HIGH SOLUBLE ENDOGLIN LEVELS AFFECT BILE ACIDS AND CHOLESTEROL METABOLISM IN MICE LIVER

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Objectives

Endoglin (CD 105) is transmembrane glycoprotein that acts as receptor for TGF- β and modulates cellular responses to this pleiotropic cytokine. It's soluble form (sEng) is a plasma protein released by the cleavage of the extracellular domain of endoglin by membrane-type metalloproteinase 1.

Plasma levels of sEng are increased in cardiovascular and metabolic diseases including hypercholesterolemia, atherosclerosis, and type II diabetes mellitus. These diseases also affect liver functions and metabolism. However, sEng direct effects on cholesterol metabolism in liver have not been published so far.

Therefore, we hypothesized that high levels of sEng will affect cholesterol and bile acids turnover in liver.

Methods

- Six-month-old transgenic male mice overexpressing human sEng on CBAXC57BL/6J background (**sEng**) and wild type mice (**WT**) underwent *in vivo* study with bile collection for 45 min, with subsequent biochemical and expression analyses.
- Cholesterol, bile acids, and liver tests were determined in plasma. Spectra of bile acids in the bile and stool were evaluated by LC-MS/MS.
- Expressions of enzymes and transporters responsible for cholesterol and bile acids homeostasis in liver and ileum were assessed by qRT-PCR and Western blot. We focused to molecules mediating **cholesterol** uptake into hepatocytes (Sr-b1, Ldlr), synthesis of cholesterol (Hmgcr), cholesterol esterification (Acat2), biliary cholesterol and phosphatidylcholine efflux (Abcg5/8, Mdr2, respectively), and plasma efflux of cholesterol (Abca1). Also were analyzed molecules enabling **bile acids** uptake from plasma into hepatocytes (Ntcp, Oatp2), efflux of bile acids into bile (Bsep, Mrp2), efflux of bile acids back to plasma (Mrp3, Mrp4), bile acids synthesis (Cyp7a1, the rate-limiting enzyme in the catabolism of cholesterol into bile acids, Cyp8b1 and Cyp27a1, enzymes of alternative pathway of BA synthesis). To describe the complex BA **enterohepatic circulation**, expression of Asbt for BA uptake and Ost α/β and Fgf15 for efflux of BA into portal blood.

Results

High levels of sEng decreased plasma cholesterol concentration

Transgenic mice with increased sEng demonstrated **decreased plasma total and LDL cholesterol levels**. This effect paralleled **upregulation of** hepatic Sr-b1 and Ldlr proteins responsible for **cholesterol uptake** into hepatocytes and **increased** Abcg8 protein level for **cholesterol efflux into bile**. Unchanged protein expression of Hmgcr and fractional cholesterol synthesis rate supported increased uptake of cholesterol from blood as the main mechanism of **increased liver and reduced plasma cholesterol concentrations**. **Biliary excretion of phospholipids** was **increased** despite the unchanged expression of their Mdr2 transporter.

Conclusions

- We have shown that high plasma levels of soluble endoglin increase systemic plasma concentrations of bile acids (BA) due to their increased reabsorption in ileum, leading to the increased uptake of BA into hepatocytes, and biliary secretion coupled with BA-dependent choleretic activity.

We propose that modulation of liver cholesterol and bile acids metabolism by sEng may have significant impact on the course of the pathology and therapy of metabolic diseases associated with increased sEng levels.

High levels of sEng increased choleresis and decreased plasma bile acids level

Net bile flow and biliary excretion of hydrophilic bile acids were increased in sEng mice as a consequence of increased hepatic disposition of bile acids caused by increased Cyp7a1 protein expression, the rate limiting enzyme for the bile acids synthesis from cholesterol. Importantly, increased hepatic disposition of bile acids compensated down-regulation of their Bsep transporter for biliary excretion. Increased bile acids within the biliary canaliculi also increased extraction of phospholipids and promoted formation of micelles. sEng **increased enterohepatic circulation of bile acids** via ileal Asbt and Ost β increased expression and thus reduced their plasma concentration thanks to increased uptake into hepatocytes via Ntcp transporter.

Proposed mechanisms of changed biliary transport of CHOLESTEROL induced by high plasma levels of sEng

Proposed mechanisms of changes in enterohepatic circulation of BILE ACIDS induced by high plasma levels of sEng

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