CHAPTER 7

Digestion and Absorption of Lipids

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COMMON ABBREVIATIONS

ACAT (acyl CoA:cholesterol acyltransferase)
FABP (fatty acid-binding protein)
SCP (sterol carrier protein)
DIETARY LIPIDS

Dietary lipids have been described as that part of the diet that can be extracted by organic solvents (Borgstrom, 1986). According to this definition, a variety of compounds qualify, including both nonpolar lipids such as triglycerides and polar lipids such as phospholipids. Although a variety of types of lipids are consumed in the diet, by far the greatest quantity of dietary lipids is in the form of triglycerides (triacylglycerols). Furthermore, most of these dietary triacylglycerols contain predominantly long-chain fatty acids (chain lengths of 14 to 20 carbons) esterified to the glycerol backbone. The average Western diet contains 100 to 150 g of dietary fat (triacylglycerols), which provides 900 to 1500 kcal, or about 40% of the calories consumed daily. (As discussed in Chapters 41 and 42, the American Heart Association and the Dietary Guidelines for Americans recommend that the intake of fat be reduced to 30% of calories to reduce the risk of atherosclerosis.) Dietary triacylglycerol is a major source of energy with a higher caloric density than the other macroconstituents, and also it is a source of essential fatty acids in the α and ω classes, mainly as linoleate (n-6, 18:2) and linolenate (n-3, 18:3). (See Chapter 15 for a discussion of essential fatty acids.)

Other dietary lipids include the fat-soluble vitamins A, D, E, and K (micronutrients discussed in Chapters 24 through 27), cholesterol and cholesteryl esters, and phospholipids. The amount of cholesterol/cholesteryl ester and phospholipid in the diet is considerably less than the amount of triacylglycerol; daily intake of cholesterol is generally less than 1 g and that of phospholipid is equal to 1 to 2 g. However, endogenous biliary lipids present additional cholesterol (1 g) and phospholipid (10 to 20 g) to the intestine; in the case of phospholipid, the biliary supply is much greater than that obtained from the diet (Northfield and Hofmann, 1975; Borgstrom, 1976).

LUMINAL DIGESTION OF LIPIDS

Digestion and absorption of lipids by the intestinal tract is a complex process that requires a number of steps to take place successfully in the lumen of the gastrointestinal tract, plus further processing of absorbed lipids by the enterocytes of the small intestinal mucosa.

Digestion of Triacylglycerols

The digestion of triacylglycerols begins in the stomach, with the action of gastric lipase secreted by the gastric mucosa. Gastric lipase is called an acid lipase because its activity is highest in an acidic medium. Acid lipases hydrolyze triacylglycerols that contain medium-chain fatty acids faster than they hydrolyze those containing long-chain fatty acids. Milk fat, rich in short- and medium-chain fatty acids, is hydrolyzed efficiently by acid lipase; this probably contributes to the efficient milk fat digestion observed in infants. Acid lipases do not hydrolyze cholesteryl esters or phospholipids such as phosphatidylcholine. Although the optimal pH of gastric lipase is around 4, the enzyme is still quite active at pH 6 to 6.5. Although the enzyme works well in the stomach, it probably continues to digest triacylglycerol in the upper duodenum where the pH is between 6 and 7. Acid lipase preferentially cleaves the fatty acid at the sn-3 position of the triacylglycerol molecule regardless of the fatty acid esterified to this position. The 1,2-diacylglycerols (diglycerides) and fatty acids produced as a result of the action of acid lipases may promote the emulsification of dietary fat in the stomach. Grinding and mixing of the gastric contents also contribute to dissection of the lipid droplets.

The lipid emulsion enters the small intestine as fine lipid droplets less than 0.5 mm in diameter. The combined action of bile and pancreatic juice brings about a marked change in both chemical and physical form of the ingested lipid emulsion. Most of the digestion of triacylglycerol is brought about by pancreatic lipase in the lumen of the upper part of the intestinal tract. Pancreatic lipase works at the interface between the oil and aqueous phases. Pancreatic lipase acts mainly on the sn-1 and sn-3 positions of the triacylglycerol molecule to release 2-monocacylglycerol and free fatty acids.
Pure pancreatic lipase works inefficiently in a bile salt-lipid mixture, and yet lipase present in pancreatic juice hydrolyzes triacylglycerols extremely efficiently. This observation led to the discovery of the cofactor called colipase. Colipase is a heat-stable protein required for lipase activity when bile salt is present; it is synthesized and secreted by the pancreas as procolipase and is activated to colipase in the small intestine by proteolytic cleavage by trypsin. As shown in Figure 7-1, the triacylglycerol lipid droplets covered with bile salts (BS in the diagram) are not accessible to pancreatic lipase. However, the binding of colipase to the triacylglycerol/aqueous interface allows the binding of lipase to the lipid/aqueous interface. Lipase binds with colipase in a 1:1 molar ratio.

**Digestion of Phospholipids**

Digestion of phospholipids occurs in the small intestine. In bile, phospholipid (predominantly phosphatidylcholine) is found in mixed micelles along with cholesterol and bile salts. Once in the intestinal lumen, the luminal phosphatidylcholine will distribute between the mixed micelles and the triacylglycerol droplets, but phosphatidylcholine tends to favor the micellar phase over the oily phase. It is phospholipid in micelles that serves as substrate for hydrolysis. Hydrolysis of phospholipids is largely brought about by phospholipase A₂, which is secreted by the pancreas as pro phospholipase A₂ and then activated by trypsin within the lumen of the small intestine. Phospholipase A₂ releases the fatty acid from the sn-2 position of phosphatidylcholine to yield a fatty acid and lysophosphatidylcholine. Although the bulk of biliary intestinal phospholipase A₂ activity is derived from pancreatic juice, there is probably some minor contribution from the intestinal mucosa, which has an intrinsic membrane enzyme that has phospholipase and retinyl ester hydrolyase activity and is known as retinyl ester hydrolyase, or phospholipase B.

**Digestion of Cholesteryl Esters**

Only free cholesterol is absorbed by the small intestine. Most dietary cholesterol is present as the free sterol; but 10% to 15% is present as the sterol ester. Biliary cholesterol is also mainly free cholesterol. Cholesteryl ester is hydrolyzed to free cholesterol in the presence of cholesteryl esterase (also called carboxyl ester hydrolase), which is secreted by the pancreas as an active enzyme. The human cholesteryl esterase has a broad specificity, and it can hydrolyze triacylglycerols, cholesteryl esters, phosphoglycerides, esters of vitamins A and D, and monoacylglycerols. Because it will hydrolyze all three ester linkages of triacylglycerols, it is sometimes called nonspecific esterase. As for phospholipase A₂, cholesterol es-

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**Figure 7-1. Interaction between lipase, colipase, and triacylglycerol droplets.** Lipase normally binds poorly to the triacylglycerol lipid droplets. In the presence of colipase, lipase molecules bind to the lipid droplets and hydrolyze the triacylglycerol to form 2-monocacylglycerol and fatty acids. BS, bile salts.
tense is active against substrates that have been incorporated into bile salt micelles. Cholesterol esterase activity is stimulated by bile salts, particularly trihydroxy bile salts such as sodium taurocholate. The activation of cholesterol esterase by bile salts is mediated by changing the conformation of the protein. A unique property of cholesterol esterase is its self-association; the presence of trihydroxy bile salts (taurocholate or glycocholate) promotes the self-aggregation of the enzyme into polymeric forms. The self-association of cholesterol esterase protects the enzyme from proteolytic inactivation. The cholesterol esterase isolated from the pancreas exists mainly as tetramers and dimers.

**UPTAKE OF LIPID DIGESTION PRODUCTS BY THE ENTEROCYTES**

The digestion products of triacylglycerols, phospholipids, and cholesterol esters are predominantly monoacylglycerol, fatty acids, lysophosphatidylethanolamine, and cholesterol. Although these lipid digestion products are somewhat polar, they have very limited capacity to dissolve in water. The epithelial surface of the small intestine is surrounded by a layer of water called the unstirred water layer, and the thickness of the unstirred water layer depends on how vigorously the small intestinal contents are mixed. Increased mixing reduces the thickness of the unstirred water layer. As illustrated in Figure 7-2, this unstirred water layer represents a barrier that lipids must cross before they can be absorbed by the enterocytes (small intestinal epithelial cells).

**Importance of Micellar Solubilization**

As shown in Figure 7-2, the limited solubility of lipid digestion products results in only a few individual molecules crossing the unstirred water layer and being absorbed by the enterocytes (arrow 1). To overcome this barrier, lipid digestion products are first solubilized in mixed bile salt micelles. Bile salts are biological detergents, and they will form micelles (aggregates of bile salts) when the concentration of bile salts in the lumen is at or above the critical micellar concentration. Pure micelles contain only bile salts, whereas mixed micelles contain bile salts as well as

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**Figure 7-2.** The role of bile salt micelles in overcoming the diffusion barrier associated with the unstirred water layer. In the absence of bile salts (1), only a limited number of lipid molecules diffuse through the unstirred water layer. If bile salts are present (2), more lipid molecules can be delivered to the brush border membranes by bile salt micelles.
other lipid moieties. The concentration of bile salts in the lumen is almost always above the critical micellar concentration. Free cholesterol and other lipid digestion products can be incorporated into the mixed micelles, thereby rendering these lipid molecules soluble in the bulk water phase. The mixed micelles containing polar lipids and cholesterol readily cross the unstirred water layer (arrow 2) and thereby increase the aqueous concentrations of fatty acids, monosaccyglycerols, cholesterol, and lysophosphatidylcholine near the epithelial aborptive surface by a factor of 100 to 1000. This provides an efficient mechanism for transport across the unstirred water layer and the subsequent uptake of these lipid molecules by the enterocytes.

Importance of Unilamellar Vesicles

When the jejunal contents of humans are sampled during digestion of a lipid meal and subjected to ultracentrifugation, one can observe a solid particulate layer on the bottom of the tube. Next, there is a mostly clear micellar phase followed by an oily phase on the top. The oily phase is composed mainly of triacylglycerols, mono- and diacylglycerols, and fatty acids. The micellar phase contains bile salts, monosaccyglycerols, and fatty acids. When Porter and Saunders (1971) performed a careful analysis of the aqueous phase after ultracentrifugation and also after the intestinal contents had been passed through a series of filters with progressively smaller pores, they observed that both the ultracentrifugation and the filtering procedure yielded a micellar phase that was slightly turbid. Furthermore, they found that there was a concentration gradient of lipids in the micellar phase, an important point that was missed previously. This observation is important because it hsted to the investigators that the micellar phase is not homogeneous.

The explanation for this intriguing observation came much later when Carey et al. (1985) proposed that the lipid in the intestinal lumen will be incorporated into mixed micelles when the bile salt concentration in the lumen exceeds the critical micellar concentration. These micelles are probably in the form of large, mixed disc-like micelles more or less saturated with lipids, with a hydrodynamic radius of about 200 Å. When the amount of lipid in the aqueous phase increases further (as occurs in some diseased states), formation of lipid crystalline vesicles (liposomes) with hydrodynamic radii of 400 to 600 Å also occurs (Creey et al., 1983).

The discovery of the existence of vesicles has important pathophysiological implications. Patients with low intraluminal bile salt concentrations (Mansbach et al., 1980) and patients with bile fistulae (Porter et al. 1971) have reasonably good fat absorption, and Carey et al. (1985) proposed that lipid crystalline vesicles may play an important role in the uptake of lipid digestion products in these patients.

Permeation of Digested Fat into Enterocytes

At least two different mechanisms have been proposed for the uptake of lipid digestion products by the small intestine: passive uptake versus carrier-mediated uptake. Once digested lipid is presented to the surface of the brush border membrane of the enterocytes, the products of lipid digestion diffuse into the lipid phase of the brush border membrane. The concentration gradient between the lipids in the brush border and those in the intracellular compartment of the enterocytes favors initial diffusion of these products into the cell. The rapid reesterification of the intracellular lipids to form triacylglycerols, phospholipids, and cholesterol esters by enzymes of the endoplasmic reticulum helps maintain low intracellular concentrations of these lipids, favoring the continued uptake or diffusion of these lipids into the intracellular compartment of the enterocytes. The more water-soluble products of lipid digestion, such as glycerol and short-chain fatty acids, if present, are efficiently taken up by diffusion.

Specific binding proteins have been identified that may participate in the uptake processes for some lipids, including fatty acids and cholesterol. These binding proteins have been purified, and antibodies against them have been raised. Antisera raised against these binding proteins was used to demonstrate that these binding proteins are located at the ap-
Recent studies revealed that there are at least two cytosolic FABPs in enterocytes. These are the hFABP (intestinal FABP) and L-FABP (liver FABP). These two FABPs differ in their binding specificity (Bernard et al., 1996). hFABP strongly binds only with fatty acids, but L-FABP will bind not only long-chain fatty acids but also lysophosphatidylcholine, retinoids, bilirubin, carotenoids, and even statins (Clatz and Veerkamp, 1985; Baas, 1988; Bansal et al., 1990). Based on nuclear magnetic resonance (NMR) binding studies, Costola et al. (1990) speculated that the hFABP facilitates the intracellular transport of fatty acids, whereas L-FABP probably facilitates the intracellular transport of monoaicylglycerol and lysophosphatidylcholine. Future studies in mice with various FABP genes "knocked out" may shed some light on the function of these proteins in the intracellular transport of lipids.

Two cytosolic carrier proteins for steols have been isolated and characterized: sterol carrier protein-1 (SCP-1) and sterol carrier protein-2 (SCP-2). Experimental evidence thus far seems to indicate that SCP-2 may play a role in the intracellular transport of cholesterol.

Reesterification of Lipid Digestion Products

2-Monoacylglycerols and fatty acids are reconstituted to form triacylglycerol, mainly via the monoacylglycerol pathway. As shown in Figure 7-3, 2-monoacylglycerol is reacylated into triacylglycerol by the consecutive action of monoacylglycerol acyltransferase and diacylglycerol acyltransferase. The enzymes involved in this monoacylglycerol pathway are present in a complex called "triacylglycerol synthetase," and this has recently been purified (Lehner and Kukis, 1985). The enzymes involved in the monoacylglycerol pathway are located on the cytosolic surface of the endoplasmic reticulum (ER). This finding is important bearing in our understanding of the intracellular formation of chylomicrons. It appears that triacylglycerols are formed at the cytosolic surface of the ER, and they then gain access to the inside of the ER.

Wettermark and Zilversmit (1984) demonstrated that there is a protein in the liver the
small intestine, and a number of other organs that promotes the transfer of triacylglycerol and cholesteryl ester between membranes; they proposed that this transfer activity may play a role in the movement of lipids into the ER. Wetterau and his colleagues have recently provided convincing evidence that this may indeed be the case (1992). In studies of patients with abetalipoproteinemia, who lack the ability to make chylomicrons, they found that apo B (apo B) was synthesized, but the triacylglycerol and apo B failed to associate with each other because of a lack of the microsomal (ER) triacylglycerol transfer protein.

The other pathway present in intestinal mucosa for the formation of triacylglycerol is called the glycerol 3-phosphate pathway. As shown in Figure 7–3, this pathway involves the stepwise acylation of glycerol 3-phosphate to form phosphatidic acid. In the presence of phosphatidate phosphatase, phosphatidic acid is hydrolyzed to release inorganic phosphate and to form diacylglycerol, which is then further esterified to form triacylglycerol. The relative importance of the monoacluberol glycerol pathway and the glycerol phosphate pathway depends on the supply of 2-monoacylglycerol and fatty acid. During normal lipid absorption, the monoacluberol pathway is much more important than the glycerol phosphate pathway in enterocytes because of the abundant supply of 2-monoacylglycerol and fatty acid and their efficient conversion to triacylglycerol, and also because 2-monoacylglycerol inhibits the glycerol phosphate pathway. However, when the supply of 2-monoacluberol is lacking or insufficient, the
glycerol's phosphate pathway becomes the major pathway for the formation of triacylglycerol.

Phospholipids

Lyso phosphatidylcholine and other lyosphospholipids inside the enterocytes can be reacylated to form phosphatidylcholine and other phospholipids, or these lyosphospholipids can be hydrolyzed to form glycerol 3-phosphorylcholine. The liberated fatty acids can be used for triacylglycerol synthesis, whereas the glycerol 3-phosphorylcholine can be readily transported via the portal blood for use in the liver. Another reaction that occurs in intestinal mucosal cells is the combination of two molecules of lyso phosphatidylcholine to yield one molecule of phosphatidylcholine and one molecule of glycerol 3-phosphorylcholine.

Cholesterol

Dietary or exogenous cholesterol absorbed by the enterocytes enters a free cholesterol pool.

This free cholesterol pool in the enterocyte also derives cholesterol from endogenous sources, including nonintestinal cholesterol (biliary cholesterol and cholesterol from cells shed from the intestinal mucosa) absorbed from the lumen of the small intestine, cholesterol derived from circulating plasma lipoproteins, and cholesterol synthesized de novo. However, enterocytes handle cholesterol from various sources quite differently. For instance, the cholesterol derived from the intestinal lumen does not mix freely with the free cholesterol pool in the enterocytes and is preferentially esterified in the enterocytes for incorporation into chylomicrons and exported into the lymph. Stange and Dietschy (1985) found that very little newly synthesized cholesterol is transported into lymph during fasting; however, during active lipid absorption and chylomicron synthesis, some of the newly synthesized cholesterol can be incorporated into chylomicrons and transported in lymph.

Cholesterol is transported almost exclusively by the lymphatic system, mainly as esterified cholesterol. Therefore, the rate of esterification of cholesterol is crucial.
terification of cholesterol probably regulates the rate of its lymphatic transport. Two enzymes have been proposed to play a role in cholesterol esterification; these are cholesterol esterase and acyl-CoA:cholesterol acyltransferase (ACAT). The distribution and regulation of ACAT in the small intestinal epithelium has been studied in considerable detail. Both the jejunal and ileal have high activities of this enzyme, with the ileum having significantly higher levels than the jejunum. The activity of this enzyme can be increased by feeding a high cholesterol diet. Using immunocytochemistry, Gallo et al. (1980) demonstrated that intracellular cholesterol esterase is derived from the uptake of pancreatic cholesterol esterase. It is hypothesized that this cholesterol esterase could catalyze esterification instead of hydrolysis with the relatively high intracellular concentrations of free cholesterol and fatty acids. As yet, we do not understand the process of how pancreatic cholesterol esterase is taken up by the enterocyte, but elucidation of this process may enhance our general understanding of how intact proteins are taken up by the enterocytes. (See Chapter 6 for a description of the uptake of intact proteins from the intestine.)

There is still lack of a general agreement about whether cholesterol esterase or ACAT plays a more important role in the esterification of cholesterol by the enterocytes. However, prevailing evidence supports a more important role of ACAT in mucosal cholesterol esterification. For instance, the higher activity of ACAT is found in the segment of the small intestine most actively involved in cholesterol absorption. Furthermore, the activity present in the intestinal epithelium can adequately account for all the cholesterol ester transported by the small intestine. Lastly, in studies employing a number of specific ACAT inhibitors, it has been demonstrated that these inhibitors significantly reduce the transport of cholesterol by the small intestine.

**Plant Sterols**

It has been well documented that plant sterols are handled differently from cholesterol by the mammalian gut, but the precise mechanisms responsible for this difference are not well understood. Although structurally quite similar to cholesterol, only a small percentage (~5%) of ingested β-sitosterol is absorbed in humans (Gould, 1955; Saleen et al., 1970). This tremendous ability of the small intestine to discriminate against the plant sterols seems to be lost in patients with familial hypercholesterolemia (Bhattacharyya and Connor 1974; Saleen et al., 1997). Two sisters with β-sitosterolemia had plasma cholesterol concentrations of 20 to 210 mg/100 mL and plasma plant sterol concentrations of 20 to 37 mg/100 mL (two thirds as β-sitosterol and the rest as campesterol). In most humans, the level of total plant sterols present in plasma is less than 0.9 mg/100 mL. Both sisters apparently had increased absorption of plant sterols and developed xanthomasis (a condition characterized by the presence of xanthomas, which are nodules composed of lipid-laden foam cells). High levels of plant sterols were found in plasma, erythrocytes, adipose tissue, and skin.

β-Sitosterolemia appears to be caused by an inherited recessive trait. Field and Mathur (1983) showed that the coenzyme A-dependent esterification of cholesterol is at least 50 times more efficient than that of β-sitosterol. They proposed that inadequate esterification of this plant sterol in the enterocytes is probably responsible for the poor absorption of β-sitosterol by the small intestine. The presence of plant sterols in the intestinal lumen also inhibits the intestinal absorption of cholesterol. The mechanisms for this inhibition are the displacement of cholesterol from the bile salt–mixed micelles by plant sterols. The consumption of a sitosterol (another plant sterol) ester margarine was recently observed to lower serum cholesterol in a Finnish population with mildly elevated plasma cholesterol levels (Miettinen et al., 1995).

**ASSEMBLY OF INTESTINAL LIPOPROTEINS**

Lipoproteins are lipid-protein complexes formed by the small intestine and the liver for the export of lipids from these organs. The cannulation of the lymphatic vessels of rats and a number of other animal species has been used extensively for the study of chylo-
micron secretion by the small intestine. This method allows the direct sampling and analysis of lipoproteins secreted by the small intestine before they enter the general circulation.

Lipoproteins Secreted by the Small Intestine

The small intestine secretes the following lipoproteins: (1) chylomicrons; (2) intestinal very low density lipoproteins (VLDLs, small chylomicrons); and (3) high density lipoproteins (HDLs). Both chylomicrons and intestinal VLDLs are triacylglycerol-rich lipoproteins. In this chapter, only chylomicrons and intestinal VLDLs are discussed because the small intestine secretes only a small amount of HDLs. High density lipoproteins are discussed in Chapter 14. During fasting, the major lipoproteins secreted by the small intestine are the intestinal (apo B-48-containing) VLDLs. Chylomicrons are the major lipoproteins secreted by the small intestine following a lipid-rich meal.

Assembly and Secretion of Chylomicrons

Only the small intestine secretes chylomicrons. The composition of the chylomicron is described in Table 7-1. The major apolipoproteins associated with chylomicrons are apo A-I, apo A-IV, and apo B-48. Traces of apo E and apo C are also added to the chylomicrons after their entry into the circulation. Data from both animals and humans indicate that the fatty acid composition of the triacylglycerol of chylomicrons closely resembles that of the dietary lipids consumed. Kaytoue et al. (1983) studied the changes in human lymph triacylglycerol composition after a subject ingested 100 g of corn oil (Table 7-2). The fatty acid composition of chylomicron triacylglycerol collected 8 hours after the lipid dose was virtually identical to that of the corn oil ingested.

The fatty acid composition of the phospholipids of lymph chylomicrons is less influenced by the dietary fatty acids because phos-
Absorption of Lipophilic Drugs and Toxins

Many lipophilic compounds, including drugs, fat-soluble vitamins, and other compounds, present in food or present on food as contaminants are incorporated into chylomicrons in the intestinal mucosal cells and transported via the lymph. The extent to which these compounds are involved in the formation of chylomicrons is known. Uptake of lipophilic drugs and toxins also may be enhanced when ingested along with dietary fat. Factors that limit lipid digestion, such as a high-protein diet, may interfere with the formation and absorption of these compounds.

Patients are often advised to take lipophilic medications together with their meals to enhance the absorption of the drug by the gastrointestinal tract. Delivery of lipophilic drugs in forms that enhance their absorption and thus require smaller doses is of interest to pharmaceutical manufacturers. Enhancement of absorption of lipophilic compounds by the presence of dietary fat also may affect absorption of environmental contaminants or toxins. For example, absorption of DDT (1,1,1-trichloro-2,2-dichloroethylene), a toxic chlorinated hydrocarbon pesticide now banned in the United States, is enhanced by concomitant fat feeding. Enhancement of absorption of lipophilic compounds by dietary fat also may account for the observation that trichloroethylene, a drug used for treating hookworm infections that is not ordinarily absorbed from the gastrointestinal tract, causes toxic effects when fed with a high-fat meal.

FACTORS AFFECTING FORMATION AND SECRETION OF CHYLOMICRONS

The formation and secretion of chylomicrons are tightly regulated by the synthesis of apolipoproteins and lipids in the enterocytes. Although considerable information regarding these factors has been gathered, the mechanisms of how these factors regulate the formation and secretion of chylomicrons largely remain unknown. A major reason for our limited knowledge of this is the lack of good cell models for studying these processes. It is extremely difficult to maintain intestinal epithelial cells in culture.

The formation and secretion of chylomicrons is not different between lymph chylomicrons and intestinal VLDLs. The intestines secrete both chylomicrons and intestinal VLDLs. During fasting, intestinal VLDLs are the only lipoproteins produced by the small intestine. Chylomicrons are the major lipoproteins produced by the small intestine following a lipid-rich meal.

Assembly and Secretion of Very Low Density Lipoproteins

As shown in Table 7-1, intestinal VLDLs are smaller than chylomicrons, and they have a different lipid composition compared with chylomicrons. In contrast, the apolipoprotein composition of chylomicrons is not different between lymph chylomicrons and intestinal VLDLs. The intestines secrete both chylomicrons and intestinal VLDLs. During fasting, intestinal VLDLs are the only lipoproteins produced by the small intestine. Chylomicrons are the major lipoproteins produced by the small intestine following a lipid-rich meal.

phatidylcholine from bile is preferentially used for the coating of chylomicrons. The fatty acid composition of biliary phosphatidylcholine is rather unique.

The intestinal triacylglycerol-rich lipoproteins are transported from the endoplasmic reticulum to the Golgi apparatus. The Golgi apparatus serves as the final assembly site for many proteins and also lipoproteins. Consequently, a block in the trafficking between endoplasmic reticulum and the Golgi apparatus results in impairment in the formation of chylomicrons. Apo B is involved in this process. Terminal glycosylation of proteins occurs at the Golgi apparatus. Golgi-derived vesicles containing the pre-chyomicrons have been clearly demonstrated by Sabesan and Froze (1977) in enterocytes actively absorbing lipid. This is illustrated in Figure 7-1. The Golgi vesicles containing the pre-chyomicrons migrate toward the basolateral membrane of the enterocytes, and pre-chyomicrons are discharged into the intercellular space through exocytosis.
Synthesis of Apolipoprotein B

Two major forms of apolipoprotein B are made by humans, apo B-100 and apo B-48. Apo B-100 and apo B-48 refer to the relative apparent molecular masses obtained by sodium dodecyl sulfate (SDS) gel electrophoresis. According to this nomenclature, the large apo B-100 was assigned an arbitrary value of 100 while apo B-48 was assigned the number 48 because it has an apparent molecular mass that is 48% that of apo B-100. In humans, the liver secretes only apo B-100, and the small intestine secretes only apo B-48. Both apo B-100 and apo B-48 are encoded by the same gene. The biosynthesis of apo B-48 involves a unique mechanism by which a CAA (cytosine-adenine-adenine) codon encoding glutamine [codon 2153 of the apo B-100 messenger RNA (mRNA)] is changed to a UAA (uracil-adenine-adenine) stop codon, and thus translation is terminated earlier to form apo B-48 (Poweit et al., 1987; Chen et al., 1987). Although we know that apo B is required for the formation of chylomicrons, the supply of apo B is probably not the rate-limiting step for lipid output in chylomicrons. Hayashi et al. (1980) demonstrated that apo B output by the small intestine did not change after intraduodenal infusion of lipid, despite the fact that lymphatic triacylglycerol output increased severalfold. It appears that the number of chylomicron particles made by the small intestine remains relatively constant during fasting and active lipid absorption. Instead of making more chylomicrons during active lipid absorption, the enterocyte simply fills each chylomicron particle with more triacylglycerol molecules, making them larger and lighter.

Synthesis of Apolipoproteins A-I and A-IV

The human small intestine synthesizes both apo A-I and A-IV. These apoproteins are secreted associated with chylomicrons. Despite the marked increase in chylomicron secretion by the small intestine following the digestion of a lipid-rich meal, the synthesis and secretion of apo A-IV is only marginally stimulated (20% to 30%) compared with the fasting condition. In contrast, the synthesis and secretion
of apo AIV is markedly stimulated by the ingestion of fat. The roles of apo AI and apo AIV in the formation and secretion of chylomicrons are not clear.

When chylomicrons are metabolized in the body by lipoprotein lipase, apo AIV detaches from these particles and circulates in the plasma either bound to HDLs or as a free protein. Although the amino acid sequence and the gene locus of apo AIV have been known since 1984, the physiological role of apo AIV remained unclear until recently. A number of recent reports indicate a unique physiological role for apo AIV: apo AIV appears to be a circulating signal released in response to fat feeding that may mediate the anorectic (inhibition of food intake) effect of a lipid meal. This function of apo AIV is discussed later in this chapter.

Role of Luminal Phosphatidylcholine
An adequate supply of luminal phosphatidylcholine is important for the formation and secretion of intestinal chylomicrons and intestinal VLDLs. It may be important because it provides the phosphatidylcholine for the surface coat of chylomicrons. The observation that bile phosphatidylcholine has a unique fatty acid composition that closely resembles the fatty acid composition of the phosphatidylcholine in the surface coat of chylomicrons suggests that bile phosphatidylcholine is preferentially used for the coating of chylomicrons. In a model system, the lipolysis of triacylglycerol emulsions and the rates of clearance of particles from the plasma by hepatocytes versus reticuloendothelial cells depend on the cholesterol content and the phosphatidylcholine species of the lipid emulsion particles (Clark, et al., 1991). Consequently, the specific fatty acid composition of the phosphatidylcholine in the chylomicron surface coat may play an important role in the metabolism of chylomicrons by the body.

Luminal phosphatidylcholine also may be important in maintaining the normal composition, turnover, and integrity of the membranes of the subcellular organelles of the enterocytes. Normal membrane composition and integrity are important not only for membrane function but also for the function of the enzymes associated with it.

Hormones
Relatively little is known of how intestinal lipid absorption is regulated by hormones. It has been reported that neurotensin enhances lymphatic lipid transport in the rat by enhancing the processing of absorbed dietary fat. However, neurotensin also induces hemodynamic changes in the gastrointestinal tract (e.g., increased lymph flow). Further experiments are needed to ascertain whether this effect of neurotensin on intestinal lipid transport is an intracellular effect or simply a hemodynamic effect. The role of gastrointestinal hormones on intestinal lipid transport is largely unknown.

DISORDERS OF INTESTINAL LIPID ABSORPTION
The determination of the amount of fat in the stool is a common test for assessment of intestinal malabsorption. Normal humans excrete in their stool less than 6 g of fat per 24-hour period, which is less than 5% of their fat intake. Thus, they absorb more than 95% of the fat consumed in their diet. The efficiency of fat absorption can be calculated as fat intake (in g per day) minus fecal fat (g per day) divided by fat intake in g per day with the dividend multiplied by 100 to express the fraction as a percentage. Intestinal lipid malabsorption can be caused by a number of clinical conditions.

Disorders of the Small Intestine
A common disorder of the small intestine that results in malabsorption of nutrients, including lipids, is celiac sprue. Celiac sprue is characterized by lesions of the small intestinal mucosa. It is caused by gluten (a protein rich in prolne and glutamine that is found in wheat, barley, oats, and some other cereals). This malabsorptive state can be corrected by feeding a gluten-free diet. The mechanism of gluten toxicity is still unclear.
Defective Digestion Caused by Pancreatic Deficiency

A major feature of pancreatic deficiency is severe abdominal pain and steatorrhea (the passage of large, pale, frothy stools), caused by the presence of a large amount of undigested fat owing to a disease of the pancreas or pancreatectomy (removal of the pancreas), which results in lack of pancreatic digestive enzymes. Pancreatic deficiency is treated by prescribing a low-fat diet or by supplementation of pancreatic enzymes with meals; severe deficiency may require driers containing partial hydrolylates of proteins and starch as well.

Defective Uptake of Lipids Caused by Bile Salt Deficiency

Bile salt deficiency results in poor micellar solubilization of lipid digestion products. Unlike pancreatic deficiency, bile salt deficiency does not affect the digestion of triacylglycerol, and therefore the fats present in the stool are mainly lipid digestion products. As described earlier, the solubilization of lipid digestion products in micelles formed by the bile salts is an important method for delivery of lipid molecules to the small intestinal epithelial cells. However, because vesicles may also play a role in the delivery of lipid digestion products to the small intestine, patients with bile salt deficiency caused by liver disease or gallstone disease often can absorb a significant amount of lipids.

Abetalipoproteinemia

Apo B is required for the formation and secretion of intestinal and hepatic triacylglycerol-rich lipoproteins, as evidenced by the lack of apo B-containing lipoproteins in the circulation in patients suffering from abetalipoproteinemia. For a long time, it was generally believed that these patients lacked the ability to synthesize apo B. However, recent studies have clearly shown that apo B is being synthesized by the enterocytes of abetalipoproteinemic patients. Furthermore, Talmud et al. (1988) have performed linkage studies in kindreds of patients with abetalipoproteinemia and have shown that the apo B gene is normal. Thus, the problem in these patients may be a defect in the association of intracellular lipid with apo B, a prerequisite for the normal packaging of apo B-containing lipoproteins. Consequently, triacylglycerol droplets accumulate in the intestinal mucosa of these subjects as lipid absorption progresses (Dobbins, 1970, Fig. 7–5).

Chylomicron Retention Disorder

The chylomicron retention disorder involves failure of the discharge of pre-chylomicrons from the Golgi-derived vesicles into the intercellular space through exocytosis. Pre-chylomicrons refer to the chylomicrons that are still inside the intestinal epithelial cells. Consequently, despite the presence of numerous Golgi-derived vesicles in the cytoplasm, there is an absence of chylomicrons in the intercellular space (Roy et al., 1987). Compared with abetalipoproteinemia, chylomicron retention disorder involves a defect that is further along the chylomicron packaging pathway probably in the trafficking of Golgi vesicles containing pre-chylomicrons to the plasma membrane.

INTESTINAL LIPID ABSORPTION AND MUCOSAL INJURY

A number of investigators have shown that long-chain fatty acids can be injurious to the intestine, especially the developing intestine. Velasquez et al. (1994) reported that the magnitude of injury caused by the presence of fatty acids in the intestinal lumen was significantly higher in piglets less than 2 weeks of age than in 1-month-old animals, suggesting that a developmental process that renders the intestinal mucosa more resistant to lipid-induced injury occurred in the piglets. The lipid-induced injury was reversible. This interesting observation is of potential clinical relevance. Immaturity or disruption of the intestinal mucosal barrier by fatty acids may result in clinical disease states to which the newborn infant is susceptible, such as necrotizing enterocolitis or toxic megacolon. Velasquez et al. (1994) also showed that esterification of the injurious long-chain fatty acids with ethanol abolished their cytotoxic effects on the intesti-
nal mucosa. Furthermore, they showed that the ethyl esters of the long-chain fatty acids are absorbed and utilized by the developing intestine.

REGIONAL DIFFERENCES IN INTESTINAL LIPID ABSORPTION

A difference in the abilities of the proximal and distal small intestine to absorb fat has been described in rats (Wu et al., 1980; Sabin et al., 1975). Not only was the distal intestine much less efficient than the proximal intestine in chylomicron production (Sabin et al., 1975), but also the chylomicrons produced by the distal intestine were larger (Wu et al., 1980). The investigators suggested that this difference between intestinal segments could be due to the availability of phospholipids for the coating of pre-chylomicrons or to altered intracellular membrane lipid composition. Most phospholipids are absorbed before the chyme reaches the distal small intestine. Although the proximal intestine is supplied with biliary phospholipids, the distal small intestine has to meet most of its phospholipid requirements by either de novo synthesis or uptake of lipoproteins from the plasma.

PORTAL TRANSPORT OF LONG-CHAIN FATTY ACIDS

The majority of absorbed fatty acids are transported by intestinal lymph as chylomicrons and intestinal VLDLs. However, there is evidence for portal transport of long-chain fatty acids, and this transport is increased when there is a defect in the intracellular esterification of fatty acids to form triacylglycerol or an impairment in chylomicron formation. In patients with abetalipoproteinemia, some dietary fat (fatty acids) may be absorbed in the virtual absence of chylomicron formation (Ways et al., 1987).
McDonald et al. (1980) demonstrated that a substantial amount of the absorbed fatty acid (58% for linoleic acid, w6, 18:2) was transported in the portal blood of normal rats when the rates of lipid absorption were low. Recently, Mannscher et al. (1981) reported that a considerable amount of endogenous and exogenous fatty acids is transported by the portal blood. The amount of exogenous fatty acid transported via the portal route can be as much as 20% of the fatty acid infused into the small intestinal lumen. The portal trans- port of absorbed fatty acids by the small intestine therefore may be more important than previously recognized.

SATIETY EFFECTS OF FAT FEEDING

There is compelling evidence in the literature showing that the ingestion of fat results in satiation and the inhibition of food intake. A number of characteristics of this lipid-induced satiety provide important clues about the mechanism involved. First, long-chain fatty acids are significantly more potent in inducing satiety than are short- or medium-chain fatty acids. Because long-chain fatty acids are transported by the small intestine mainly as chylomicrons whereas medium-chain fatty acids are transported primarily in the portal blood, this observation would imply that chylomicrons are somehow involved in this lipid-induced satiety. Second, fatty acids introduced into the small intestinal lumen are more potent in inducing satiety than are fatty acids delivered by direct peripheries venous, portal, or caval routes. Again, this would imply that the gastrointestinal tract, and probably the production of chylomicrons, is somehow involved in this lipid-induced satiety. Third, the lipid-induced satiety is abolished by the presence of arilat (an inhibitor of pancreatic lipase) in the intestinal lumen, indicating that it is the digestion products and not triacylglycerols per se that elicit the satiety response. Thus, most of the observations concerning lipid-induced satiety seem to imply that intestinal lipid absorption, in particular the formation and secretion of chylomicrons, is involved in this physiological response to the ingestion of lipid.

Fujimoto et al. (1992) reported an exciting finding that apo AIV, an apolipoprotein made and secreted as part of the chylomicrons by the small intestine epithelial cells, may be involved in this lipid-induced satiety. The synthesis and secretion of apolipoprotein AIV is modestly stimulated by the ingestion of fat. It apparently acts on the central nervous system to elicit the satiety response, but the mechanism by which apo AIV inhibits food intake is not understood. A number of recent reports (Okumura et al., 1994, 1996) suggested that apo AIV may act as a modulator of upper gastrointestinal tract function by inhibiting gastric emptying as well as gastric acid secretion. Thus, apo AIV appears to play an important role in the integrated control of digestive function and ingestive behavior.

REFERENCES


7 — Digestion and Absorption of Lipids


RECOMMENDED READINGS