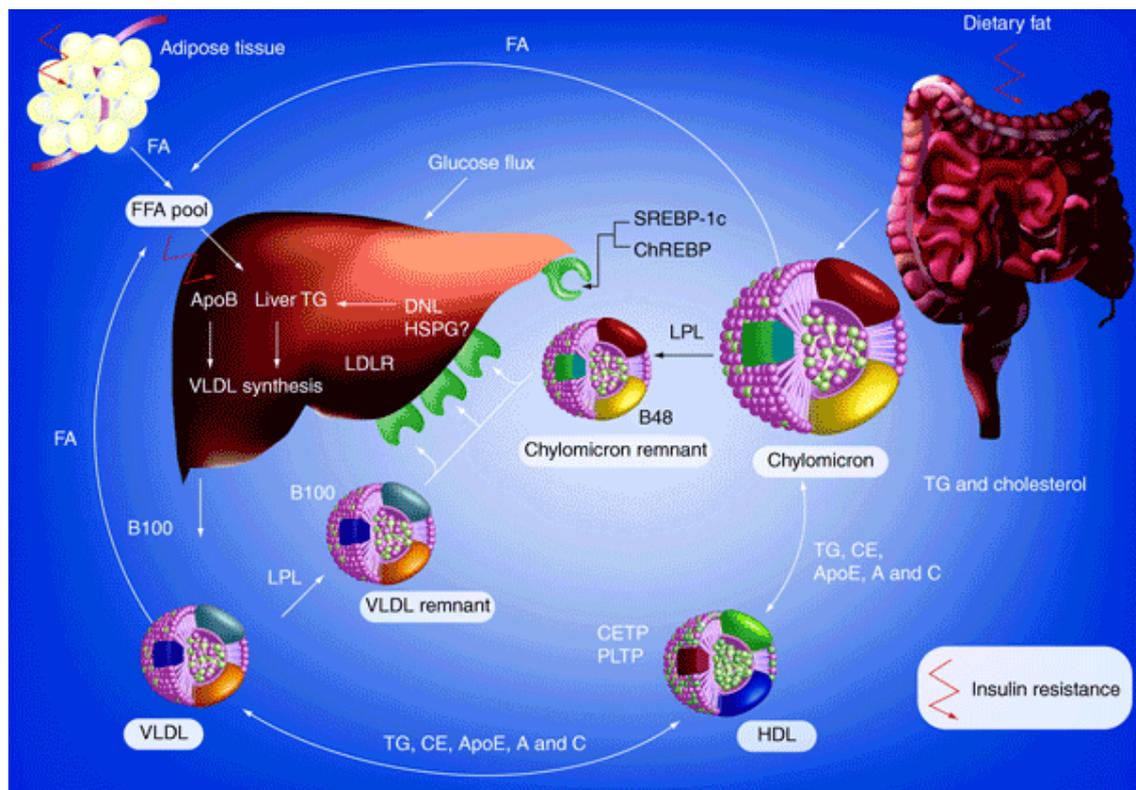
Exogenous pathway

Epithelial cells lining the small intestine (enterocytes) readily absorb lipids from the diet. These lipids, including TAGs, phospholipids, and cholesterol, are assembled with apolipoprotein B-48 into chylomicrons. These nascent (recently created) chylomicrons are secreted from the intestinal epithelial cells into the lymphatic circulation in a process that depends heavily on apolipoprotein B-48. As they circulate through the lymphatic vessels, nascent chylomicrons bypass the liver circulation and are drained via the thoracic duct into the bloodstream.

In the bloodstream, HDL particles donate apolipoprotein C-II and apolipoprotein E to the new chylomicron, after which point the chylomicron is considered mature. Via apolipoprotein C-II, mature chylomicrons activate lipoprotein lipase (LPL), which catalyzes the hydrolysis of TAG, ultimately releasing glycerol and fatty acids from the chylomicrons. Glycerol and fatty acids can then be absorbed in peripheral tissues, especially adipose and muscle, for energy and storage.

The hydrolysed chylomicrons are now considered chylomicron remnants. The chylomicron remnants continue circulating until they interact via apolipoprotein E with chylomicron remnant receptors, found chiefly in the liver. This interaction causes the endocytosis (engulfment) of the chylomicron remnants, which are subsequently hydrolysed within lysosomes. Lysosomal hydrolysis releases glycerol and fatty acids into the cell, which can be used for energy or stored for later use.

Endogenous pathway

The liver is another important source of lipoproteins, principally VLDL. TAG and cholesterol are assembled with apolipoprotein B-100 to form VLDL particles. Nascent VLDL particles are released into the bloodstream via a process that depends upon apolipoprotein B-100.

As in chylomicron metabolism, the apolipoprotein C-II and apolipoprotein E of VLDL particles are acquired from HDL particles. Once loaded with apolipoproteins C-II and E, the nascent VLDL particle is considered mature.

Again like chylomicrons, VLDL particles circulate and encounter LPL expressed on endothelial cells. Apolipoprotein C-II activates LPL, causing hydrolysis of the VLDL particle and the release of glycerol and fatty acids. These products can be absorbed from the blood by peripheral tissues, principally adipose and muscle. The hydrolysed VLDL particles are now called VLDL remnants or intermediate density lipoproteins (IDLs). VLDL

remnants can circulate and, via an interaction between apolipoprotein E and the remnant receptor, be absorbed by the liver, or they can be further hydrolysed by hepatic lipase.

Hydrolysis by hepatic lipase releases glycerol and fatty acids, leaving behind IDL remnants, called low density lipoproteins (LDL), containing a relatively high cholesterol content. LDL circulates and is absorbed by the liver and peripheral cells. Binding of LDL to its target tissue occurs through an interaction between the LDL receptor and apolipoprotein B-100 or E on the LDL particle. Absorption occurs through endocytosis, and the engulfed LDL particles are hydrolysed within lysosomes, releasing lipids, chiefly cholesterol.

Reverse Cholesterol Transport

Reverse cholesterol transport refers to the process by which cholesterol is removed from the tissues and returned to the liver. HDL is the key lipoprotein involved in reverse cholesterol transport and the transfer of cholesteryl esters between lipoproteins. The smallest and most dense lipoprotein particle is HDL. HDL is formed through a maturation process where nascent HDL is secreted by the liver and intestine and proceeds through a series of conversions (known as the "HDL cycle") to attract cholesterol from cell membranes and free cholesterol to the core of the HDL particle, although the exact mechanism for this not yet clear. It has been suggested that the action of cholesteryl ester transfer protein transforms HDL into a TAG-rich particle that interacts with hepatic-triglyceride lipase. Cholesterol ester-rich HDL may also be taken up directly by the receptors in the liver. Another mechanism may be that cholesterol esters are delivered directly to the liver for uptake without catabolism of the HDL cholesterol particle.

In relation to pathological change it has been accepted that higher levels of HDL are associated with lower levels of heart disease, therefore higher

levels of HDL are considered to be protective. In contrast, it is accepted that other lipoproteins, including VLDL, IDL, LDL, and the remnant particles rendered in lipid processing, are highly atherogenic. The term "non-HDL cholesterol" has been adopted to describe this increased risk reflected in the lipid profile that may not be otherwise identified by simply examining the LDL alone. Non-HDL cholesterol therefore encompasses a broader indication of cardiovascular disease risk.

Storage of Lipids

Fat metabolism is controlled primarily by the rate of stored TAG breakdown (lipolysis) although there is also local generation of fatty acids from the enzymatic breakdown of circulating lipoproteins. The fatty acids used for adipose tissue TAG synthesis are derived from TAG contained in chylomicrons and very-low-density lipoproteins (VLDLs), acted upon by the enzyme lipoprotein lipase (LPL). Adipose tissue LPL is found attached to the basement membrane glycoproteins of capillary endothelial cells. Apolipoprotein C-II must be present within the lipid particles to activate LPL. A deficiency in apolipoprotein C-II leads to increased plasma TAG levels. A rise in plasma insulin stimulates adipocyte LPL to release fatty acids from chylomicrons and VLDLs.

Key Learning Points :

1. Fatty acids are not water-soluble, and therefore must be transported as TAG, or attached to plasma protein.
2. TAGs constitute the major transport form of fat. They are formed by the esterification of one molecule of glycerol with three molecules of fatty acids. Breakdown of TAG will yield glycerol.
3. Lipoproteins refer to complexes of cholesterol, TAG, and proteins that transport lipids in the aqueous environment of the blood stream.
4. The four major classes of lipoprotein are chylomicrons, VLDL, HDL, and LDL.
5. Lipoproteins have apolipoproteins on their outer surface, increasing particle stability and controlling metabolism.
6. Lipoproteins contain TAG and cholesteryl esters. The higher the TAG content, the less dense the particle.
7. Lipoprotein metabolism occurs mainly via the endogenous or exogenous pathway, depending on the source of the lipoprotein.
8. Exogenous metabolism occurs when the source is dietary. Endogenous metabolism occurs when the source is hepatic.
9. Reverse cholesterol transport refers to the removal of cholesterol from the tissues and returned to the liver.