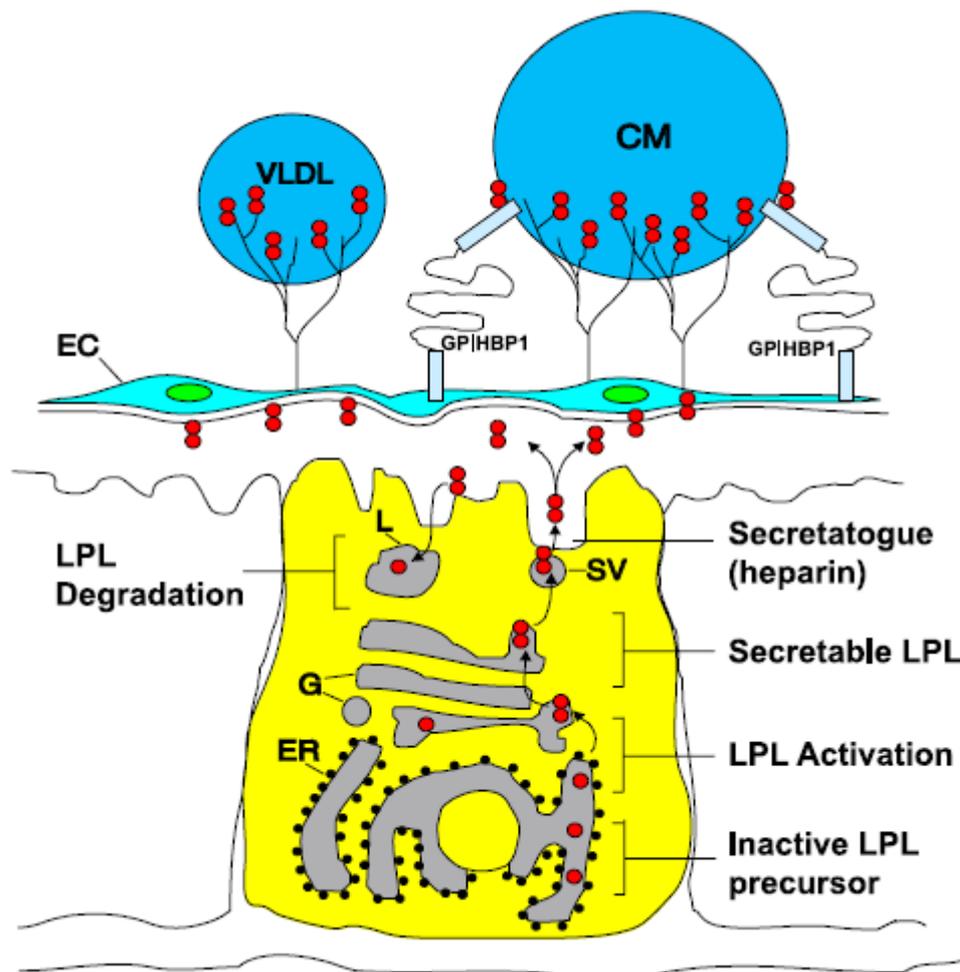


## Lipoprotein lipase

**Background information:** students are supposed to know the major types of lipoproteins, their formation and catabolism (see lectures and textbooks). The summary below emphasizes only a few points for seminar discussion.

### The lipoprotein lipase

Lipoprotein lipase (LPL) hydrolyzes triglycerides found in chylomicrons (CHY) and VLDL, it cleaves the *sn*-1 and *sn*-3 ester bonds, preferentially. It possesses also phospholipase activity (approximately 20-fold lower than its triacylglycerol hydrolase activity), which is necessary for opening the phospholipid shell and to reach the hydrophobic core of the lipoprotein particles. Lipoprotein lipase is present in many tissues, including adipose tissue, striated muscle, lactating mammary gland and heart. The enzyme is synthesized in the parenchymal cells within the tissue (i.e. adipocytes, and muscle cells) and is secreted after posttranslational modifications. It passes through the capillary endothelium and binds non-covalently to heparan sulfate carbohydrate chains of the glycocalyx present at the luminal surface of endothelial cells. Throughout its action the enzyme remains on the surface of the capillary endothelial cells. In the adipose tissue insulin increases LPL activity by transcriptional and posttranslational mechanisms. On the contrary, LPL activities in skeletal muscle and heart are higher in starvation compared to activities in the well-fed state. Physical exercise enhances LPL activity in skeletal muscle, whereas prolactin induces the enzyme in lactating mammary gland.



The synthesis and posttranslational modifications of LPL (from Ref. 1)

## **The hepatic lipase**

LPL is not expressed in adult liver, however, hepatic lipase (HL) present in the liver sinusoids is a lipid hydrolase with properties somewhat similar to lipoprotein lipase. Hepatic lipase acts on IDL and HDL2. In addition to the triglycerides found in the core of these lipoproteins it cleaves surface phospholipids, as well. Hence, hepatic lipase is involved in the hepatic processing of IDL (either uptake by the liver or its conversion to LDL). Likewise, hepatic lipase plays a role in the conversion of HDL2 back to HDL3.

LPL, HL, and the pancreatic lipase (PL), although encoded by different genes, belong to the same family of lipases and share structural homology. They all have a Ser-His-Asp triad (characteristic otherwise for Serine proteases) in their catalytic sites, and both LPL and PL require a protein cofactor to maintain their catalytically active conformation. This cofactor for the PL is the co-lipase produced by the pancreas, whereas apo CII on the surface of CHY and VLDL is required for LPL activity.

## **Case report**

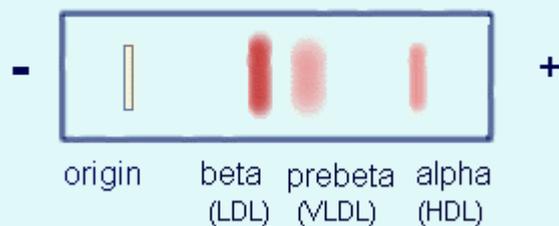
An 11-year-old boy had a long history of abdominal complaints, including bouts of abdominal pain so severe that narcotics were required for relief. These episodes were intermittent, occurring every 6 months on average. On one occasion abdominal surgery (an exploratory laparotomy) was performed, and the patient's appendix was removed. However, this did not correct the problem. The patient had recently felt well until he suddenly developed another episode of abdominal pain. His mother stated that the illness came on 4 hr after he had eaten a meal consisting of pork chops, fried potatoes, milk, ice cream topped with generous serving of whipped cream. No one else in the family had been made ill by this meal. Examination of the patient indicated an acute abdominal emergency. The patient was brought to the hospital at 8:00 AM, 14 hr after his last meal. On arrival a blood specimen was drawn. Within 15 min the laboratory technician reported that valid results could not be obtained from the blood plasma because it was cloudy. The plasma was "milky" but on centrifugation for 30 min at 15,000 rpm, it cleared considerably and there was a thick band of "cream" floating on the top of the specimen. Careful physical examination revealed eruptive xanthomatosis (white to yellow cutaneous nodules raised on erythematous base on the buttocks and elbows). The enlargement of liver and spleen (hepatosplenomegaly) was also detected.

The patient's clinical condition has greatly improved after a low-fat diet was introduced.

## **Case discussion**

1, In clinical laboratory practice blood is drawn „on empty stomach” which means that the patient had dinner the previous evening, and was fasting until the morning blood withdrawal, only drinking of water is allowed. Under normal metabolic conditions chylomicrons containing dietary lipids should be cleared from blood by the morning, and fasting plasma samples are normally devoid of CHYs. The presence of CHYs in patient plasma is rather spectacular: instead of getting a yellowish colored, transparent platelet-poor plasma following the sedimentation of the cellular blood elements, chylomicrons (with their density being lower than that of plasma) float on top of the plasma forming a white creamy layer. This phenomenon was observed in the case describe above, and this should raise the suspicion that this patient might have chylomicronemia. In the clinical laboratories the method called lipid electrophoresis is used for the detailed analysis of lipoproteins: lipoproteins are separated based on their different electrophoretic mobilities, thereafter visualized by a lipid staining procedure, and quantitated based on scanning and evaluating the staining densities of the various lipoprotein fractions. Since the electrophoretic mobility of the CHY is practically zero, CHYs remain at the site of sample application (see Fig below).

## Electrophoretic Pattern of Serum Lipoproteins



### 2, The chylomicronemia syndrome

Chylomicronemia means the accumulation of CHYs in blood, it is diagnosed if the plasma triglyceride level reaches the value of 1000 mg/dl (normal fasting plasma TG levels are below 200 mg/dl). Chylomicronemia syndrome is diagnosed if this high plasma TG level is associated with at least one of the following clinical symptoms:

- Eruptive xanthomas : a skin sign, presented mostly on the buttocks and elbows, which looks like yellowish papules rising from a red background („xanthomas”), and their ulceration occurs frequently („eruptive”). Histological examination indicates the accumulation of triglycerides in the histiocytes.
- Lipemia retinalis: the retinal vessels are white because of the high blood lipid level and the retina is salmon-colored
- Abdominal symptoms: abdominal pain, acute pancreatitis with or without liver enlargement (hepatomegaly) and spleen enlargement (splenomegaly). Hepatosplenomegaly is reversible, if the plasma triglyceride level normalizes, so are the sizes of these organs. In the course of the acute pancreatitis the pancreatic zymogens are activated to active proteases, which will destroy the pancreatic tissue and induce inflammation. Acute pancreatitis, in general, is a rather severe condition, that may even lead to the death of the patients (in ca 50 % of the cases). The probability of the pancreatitis correlates well with the plasma TG levels, that is why an upper limit of 500 mg/dl is set as the therapeutical goal.

The chylomicronemia syndrome may develop in several, very different diseases: hormonal dysregulations, cancer, autoimmune diseases, certain drugs can all lead to the so called secondary chylomicronemia. Out of the potential diseases only the familial LPL deficiency and the apoCII defency are discussed in the seminar, both belong to the genetically inherited primary chylomicronemias.

### 3, The LPL deficiency

The familial LPL deficiency is a rare (<1: 10<sup>6</sup>) autosomal recessive disorder, in which the clearance of chylomicrons from blood is diminished. This results in an elevated fasting plasma triglyceride level (VLDL is, however, not accumulated). The plasma triglyceride levels of the heterozygotes are normal or only mildly elevated, and hence, present no spectacular clinical symptoms. Interestingly, although elevated plasma TG level, in general, counts as a risk factor for atherosclerosis, familial LPL deficient patients are not at high risk for atherosclerosis and their propensity for cardiovascular diseases is not enhanced.

The diagnosis of the disease is based on the determination of the LPL activity. Since this enzyme does not circulate in blood, but is anchored on the capillary endothelial cells, heparin administration prior to blood sampling is required.

#### *Effect of heparin and the diagnosis of LPL deficiency*

When heparin is given intravenously, lipoprotein lipase activity is released into the blood plasma, because the affinity of LPL for heparin is ca 40 times higher than that for the heparan sulfate. This is a pharmacologic action of heparin, and heparin is not a physiologic cofactor or

activator for the enzyme. The lipolytic activity that is measurable in the postheparin plasma represents two different enzymes that hydrolyze triglycerides. One of the lipases is hepatic lipase, derived from the liver, which is resistant to inactivation by protamine or 1 M NaCl. The other enzyme, which is of extrahepatic origin and is inhibited by protamine or 1 M NaCl is the lipoprotein lipase. After heparin injection a major part (45-90 %) of lipolytic activity that appears in the blood plasma is protamine and salt resistant, that is, due to hepatic lipase. This procedure also shows a decreased LPL activity in those patients who have a normal LPL enzyme, but are deficient in apoCII, the LPL cofactor. In such patients addition of normal plasma (which contains normal apoCII) to the patient plasma normalizes the LPL activity. Several mutations have been described as the molecular background for the LPL deficiency, these mutations may alter not only the synthesis of the enzyme, but its posttranslational processing and transport to the capillary endothelial cells, as well. An interesting LPL polymorphism has been described in the Framingham Offspring Study looking into the development of cardiovascular diseases. The Ser447X mutant enzyme shows an increased LPL activity, and it seems to be beneficial from the point of view of cardiovascular diseases. Those men, who carry this LPL variant, have a decreased TG level and an increased HDL-cholesterol level, and the incidence of cardiovascular diseases is also decreased (Ref 4).

#### *Therapy of the familial LPL deficiency*

Since the only potentially dangerous symptom is the development of acute pancreatitis, which correlates well with the plasma triglyceride levels, the therapeutic goal is to keep this value below 500 mg/dl. The easiest way to reach this is to keep a low-fat diet, but lipid-lowering medications may also be applied.

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