Insights from human congenital disorders of intestinal lipid metabolism

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ABBREVIATIONS, ABL: Abetalipoproteinaemia; ANGPTL3: Angiopoietin-like 3 protein; CE: Cholesteryl ester; CM: Chylomicron; CRD: Chylomicron retention disease; DGAT: Diacylglycerol acyltransferase; ER: Endoplasmic reticulum; FA, fatty acids; fatty acid binding protein (FABP); FHBL: Familial hypobetalipoproteinemia; low-density lipoprotein (LDL); MGAT: Monoacylglycerol acyltransferase; MTP: Microsomal triglyceride transfer protein; MG: monoacylglycerols; PCSK9: Proprotein convertase subtilisin/kexin type 9; PL: Phospholipid; TG: triacylglycerols; VLDL, very-low density lipoprotein.
**ABSTRACT**

The intestine must challenge the profuse daily flux of dietary fat that serves as a vital source of energy and as an essential component of cell membranes. The fat absorption process takes place in a series of orderly and interrelated steps, including the uptake and translocation of lipolytic products from the brush border membrane to the endoplasmic reticulum, lipid esterification, apolipoprotein (Apo) synthesis and ultimately the packaging of lipid and Apo components into chylomicrons (CM). Deciphering inherited disorders of intracellular CM elaboration afforded new insight into the key functions of crucial intracellular proteins, such as Apo B, microsomal triglyceride transfer protein and Sar1b GTPase, the defects of which lead to hypobetalipoproteinemia, abetalipoproteinemia and chylomicron retention disease, respectively. These “experiments of nature” are characterized by fat malabsorption, steatorrhea, failure to thrive, low plasma levels of triglycerides and cholesterol, and deficiency of liposoluble vitamins and EFAs. After summarizing and discussing the functions and regulation of these proteins for reader's comprehension, the current review focuses on their specific roles in malabsorptions and dyslipidemia-related intestinal fat hyperabsorption while dissecting the spectrum of clinical manifestations and managements. The influence of newly discovered proteins (Proprotein convertase subtilisin/kexin type 9 and Angiopoietin-like 3 protein) on fat absorption has also been provided. Finally, it is stressed how the overexpression or polymorphism status of the critical intracellular proteins promotes dyslipidemia and cardiometabolic disorders.
A. BRIEF OVERVIEW OF INTESTINAL FAT ABSORPTION

The small intestine is the major site for the transport of alimentary fat, the most calorically dense nutrient, in the form of lipoproteins. The process of lipid absorption may schematically be divided in three sequential phases: intraluminal, intestinal and secretory events (Figure 1). The intraluminal phase includes chemical hydrolysis of lipids [mainly triacylglycerols (TGs), phospholipids (PL) and cholesteryl esters (CE)] by lipolytic enzymes (pancreatic lipase, phospholipase A2 and cholesterol esterase, respectively), the micellar solubilization of lipolytic products by bile acids (1-13) and the shuttle of micelles through the unstirred water layer to the surface of the microvillus membrane (14-19). The resistance of the unstirred water layer is influenced by the chain length and saturation of fatty acids (FAs) as well as lipid species (14, 16, 20). The intestinal phase comprises passive diffusion and protein-mediated facilitated transport of hydrolyzed products (FAs, monoacylglycerols, free cholesterol and lyso-PLs) across the microvillous membrane of the enterocyte involving several transporters [e.g., cluster of differentiation 36, fatty acid transport protein (FABP)4 and plasma membrane FABP] (21-27). However, it is important to specify that the transfer of long-chain FAs, in particular, is a highly controversial question that has attracted the interest of several reviews (28-34). Once in the cytoplasm, the lipolytic products are bound by cytosolic FABPs (intestinal- and liver-FABP) (35-43) before being targeted to the endoplasmic reticulum (ER) for the re-esterification of TG [essentially by monoacylglycerol acyltransferase (MGAT) and diacylglycerol acyltransferase (DGAT)], CE [with the involvement of acyl-CoA cholesterol acyltransferase (ACAT-2)] and PL (via combination of diacylglycerol with choline and ethanolamine), and finally the formation of lipid-carrying lipoproteins in the secretory pathway (44-46). The delivery phase involves the exocytosis of chylomicron (CM) from the absorptive cells and their subsequent delivery into the
systemic circulation via the intestinal lymph. Experimental and clinical approaches have significantly advanced our understanding of the intra-enterocyte step that brings together the large amphipathic Apo B-48 polypeptide and newly esterified lipids in a fixed temporal sequence (47). Once the translation and translocation of Apo B-48 into the lumen of the ER are initiated, microsomal triglyceride transfer protein (MTP) shuttles lipids from the ER membrane to the growing Apo B-48 chain in the ER, allowing the protein to translocate completely into the lumen (47, 48). However, deprivation of lipid substrate causes degradation of Apo-B-48 and inhibition of nascent CM assembly (47, 49). The pre-CM is transported via a pre-CM transport vesicle to the Golgi apparatus (50) where the coat is removed and a mature CM is formed through acquisition of more neutral lipids (51). More specifically, the maturation of CM in the Golgi apparatus is accompanied by further alterations in Apo B-48 glycosylation (52, 53), Apo A-I accretion (54), lipid composition (53) and size (54). The transfer process is highly dependent on the small Sar1b GTPase that initiates the vesicular coat protein complex II (COPII)-dependent transport of cargo from the ER to the Golgi apparatus (55) (Figure 1).

This article is aimed at summarizing insights gained in the last two decades on the important pathways modulating key intracellular proteins in CM formation and intestinal fat absorption. One of our major goals for this article is to familiarize readers with this expanding and evolving understanding related to the regulation, ontogeny and functions of the crucial proteins: Apo B-48, MTP and Sar1b GTPase. We will also summarize the impact of their genetic defects on enterocyte lipid transport while pointing out clinical disorders and management.

B. ABETALIPOPROTEINEMIA

1. INTRODUCTION
Abetalipoproteinaemia (ABL; OMIM#200100) is an autosomal recessive disorder that is usually detected during infancy due to failure to thrive, severe diarrhea and fat malabsorption. The defective gene was discovered 40 years after the first description of the disease (56). The primary cause is due to the defects in MTP (48) (Figure 2), leading to abnormal assembly of intestinal and hepatic Apo B-containing lipoproteins (57-61), thereby explaining the virtual absence of plasma Apo B-100 and Apo B-48, as well as the low plasma concentrations of TG and cholesterol (62).

2. MTP PROPERTIES

MTP is located within the lumen of microsomes of the liver and intestine (63, 64). It appears as a heterodimer with two subunits of apparent molecular weight 58,000 and 88,000 kDa (65). The small subunit has been identified as the multifunctional protein disulfide isomerase (63), while the unique large subunit confers the catalytic property to the protein complex (65). It catalyzes the transfer of TG, CE and phosphatidylcholine between PL surfaces. In fact, by transporting lipids by a shuttle mechanism (Ping Pong Bi Bi kinetics), MTP acts as a carrier of lipids from their site of synthesis to nascent lipoproteins within the ER (66). Additional evidence for the critical role of MTP in CM and very-low density lipoprotein (VLDL) production in the enterocyte and hepatocyte, respectively, has been provided with pharmacological approaches utilizing specific MTP inhibitors that simultaneously inhibit lipid transfer activity in situ while blocking Apo B secretion (67-70). Treatment with MTP inhibitors results in a dose-dependent decrease in Apo B secretion, suggesting that MTP is rate-limiting for TG-rich lipoprotein secretion (67). Investigators have rapidly exploited the discovery of ABL molecular basis to elaborate on the concept that pharmacologic MTP inhibition may constitute a powerful strategy to reduce Apo B-containing lipoproteins [CM, VLDL and low-density lipoprotein (LDL)] in
dyslipidemia with a considerable impact on the prevention of cardiovascular diseases. Given gastrointestinal and hepatic adverse effects, the development of most of MTP inhibitors was halted except Lomitapide that has succeeded to cross phase 2 (71, 72) and exhibited efficacy and safety in a recent phase 3 focusing on homozygous familial hypercholesterolaemia (73). Importantly, MTP interacts with Apo B through its N-terminal β-barrel domain (residues 22–297), while the middle α-helical domain (residues 298–603) mediates the interaction with protein disulfide isomerase, and the C-terminal mediates the lipid-binding and transfer catalytic activity of MTP (74-76). It is now well established that MTP elicits lipidation of growing Apo B polypeptide chain, thus favouring the assembly and secretion of lipoproteins (77, 78). In addition to achieving net transfer of lipid to Apo B during its translation, MTP can also activate the trafficking of TG droplets from the cytosol to the ER lumen and catalyse the fusion of nascent Apo B-containing particles with TG droplets, thereby contributing to CM expansion (79, 80). If MTP is absent, as seen in the condition of ABL, Apo B does not fold properly because of the defect of adequate lipidation in the ER, which irremediably leads to its proteosomal degradation (81). Therefore, the transient physical interaction of MTP with Apo B is crucial at early stages of lipoprotein synthesis (78, 82-84) (Figure 1). The absence of MTP is triggered by mutations in the gene for the large MTP subunit, which contains 18 exons and spans approximately 55-60 kb in chromosome 4 (4q22-q24) (85). Interestingly, one of the quantitative trait locus for LDL is located in a region of chromosome 4 that contains MTP that constitutes a clearly strong positional candidate gene for association for the variation effects on LDL size fractions (86, 87). Furthermore, a systemic exploration of the chromosome 4 linkage has identified the MTP gene as a modifier of lifespan in a cohort of long-lived individuals (88). Since the description of MTP as the cause of the rare inherited ABL disease (86, 89), over 30 mutations in MTP have been
identified in more than 50 ABL patients described so far: 21.3% missense, 7% nonsense, 12% small indel, 3% gross indel and 27.7% splicing variants (90-93). The functional characterization of various mutations found in ABL has not only disclosed their effects on the expression, subcellular location and interaction of MTP with protein disulfide isomerase, but has also emphasized requirements for the transfer of both TGs and PLs to support Apo B secretion (94).

3. MTP EXPRESSION IN DIFFERENT TISSUES AND DURING HUMAN DEVELOPMENT

The large subunit expression of MTP along with its transfer activity is predominantly present in differentiated epithelial cells of the small intestinal regions (duodenum, proximal jejunum), but absent in crypt and colonic cells (95-97). Interestingly, MTP was identified in villus and crypt epithelial cells as well in different regions of the human foetal intestine, including the colon (95). Staining was detected as early as the 13th week of gestation in all gut segments and was almost entirely confined to the columnar epithelial cells of the jejunum and colon (95). Also, a trend toward increasing MTP activity was noticed at 20–22 weeks of gestation (95). These observations combined with previous reports demonstrate the small intestine's ability to synthesize and secrete Apo B-containing lipoproteins during development (95, 98-101). The liver also represents a major organ of MTP expression, but hepatocytes constitute the unique cell population containing MTP protein with an expression gradient: a decrease toward the periphery of the lobule opposing the portal triad and an increase in cells proximal to the central vein (96). Interesting findings support the presence of MTP in the kidney and heart, which allows the secretion of their content of Apo B-containing particles, thereby protecting proximal tubules and myocardium, respectively, against accumulation of toxic lipids (102-104). MTP mRNA and protein were also detected in antigen-presenting cells, including monocytes, splenocytes, B and T cells (105) with an involvement of CD1d-restricted lipid antigen presentation (106).
4. FUNCTIONAL CONSEQUENCES OF MTP ABSENCE: ABL CLINICAL SPECTRUM

Mice homozygous for an MTP gene disruption died at ~E10.5, thereby underscoring the essential role of MTP, and the importance of the synthesis and secretion of ApoB-containing lipoproteins during early stages for lipid delivery to embryos (107, 108). Additionally, half-normal levels of the MTP mRNA, protein, and TG transfer activity in tissues of heterozygous mice are insufficient for normal levels of lipoprotein secretion and developmental functions, emphasizing the lack of physiological compensation for reduced levels of MTP gene expression in Mttp+/- (107). In contrast, obligate human heterozygotes exhibited normal plasma lipid and lipoprotein levels, and adult human subjects survive with a near-total absence of Apo B-containing lipoproteins (109), which clearly implies that both alleles of the MTP 97 kDa gene need to be defective in order to observe the major decline in plasma lipids and intestinal absorption, and the recessive character of the ABL disease. So far, we do not know if MTP is present in excess under normal circumstances as losing approx. 50% of its expression is not enough to affect lipoprotein assembly. Since no one has ever measured the levels of MTP in the intestine or liver from human heterozygotes, we also do not know whether the explanation for the flagrant paradox (e.g. humans vs. mice) stems from a potential compensatory up-regulation of MTP expression from the normal allele or is a result of a reduction in MTP mRNA or protein turnover. This could also be due to different embryonic growth rates or maternal–fetal transport methods. Although a number of different in vitro model systems have been employed to address these issues, extrapolation to in vivo physiology might be hazardous. In this context, it is also worth recalling that the placental circulation is established very early during human gestation, and the transport mechanisms for lipids in the placenta and lipid requirements may be quite different from those in the mouse yolk sac.
Consequently to MTP abnormalities in ABL, various multi-system manifestations are noted (Table 1). Most of the reported young patients have diarrhea, steatorrhea, acanthocytosis, low serum cholesterol and Apo B deficiency, which are accompanied with failure to thrive and EFA deficiency. In view of the severe fat malabsorption, transport of fat-soluble vitamins is flawed and even worsened with fat consumption (57).

Elevated serum transaminases with hepatomegaly due to hepatic steatosis have frequently been reported (110-112). Few patients presented with cirrhosis. Apparently, with post transplantation, the profile of the liver and lipoproteins was normal, but intestinal fat absorption persisted as the mutant MTP remains expressed in the intestine (113).

One of the most serious clinical manifestations is at the level of both central and peripheral nervous systems. In fact, a progressive ataxic neuropathic disease and retinopathy develop in later childhood and are probably due to vitamin E deficiency (111, 114, 115). However, one can observe absent tendon reflexes as early clinical signs, which are followed by deep sensory loss in the lower limbs and then a cerebellar syndrome with an ataxic gait, dysmetria, and dysarthria (109). Pes cavus, pes equinovarus, and kyphoscoliosis are frequently encountered. Upper motor neuron signs, including Babinski sign or weakness of legs can be observed in ABL patients. Nevertheless, the primary driving pathology is demyelination (111). Intriguingly, some patients escaped serious affliction until much later in life (111).

Among the wide range of ophthalmic symptoms and manifestations, the most prominent abnormality is pigmentary retinal degeneration (116). Early in the course of disease, patients have loss of night vision and some of them also exhibit loss of color vision. The retinopathy often produces slowly enlarging annular scotomas with macular sparing, such that patients are relatively unaware of the progression of the disease. Complete loss of vision can ultimately occur...
Fundoscopic examination reveals an atypical pigmentation of the retina characterized by small, irregularly distributed, white spots. Electroretinogram and fluorescein angiography investigations have shown the retina to be affected in asymptomatic ABL patients (116). Profound muscle weakness has been described in ABL. Striated and smooth muscles are affected and may represent the cause of premature death among a few ABL patients (117-119). The etiology of myopathy remains unclear although myositis appears to be related to ceroid pigment deposition and muscle weakness to vitamin E deficiency and neuropathy. Death related to cardiomyopathy has been described for some patients (117, 118). Acanthocytosis is among the characteristic hematologic manifestations of ABL. Circulating erythrocytes present abnormal shaped structures that inhibit rouleaux formation and culminate in extremely low erythrocyte sedimentation rates. Anemia has been found in some cases of ABL probably as a consequence of deficiencies of iron, folate and other nutrients secondary to fat malabsorption (110, 117).

5. TREATMENT

As highlighted in a very recent review, early diagnoses, combined with appropriate supplements, help prevent the severe sequelae of ABL (120). Fat intake should be reduced to 5 to 20 g/d, which will decrease steatorrhea while favouring marked clinical improvement and growth acceleration. Nevertheless, the diet has to be supplemented with EFAs (e.g., 5 g corn oil or safflower oil/d) to avoid EFA deficiency. Medium chain TGs are often recommended to subjects with ABL as a caloric substitute for long-chain fatty acids, but under high precautions given their secondary effects such as hepatic fibrosis. The classical treatment also includes supplements of fat-soluble vitamins (E, A, D, and K). Noteworthy, oral alpha-tocopherol supplementation has to be initiated as early as possible to prevent neurological and retinal disability and halt/abrogate
progression of the neuromuscular and myocardiopathy complications associated with this disease (109, 121, 122). If short-term efficacy of high-dose oral vitamin supplements has been proven (123-126), longer-term management in ABL has been little reported. Nevertheless, some studies stressed that combined vitamin E and A therapy initiated before age 2 leads to attenuation of retinal degeneration 10 years later (127).

C. HYPOBETALIPOPROTEINEMIA

1. INTRODUCTION

Familial hypobetalipoproteinemia (FHBL; OMIM 107730), an autosomal co-dominant disorder, is characterized by molecular defects in the APOB gene (Figure 2) on chromosome locus 2p23-24, which interfere with the translation of full-length of Apo B mRNA (128-130). Consequently, the formation of truncated Apo B of various sizes prevents the active export of TG from the intestine by CM and from the liver by VLDL, resulting in intestinal and hepatic TG accumulation. Therefore, abnormally low TG-rich lipoproteins and LDL are observed in blood circulation. As a function of the genetic status, the clinical manifestations may vary from none to neurological, endocrine, haematological and liver dysfunctions.

2. APO B PROPERTIES

The majority of bulk lipid transport is achieved by Apo B-containing lipoproteins (Figure 3). Apo B constitutes the largest glycoprotein that plays a central role in human TG-rich lipoprotein metabolism. The greatest form of Apo B is also the dominant protein in LDL and the ligand for the LDL receptor. Its structure is characterized by a globular amphipathic NH₂ domain spanning the first 15–20% of the polypeptide followed by an extended hydrophobic β-sheet domain from about 20 to 48%, while the rest of the polypeptide is comprised of an α-helical amphipathic region, another long β-sheet domain, and another α-helical domain (131-133). Contrary to the
other Apos that are exchangeable among circulating lipoproteins, Apo B is not transposable since it remains bound to the nascent lipoprotein until its recognition by specific receptors and uptake of the whole Apo B-containing particle in several tissues. Another unique feature is that only one single Apo B molecule is detectable in Apo B-containing lipoproteins. However, this single Apo B molecule is fully sufficient to provide the structural framework for the assembly of TG-rich lipoproteins in the liver and the small intestine (134) (Figure 3). The synthesis of Apo B is not regulated at the transcriptional level, but it rather requires complex posttranslational processing, including lipidation and glycosylation, for proper folding and secretion (135). In fact, degradation by the ubiquitin proteasome system represents the major mechanism for regulation of Apo B secretion whereas the availability of newly synthesized lipids protects Apo B from destruction and serves as the critical factor in targeting Apo B for secretion (136-139). Mechanistically, the translocation arrest provokes a prolonged association of Apo B with the Sec 61β translocon and ribosomes, thereby resulting in an impairment in the elongation stage of Apo B (140-143). As against, efficient ongoing lipid synthesis (during co-translational translocation across the ER) prevents Apo B membrane from slowing or immobilizing, which favours its elongation. Of particular interest, a high physiologic concentration of lipids, provided over longer periods, induces ER stress that decreases the secretion of Apo B-100 (144, 145). It is recognized that the assembly of TG-rich lipoprotein occurs in two steps, one co-translational (the first step) and another post-translational (the second step) where the larger amount of TG is added, likely via fusion of a primordial Apo B lipoprotein particle with an Apo B-free TG droplet in the secretory pathway (146) (Figure 1).

The human Apo B gene covers 43 kb of chromosome 2p and the coding portion of the gene extends over 43 kb and contains 29 exons and 28 introns. It produces two forms of circulating
Apo B, namely Apo B-48 (2152 amino acids) and Apo B-100 (4536 amino acids), by unique mRNA editing process (147-149). This RNA editing mechanism converts a codon (CAA) in the human intestine to a translation stop codon (UAA) at 48% of the full-length coding sequence. The resulting Apo B-48 protein is identical to the amino-terminal 48% of Apo B-100 and is obligatory for CM produced by the small intestine, whereas full-length Apo B-100 is structurally essential for VLDL synthesized by the liver. The remarkable posttranscriptional modification of Apo B mRNA is performed by a multicomponent enzyme complex, termed the C-to-U editosome (150) that contains APOBEC-1, a single catalytic subunit, and other proteins known as auxiliary or complementation factors (150, 151). The editosome has been suggested to favour more efficient absorption (152) and, conversely, low or no Apo B mRNA editing activity may result in relatively higher levels of VLDL and/or LDL as noted in the liver of animals (153). These Apo B-100-containing lipoproteins are more atherogenic than Apo B-48-containing CM. Noteworthy, there is a small amount of Apo B mRNA that escapes editing, resulting in a low level of Apo B-100 expression by the intestine (109).

3. APO B EXPRESSION IN DIFFERENT TISSUES AND DURING HUMAN DEVELOPMENT

If previously the faith was that only the small intestine and liver have the capability to assemble and secrete Apo B-containing lipoproteins (Figure 3), it now turns out that various organs are able enough to do it. Proximal tubule cells of mammalian kidney produce Apo B-containing lipoproteins and, conversely, inhibition of Apo B expression increases fasting-induced lipid accumulation in the kidney cortex (64, 102, 128). Apparently, the size and density of kidney-derived Apo B-containing lipoproteins depend on lipid availability (154). The human heart also expresses the Apo B gene in addition to the MTP gene (103). Likely, the mandatory presence of Apo B and MTP in cardiac myocytes allows the output of Apo B-containing lipoproteins in order
to prevent pathological TG accumulation in the heart (103, 104, 155, 156). The placenta elaborates Apo B-100-containing particles that participate in lipid transport between mother and foetus (157, 158). The importance of this process is underscored by the increased lethality following the disruption of Apo B and MTP in the yolk sac of mice (107, 159, 160), which probably assures the early delivery of fat-soluble nutrients from the yolk sac to the embryo by Apo B-100-containing particles packaging and discharge. Ocular Apo B synthesis was also noted (161). Lipid overload in retinal pigmented epithelium possibly triggers the local machinery to produce Apo-B-containing lipoproteins to avoid apoptosis or degeneration of the tissue (162).

The human foetal intestine was demonstrated to possess an efficient lipoprotein-lipid transport system during development (163, 164). The assembly and secretion of TG-rich lipoproteins were active in the jejunum and colon of the human foetus (165, 166). For this task, it early produces Apo B-100, but the switch in dominance from Apo B-100 to Apo B-48 mRNA takes place later during development (99, 100, 167-170). These processes are highly regulated by many hormones (167, 171, 172).

Taken together, there is now clear evidence that the intracellular assembly process is driven by Apo B. Thus, accidents of nature limiting Apo B production can have adverse effects on the export of lipids with potentially toxic fat accumulation in the different tissues and with a significant impact on TG-consuming organs. As expected from the central role of the Apo B-containing lipoproteins in delivering lipids, antioxidant vitamins and fuel to cells, mutations of Apo B may profoundly affect development.

4. FUNCTIONAL CONSEQUENCES OF APO B ABSENCE: FHBL CLINICAL SPECTRUM

To date, more than 45 truncations in the Apo B gene have been reported and most of them are frequently due to mutations, exon deletions and splicing variations (130, 173). The Apo B
truncations have traditionally been named according to a centile system, with a wide range from Apo B-2 to Apo B-90 relatively to normal Apo B-100 (174-197). The various truncations give rise to different sizes, densities, functions and metabolism of lipoprotein fractions (198). In particular, they are characterized by a lower production rate and higher clearance rate, thereby contributing to abnormally reduced concentrations of circulating Apo B (179, 199-202), while fragments of Apo B<27.6 are undetectable (199). Even in heterozygous subjects with FHBL, the concentrations of Apo B do not exceed 30% of the normal values (203, 204).

FHBL heterozygotes may be asymptomatic whereas homozygous FHBL patients present with steatorrhea, intestinal fat malabsorption, deficient absorption of EFA and lipid-soluble vitamins (A, D, E and K), hypocholesterolemia and Apo B deficiency along with neurological, ocular, endocrine, hematological abnormalities (205-207) (Table 1). Affected individuals may exhibit red cell acanthocytosis and retinitis pigmentosa. Sometimes fatty liver is accompanied by mild elevation of serum liver enzymes. In fact, sophisticated techniques could detect nonalcoholic fatty liver disease in FHBL subjects (187, 208-213).

5. TREATMENT

Although homozygous FHBL and ABL have a different genetic basis, they share similar signs, symptoms and laboratory findings. Likewise, the clinical follow-up and management are comparable for the two disorders. As for ABL, it is mandatory to pay special attention to growth monitoring and prevention of complications in paediatrics by offering specialized dietary advice and fat-soluble vitamin treatments.

D. CHYLOMICRON RETENTION DISEASE

1. INTRODUCTION
Chylomicron retention disease (CRD, OMIM 246700), or Anderson disease, is an autosomal-recessive condition caused by mutations in the \textit{SARA2} gene encoding the Sar1b protein. Contrary to ABL and FHBL, the synthesis of pre-CM occurs in the ER, but without the possibility to reach the Golgi apparatus (Figure 2). Lipids accumulate in the intestine and liver, while there is a selective absence of postprandial Apo B-48 and CMs. Thus, the young patients experience fat malabsorption, failure to thrive and steatorrhea. Almost 50 cases have been reported so far in the literature.

2. \textit{SAR1B PROPERTIES}

Trafficking of CM-contained vesicles through the early secretory pathway is mediated by coat protein (COPII), a process requiring the small Sar1b GTPase for the exchange of GDP for GTP (214). Activated Sar1b initiates vesicle formation by recruiting first the inner COPII coat components (Sec23 and Sec24) and subsequently the components of the outer flexible coat (Sec13/Sec31) able to accommodate various sizes of vesicles (215). The mature coated vesicles bud from the ER and reach the Golgi apparatus where CM transit prior to their discharge into the intercellular space and their transfer to the lymphatics before flowing into the blood circulation. Genetic defects in Sar1b GTPase inhibit the step of pre-CM trafficking to the Golgi (Figure 2). The obligatory role of Sar1b GTPase is evidenced by the presence of its paralogue Sar1a GTPase (216-218) that is 90\% identical, differing only by 20 amino acid residues, but does not compensate for the lack of the Sar1b protein in CRD (219). Of note, if Sar1a could not compensate for loss of Sar1b in the gut, only limited data are, however, available regarding its contributions in the liver despite the fact that VLDL secretion was not substantially affected in CRD (220). Nevertheless, a recent study has provided strong evidence that Sar1b promotes the secretion of TG-rich, Apo B-containing lipoproteins from the liver, which would neatly explain
the counter-intuitive observation that some CRD children develop hepatic steatosis, despite severe intestinal fat malabsorption (221). Furthermore, \textit{SAR1b} has been shown to be the predominantly expressed isoform in human jejunum and liver. Although Sar1a antagonizes the lipoprotein secretion-promoting activity of Sar1b, both isoforms were noted to modulate the expression of genes encoding cholesterol biosynthetic enzymes and the synthesis of cholesterol \textit{de novo} (221). Noteworthy, Sar1b GTPase is not only central for COPII responsible for the biosynthetic transport of proteins from the ER to the Golgi apparatus, but also for fusion of the specific CM transport vesicle, the pre-CM transport vesicle, with the Golgi (50, 54, 222, 223). The 3D structure of Sar1b protein is formed by 6 $\alpha$-helices and 6 parallel $\beta$-strands that lead to a hydrophobic $\beta$-sheet sandwiched between 3 $\alpha$-helices (224-226). The NH2-terminus segment comprises the site interacting with Sec 12 and the two GTP binding and hydrolysis sites (224, 226-228), while the C-terminus segment includes the $\alpha$-6 helix and the regulation loop (224, 225). Importantly, the N-terminus part allows the anchorage of Sar1b-GTP complex on the ER membrane, GTP-binding and hydrolysis, whereas the C terminus regulates interactions of Sar1b with the membrane. All the mutations affecting C- or N-terminus provoke failure to secrete CMs albeit with diverse phenotypes among CRD patients (229). Additional studies are necessary to examine the functionality of the mutated proteins, thereby providing further insights into this disease as well as on the normal pathway for the export of CM.

3. \textit{SAR1B} EXPRESSION IN DIFFERENT TISSUES AND DURING HUMAN DEVELOPMENT

Despite growing knowledge on Sar1b GTPase, little is known about its tissue distribution and developmental regulation. Evaluation of \textit{Sar1b} mRNA revealed the skeletal muscle as the tissue with the highest \textit{Sar1b} expression, followed by the heart and liver, the organs composing the digestive tract, the brain and finally the lung and the adipose tissue (230). Sar1b protein
expression levels follow a similar pattern among the organs, except for its higher expression in the heart. The abundant expression of Sar1b in the skeletal muscle and heart suggests the highly specialized role of Sar1b in these particular tissues, including the regulation of calcium trafficking among multiple calcium storage organelles, for instance the sarcoplasmic reticulum and the ER as reported previously (231-234). Accordingly, it was reported that patients with CRD suffer from myolysis, cardiac abnormalities and elevated creatine kinase levels (235).

The proteins from the Sar1 family have not previously been reported as being involved in morphogenesis and development. Intuitively, the importance of the small GTPase classes in this process is predicted given their crucial functions in vesicular trafficking between the membranes of the ER and the Golgi apparatus as noted in the initial stage of root hair (236) and axon (237) development, the membrane recruitment of cargo-sorting coat proteins, the modulation of membrane lipid composition, and the interaction with regulators of other G proteins. Evidently, additional efforts must be invested to support the possibility that the modulation of COPII-like trafficking machinery by Sar1b is active during development.

4. FUNCTIONAL CONSEQUENCES OF SAR1B ABSENCE: CRD CLINICAL SPECTRUM

To date, about twenty gene defects have been described in SARA2, including missense or nonsense mutations, which affect splice acceptor and donor sites or correspond to insertions, deletions and duplications, thereby disrupting coding sequences. Heterozygous carriers remain asymptomatic, whereas homozygous present within the first few months or years of life with failure to thrive and diarrhea (Table 1). Furthermore, the young children exhibit white coating mucosa because of fat-laden villous enterocytes. Epithelial cells also show marked accumulation of large lipid droplets in the cytoplasm along with lipoprotein-sized structures in membrane bound compartments. The postprandial state is characterized by absence of TG elevation, Apo B-
48 and CMs in response to a fat meal test, confirming the typical steatorrhea in CRD patients. These abnormalities are in line with closed juxtaposition of the intercellular spaces in the mucosa and are accompanied with severe hypocholesterolemia and low concentrations of total lipids, PLs, lipid soluble vitamins, EFAs, LDL and HDL, and Apos (B and AI) in the plasma. Clinically, electromyographic irregularities and diminished osteotendinous reflexes were common, but areflexia was rarely observed. Ophthalmological manifestations of vitamin A and E deficiencies were limited to electrophysiological anomalies detected by electroretinograms and evoked potentials. Hepatic steatosis is detected only in few patients. Common at the time of diagnosis, the aforementioned anomalies proved largely reversible with treatment. Although the investigation of the Canadian subjects with the allele 409G>A reveals a more severe degree of hypocholesterolemia and few clinical parameters, no genotype–phenotype correlation has been evidenced (238).

5. TREATMENT

As per ABL and FHBL, CRD patients have a significant amelioration when treated with a low fat diet and supplementation with fat-soluble vitamins. To distinguish between the biochemical and clinical phenotypes, as well as management of the three congenital malabsorptions, a summary is presented in Table 2.

E. CONGENITAL HYPOCHOLESTEROLEMIA IDENTIFIED IN PATIENTS WITH LOSS–OF–FUNCTION MUTATIONS OR PCSK9 VARIANTS

Proprotein convertase subtilisin/kexin type 9 (PCSK9), a serine protease expressed mainly in liver and intestine, is strongly involved in LDL metabolism (239). Its gene is localized on human chromosome 1p32 and encodes a 692–amino acid proteinase K–like serine protease that has as a central role in regulation of cholesterol homeostasis, essentially by targeting the receptor to
degradation, leading to reduced LDL-cholesterol clearance from the circulation (240). If the dominant gain of function mutations in the PCSK9 gene cause a phenotype similar to autosomal dominant familial hypercholesterolemia (241), loss of function variants are associated with hypocholesterolemia and protection against coronary artery disease (242–244). For example, R46L is a loss of function PCSK9 mutation since R46L-PCSK9 undergoes near a 16% increase in cell surface LDL receptors and a 35% increase in internalized LDL compared with wild-type-PCSK9, suggesting that R46L causes hypocholesterolemia through a decreased ability to degrade LDL receptors (245). Other loss of function mutations may have even drastic effects on cholesterolemia since they lower circulating LDL-cholesterol levels below \( \approx 0.4 \) mmol/L (244, 246). So far, humans can survive and stay healthy as also confirmed in \( Pcsk9 \)-knockout mice (247, 248). However, this does not seem to be the case in some lower vertebrates where knockdown of PCSK9 mRNA in zebrafish leads to disorganization of the nervous system and lethality.

**F. CONGENITAL HYPOCHOLESTEROLEMIA IDENTIFIED IN PATIENTS WITH ANGPTL3**

A form of familial combined hypolipidemia is due to defects in the *Angiopoietin-like 3 protein* (*ANGPTL3*) gene in a family with primary hypocholesterolemia (249). ANGPTL3 is located on **1p31.3**, exhibiting a signal peptide, an N-terminal coiled-coil domain and a C-terminal fibrinogen-like domain. The mode of inheritance and the clinical implications of familial combined hypolipidemia are not well defined. ANGPTL3 is located primarily in the liver and regulates lipid metabolism (250). Through its N-terminal region, it acts as a dual inhibitor of lipoprotein lipase and endothelial lipase and increases plasma HDL-cholesterol (251, 252). Patients with the loss-of-function mutation in ANGPTL3 have extremely lower plasma TG,
LDL- and HDL- cholesterol levels than individuals with no mutation (249, 253). Accordingly, mice with the loss of ANGPTL3 expression display lower levels of TGs and HDL-cholesterol (250, 254, 255), whereas ANGPTL3 injection or overexpression increases circulating lipid levels (250, 251). Diabetes and cardiovascular disease were absent in homozygotes, raising the possibility that absence of ANGPTL3 is protective for these conditions. The prevalence of ANGPTL3 mutations giving rise to a combined hypolipidemia phenotype in subjects with severe primary HBL is about 10% (256). Of subjects with total cholesterol concentration below the second percentile, those with HDL-cholesterol concentration below the second decile may be carrying ANGPTL3 mutations, whereas those with higher HDL-cholesterol concentrations may be carrying APOB mutations (256).

**G. POLYMORPHISMS OF APO B AND MTP AND LIPID CHANGES**

To date, several variants on Apo B and MTP genes have been detected and studied due to their plausible role in the modulation of lipid/lipoprotein profiles and postprandial lipemia. One must be aware about this phenomenon since it may explain the low and high postprandial responses in relation with intestinally or hepatically derived TG-rich lipoproteins and the risk of myocardial infarction/premature coronary heart diseases.

Numerous polymorphisms of the Apo B gene have been described with an effect of the Ins/Del polymorphisms on lipid levels (257-259), and the kinetics of the secretion of VLDL has already been found (260). Noteworthy, the XbaI polymorphism was related to the inter-individual variability observed during postprandial lipemia, showing a significantly augmented or reduced postprandial response (261, 262). Particularly, a variable number of tandem repeats polymorphism, which is located 75 bp downstream the second polyadenylation signal at the 3’ end of the Apo B gene (2p24-p23), has been found to be common in some ethnic groups (263-
They were associated with modifications of lipid concentrations (267-271) and the risk of coronary heart diseases (270-272). Nevertheless, not one of these associations has been consistently observed in a large number of studies (273, 274). Discrepancies may be related to differences of ethnic groups and environmental factors.

As mentioned before, mutations in the MTP gene have been established in cases of ABL (90-93). Several of these genetic abnormalities (premature stop codons, mutations in canonical splice sites, or frameshift mutations) have been informative as to MTP function status. However, in many cases, predictions were difficult to establish (275, 276) despite the co-segregation of the genetic defects with the clinical phenotypes. With additional determinations of MTP activity (48, 92) and by scrutinizing the repercussions of intronic mutations (277) on intestinal or hepatic biopsies, as was performed by various groups, a clear picture of the genotype-phenotype relationship will certainly be obtained.

The most studied promoter polymorphism at the MTTP locus (−493G/T, located 493 bp upstream from the transcriptional start site) lowers the expression of MTP while reducing the formation and secretion of CMs and VLDLs (278-281). There is also evidence that the MTP-493G/T polymorphism modulates postprandial Apo B-48 and Apo B-100 of TG-rich lipoproteins (282). Various groups reported an association between the MTP -493T allele and low levels of serum TG, total cholesterol, LDL-cholesterol and Apo B (58, 283-286). However, other studies revealed the opposite (287-289) or detected no relationship between this polymorphism and any lipid phenotype (290), demonstrating a huge gap across races on MTP -493T, which might be attributed to profoundly different evolutionary pressures at this locus.
Little is known as to the role of Sar1b polymorphisms on the inter-individual variability of the postprandial response although studies have shown potential genotype-phenotype links (229) and elevated output of TG secretion in response to Sar1b overexpression (291, 292).

H. INTESTINAL LIPID FLUX, LIPID HOMEOSTASIS AND CARDIOMETABOLIC DISORDERS

It is now well established that the small intestine is a significant determinant of postprandial dyslipidemia, cardio-metabolic disorders and atherogenesis. Various groups have pointed out the direct role of intestinally derived TG-rich lipoproteins in the progression of atherosclerosis, particularly during insulin resistance and type 2 diabetes mellitus (293, 294). Clearly, these disorders favour the increased basal rate of Apo B-48-containing lipoprotein secretion in fed and fasting states (295, 296). Demonstration has been obtained in various animal models (297-299) as well as in humans (300, 301). The contribution of the intestinal Apo B 48-containing lipoproteins to the promotion of atherosclerosis is increasingly being recognized as (i) Apo B-48 is correlated with postprandial lipemia (302), carotid intima-media thickness (303) and arterial disease (304, 305); and (ii) CM remnants have access to and accumulate in the sub-endothelial space, thereby triggering the formation of atherosclerotic lesions (306-309). Therefore, one should be aware of the danger of overproduction of intestinal lipoprotein particles in cardio-metabolic states such as obesity, insulin resistance and diabetes.

I. CONCLUSIONS

We have emphasized that mutations in Apo B-48, MTP and SARA2 genes result in low or absent lipid, LDL-cholesterol and Apo B levels. They cause intestinal fat malabsorption along with deficiency of EFA and liposoluble vitamins, thereby triggering various clinical disorders. Genetic defects in PCSK9 and ANGPTL3 also lead to familial hypobetalipoproteinemia with an
evident impact on metabolic and biochemical disorders. We have presented an update on management strategies. We have also documented the postabsorptive concentrations of plasma TGs in response to particularly common polymorphisms of Apo B and MTP. Definitely, the intestine exerts important influences not only on fat malabsorptions, but also on disease-related abnormalities of postprandial lipoprotein metabolism, including insulin resistance, type 2 diabetes and atherosclerosis. Molecular testing should be available to rapidly define molecular aberrations and prevent complications. Hopefully, new nutritional and genetic developments will offer opportunities in the next future to develop strategies to target the intestine in order to enhance fat absorption in congenital malabsorptions on the one hand, and reduce postprandial lipemia and prevent atherosclerosis, on the other hand.
ACKNOWLEDGMENT

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FINANCIAL DISCLOSURE

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REFERENCES


55. Siddiqi, S., and C. M. Mansbach. 2012. Phosphorylation of Sar1b protein releases liver fatty acid-binding protein from multiprotein complex in intestinal cytosol enabling it to bind to endoplasmic reticulum (ER) and bud the pre-chylomicron transport vesicle: *J. Biol. Chem.* **287**: 10178-10188.


of vitellogenin provides a molecular model for the assembly and secretion of atherogenic lipoproteins: *J. Mol. Biol.* **285**: 391-408.


180. Young, S. G., S. T. Hubl, R. S. Smith, S. M. Snyder, and J. F. Terdiman. 1990. Familial hypobetalipoproteinemia caused by a mutation in the apolipoprotein B gene that results in a truncated species of apolipoprotein B (B-31). A unique mutation that helps to define the


transport machinery coordinates cellular lipid secretion and cholesterol biosynthesis: *J. Biol. Chem.* **289**: 4244-4261.


five mutations in the SAR1B gene on the structure and the functionality of Sar1b protein:  


protein gene-493T variant lowers cholesterol but increases the risk of coronary heart disease: *Circulation* **109**: 2279-2284.


FIGURE LEGENDS

Figure 1: Schematic intracellular network required for lipid metabolism, Apo B biogenesis and chylomicron assembly in the enterocyte.

Digestion takes place in the intestinal lumen where triglycerides (TG), cholesteryl esters (CE) and phospholipids (PL) are hydrolyzed by pancreatic lipase, cholesterol esterase and phospholipase A2, respectively. The lipolytic products (FA, 2-MG, FC, lyso-PL) are emulsified into mixed micelles by bile salts before their diffusion through the unstirred water layer. At the apical surface, they enter the enterocyte via passive diffusion when they are in high concentrations. However, protein transporters (e.g., FABPm) are required at weaker concentrations. Once inside the enterocyte, the lipolytic products are bound to cytoplasmic FABPs (I- and L-FABP) in order to migrate to the endoplasmic reticulum (ER) where they are re-esterified by various enzymes: Monoacylglycerol acyltransferase (MGAT) and diacylglycerol acyltransferase (DGAT) for the conversion of 2-MG to TG, ACAT-2 for the esterification of FC into CE, and an enzymatic system (glycerophosphate acyltransferase, phosphatidate phosphodiesterase, lyso-PC acyltransferase) for the production of PL. The synthesis of Apo B-48 and its interaction with microsomal triglyceride transfer protein (MTP), a heterodimeric complex with the chaperone protein protein disulfide isomerase (PDI), through the NH2-terminal domain, is essential for pre-chylomicron (CM) assembly. This packaging process starts when the growing Apo B-48 polypeptide is cotranslationally lipilated by MTP in the lumen of the ER: the association of Apo B-48 with lipids is required not only to ensure proper initiation of folding, but also to prevent its destruction by the proteasomal degradation pathway. The formation of pre-CM transport vesicles is critical for the transport of the nascent lipoproteins to the Golgi, a process mediated by Sar1b GTPase. The maturation of pre-CMs is completed in the Golgi by
addition of lipids and glycosylation of Apos, yielding a mature CM particle, characterized by TG and CE in its core, and FC, PL and Apo B on its surface. Finally, CMs are released by the enterocyte into the intestinal lymph through exocytosis.

MG: monoacylglycerol; FC: free cholesterol; I-FABP: Intestinal-Fatty cid Binding Protein; L-FABP: liver-FABP; PC: phosphatidylcholine

**Figure 2:** Congenital disorders of intestinal lipid absorption. Defects in the key proteins Apo B-48, microsomal triglyceride transfer protein (MTP) and Sar1b GTPase result in the genetic disorders characterized by familial hypobetalipoproteinemia (FHBL), abetalipoproteinemia (ABL) and chylomicron retention disease (CRD), respectively.

**Figure 3:** Schematic structure of plasmatic CM and VLDL

Chylomicrons (CM), secreted by the intestine, and VLDL delivered by the liver, appear as spherical particles in the plasma. They are composed of a hydrophobic center [triglycerides (TG) and cholesteryl esters (CE)] with an external monolayer of lipids [(phospholipids (PL) and unesterified cholesterol (FC)] along with different Apos. The main form of Apo Bs in CMs and VLDLs are Apo B-48 and Apo B-100, respectively.
Table 1: Clinical phenotypes of the main congenital malabsorption disorders

<table>
<thead>
<tr>
<th>Condition</th>
<th>CRD</th>
<th>ABL</th>
<th>HBL homozygous</th>
<th>HBL heterozygous</th>
</tr>
</thead>
<tbody>
<tr>
<td>Retinopathy</td>
<td>+</td>
<td>+++</td>
<td>+++</td>
<td>-</td>
</tr>
<tr>
<td>Myopathy</td>
<td>+</td>
<td>+++</td>
<td>+++</td>
<td>+/-</td>
</tr>
<tr>
<td>Neuropathy</td>
<td>+</td>
<td>+++</td>
<td>+++</td>
<td>+/-</td>
</tr>
<tr>
<td>Cardiomyopathy</td>
<td>+/-</td>
<td>++</td>
<td>++</td>
<td>-</td>
</tr>
<tr>
<td>Hepatic steatosis</td>
<td>+</td>
<td>++</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Steatorrhea</td>
<td>++</td>
<td>+++</td>
<td>+++</td>
<td>+/-</td>
</tr>
<tr>
<td>Acanthocytosis</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+/-</td>
</tr>
</tbody>
</table>

ABL: abetalipoproteinemia, HBL: hypobetalipoproteinemia, -: none, +/-: possible, +: low intensity, ++: moderate intensity, +++: high intensity
<table>
<thead>
<tr>
<th>Disorder</th>
<th>Gene</th>
<th>Biochemical phenotype</th>
<th>Clinical phenotype</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abetalipoproteinemia</td>
<td>MTP</td>
<td>- Ø LDL cholesterol</td>
<td>- Failure to thrive</td>
<td>- Low fat (&lt;30 % of total calories) with reduced</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Ø Chylomicrons</td>
<td>- Abdominal distension</td>
<td>LCFA</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Ø VLDL</td>
<td>- Lipid malabsorption</td>
<td>- PUFA</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ Triglycerides</td>
<td>- Spinocerebellar degeneration</td>
<td>Vitamin E 100–300 IU/kg/d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ Vitamins (A, D, E, K)</td>
<td>- Night blindness</td>
<td>Vitamin A 100–400 IU/kg/d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Ø Apo B</td>
<td>- Coagulopathy</td>
<td>Vitamin D 800–1200 IU/d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ Other apolipoproteins</td>
<td></td>
<td>Vitamin K 5–35 mg/w</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Altered lipoprotein composition</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypobetalipoproteinemia</td>
<td>APOB</td>
<td>Heterozygous</td>
<td>Heterozygotes</td>
<td>- Low fat (&lt;30 % of total calories), with reduced long-chain fatty acids</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ LDL cholesterol (less than 30 % of levels normal for age and sex)</td>
<td>- Asymptomatic</td>
<td>- PUFA</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Homozygous</td>
<td>Homozygous</td>
<td>Vitamin E 100–300 IU/kg/d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Ø or ↓ LDL cholesterol</td>
<td>- Gallstones</td>
<td>Vitamin A 100–400 IU/kg/d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ Triglycerides</td>
<td>- Insulin sensitivity</td>
<td>Vitamin D 800–1200 IU/d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Ø Apo B</td>
<td>- Hepatic cirrhosis</td>
<td>Vitamin K 5–35 mg/w</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ Vitamins (A, D, E, K)</td>
<td>- Hepatocarcinoma</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ Other apolipoproteins</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chylomicron retention disease</td>
<td>SARA2</td>
<td>- Ø Chylomicrons</td>
<td>- Anthropometry (failure-to-thrive)</td>
<td>- Low fat diet</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ LDL cholesterol (less than 50 % of levels normal for age and sex)</td>
<td>- Digestive (diarrhea, vomiting, abdominal distension)</td>
<td>- Enriched in EFA (vegetable oils, fish…), vitamin E: 50 IU/kg/day</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- anemia</td>
<td>- Hepatomegaly</td>
<td>- Vitamin A: 15,000 IU/d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ Total cholesterol</td>
<td>- Neurological abnormalities</td>
<td>(adjust according to plasma levels)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ HDL</td>
<td>(areflexia, ↓ deep proprioception)</td>
<td>- Vitamin A: 800–1200 IU/kg/d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- EFA deficiency</td>
<td>- Malnutrition (accumulation of lipid droplets in enterocytes)</td>
<td>- Vitamin K: 15 mg/w</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- ↓ Vitamins</td>
<td>- Anemia</td>
<td>(adjust according to INR and plasma levels)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>One perfusion</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>- Fatty acids: intralipid 20% 2g/kg/month</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>- Vitamin E: 4 to 6 mg/kg/month</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>- Vitamin A: 500 IU/kg/month</td>
</tr>
</tbody>
</table>

Apo: apolipoprotein; EFA: essential fatty acids; HDL: high density lipoprotein; INR: International normalized ratio; LCFA: long chain fatty acids; LDL: low density lipoprotein; PUFA: polyunsaturated fatty acids; VLDL: very-low density lipoprotein
Figure 1

Intraluminal Alimentary Lipids

FA
Phospholipase A2
PL
Lyso-PL

2 FA
Pancreatic Lipase
TG

2-MG
FA
Cholesterol esterase
CE

Figure 1

Apical compartment

Digestive products

I-FABP
L-FABP

FABPm

Basolateral compartment

Chylomicron

Sterol plants
Free cholesterol
Fatty Acids
Lyso Phospholipids
Monoacylglycerols
Figure 2

APO B defects

MTP defects

Sar1b GTPase defects

CRD

FHBL

ABL
Very-low density lipoprotein