NAFLD: Experimental and pathophysiology

SAT001
Lipoprotein lipase deletion induces a proinflammatory phenotype in liver-infiltrating monocytes during non-alcoholic steatohepatitis
Anja Koop1, Joerg Heeren2, Ansgar W. Lohse1, Johannes Kluwe3.
1University Medical Center Hamburg-Eppendorf, 2Institute of Internal Medicine, Hamburg, Germany; 3University Medical Center Hamburg-Eppendorf, Institute of Biochemistry and Molecular Cell Biology, Hamburg, Germany

Background and Aims: While absent in the healthy liver, hepatic expression of lipoprotein lipase (LPL) is upregulated during non-alcoholic steatohepatitis (NASH). The functional consequence is unclear, however. We have previously shown that LPL deficiency in myeloid cells leads to a more severe NASH phenotype. Published evidence suggests that LPL may influence macrophage polarization. The aim of our current study was to evaluate if LPL deficiency affects myeloid cells during NASH progression.

Method: C57BL/6j mice heterozygous for lysozyme2-cre recombinase were crossed with LPL floxed mice to generate mice with a myeloid cell-specific LPL knockout (LPLΔlysM) and wild type-like littersmates (LPLfl/fl). Mice were fed a high cholesterol, high fat, high caloric diet for 26 weeks to induce a NASH. The NASH phenotype was characterized by histology, qPCR and transaminases. Liver cells were isolated and analyzed by flow cytometry.

Results: We could reproduce our previous finding that myeloid cell-specific LPL knockout aggravates the NASH phenotype with enhanced fibrosis and inflammation in LPLΔlysM mice. Immune cell phenotyping showed increased number of CD8+ T-cells in the livers of LPLΔlysM NASH mice compared to LPLfl/fl NASH mice. LPL deficiency in myeloid cells did not influence the frequency of different myeloid cell populations in liver or blood. The percentage of neutrophils (Ly6Chigh), monocytes (Ly6Cnegative, Ly6Chigh) and liver resident macrophages (F4/80high) remained on a comparable level in both NASH groups. Liver monocytes produced more iNOS indicating a stronger proinflammatory phenotype in liver infiltrating LPL-deficient monocytes in NASH. In contrast, the immunophenotyping of liver resident macrophages and peripheral monocytes did not show any differences between LPLfl/fl and LPLΔlysM NASH mice.

Conclusion: LPL deficiency in myeloid cells aggravates NASH progression. In NASH, LPL appears to restrict a proinflammatory phenotype in liver-infiltrating monocytes but not in liver resident macrophages.

SAT002
A translational mouse model for NASH and advanced fibrosis in association with atherosclerosis
Anita M. van den Hoek1, Nicole Worms1, Anita van Nieuwkoop1, Christa de Ruiter1, Aswin Menke1, Sridhar Radhakrishnan2, Martine C. Morrison1, Kanita Salic1, Robert Kleemann1.
1Department of Metabolic Health Research, Leiden, Netherlands; 2Research Diets, Inc., New Brunswick, United States

Background and Aims: Non-alcoholic steatohepatitis (NASH) is a fast-growing liver disorder in the Western world and is associated with an increased incidence of cardiovascular disease and type 2 diabetes. Animal models adequately mimicking this condition and that display both the metabolic and histological features of human NASH are scarce. We herein investigate whether Ldlr−/−.Leiden mice on a high fat diet represent a suitable NASH model.

Method: Ldlr−/−.Leiden mice were fed high-fat diets (no added cholesterol) containing lard or milk fat for 28 weeks. Effects on body weight, plasma and liver biochemical variables, liver histology, adipose tissue (inflammation) and atherosclerosis (aortic root) were assessed. Additionally, disease induction at earlier timepoints in the milk-fat group were investigated by taking a liver biopsy at t = 12 weeks and sacrifice at t = 22 weeks. The response to treatment (week 18–28) with 10 mg/kg/d FXR agonist obeticholic acid (OCA) on NASH and fibrosis was also evaluated.

Results: Both high-fat diets induced obesity, hyperlipidemia, hyperinsulinemia, and increased ALT and AST levels. Mice on both diets developed progressive macro- and micro-vascular steatosis, hepatic inflammation and fibrosis. OCA treatment significantly reduced hepatic inflammation and fibrosis in both models. Lard-fat diet group had more severe hyperinsulinemia and adipose tissue inflammation, while milk-fat diet group had more severe hepatic inflammation with advanced bridging fibrosis (F3) in all mice after 28 weeks. Another longitudinal study with the milk-fat diet revealed that after 22 weeks on the diet fibrosis was significantly induced, but primarily in F1-F2 stage with occasionally bridging fibrosis. In addition, milk-fat diet induced severe atherosclerotic lesions (primarily type IV and V based on AHA classification) in the aortic root area after 22 weeks.

Conclusion: Ldlr−/−.Leiden mice fed high-fat diets recapitulate features of the metabolic syndrome and NASH with progressive liver fibrosis and simultaneous atherosclerosis development. By adaptation of the fat content of the diet, either insulin resistance and adipose tissue inflammation (lard-based diet) or hepatic inflammation and fibrosis (milk-fat diet) can be emphasized. This represents a novel translational animal model of fibrosing NASH in association with atherosclerosis that can be used to investigate the effects of new drugs, alone (or drugs in combination).

SAT003
Aberrant hepatic protein tyrosine phosphatase receptor type delta expression is a driver of metabolic liver disease
Armando Andres Roca Suarez1,2, Atish Mukherji1,2, Nicolas Brignon1,2, Laurent Mailly1,2, Frank Jühling1,2, Marine Oudot1,2, Sarah Durand1,2, Patrick Pessaux1,2,3, Thomas Baumert1,2,3,4, Joachim Lupberger1,2.
1Institut national de la santé et de la recherche médicale, Institut de recherche sur les maladies virales et hépatiques, Strasbourg, France; 2Université de Strasbourg, Strasbourg, France; 3Pole hepatolo-digestif, institut hospitalo-universitaire, Strasbourg, France; 4Institut universitaire de France (IUF), Paris, France

Background and Aims: Non-alcoholic steatohepatitis (NASH) is a fast-growing liver disorder in the Western world and is associated with an increased incidence of cardiovascular disease and type 2 diabetes. Animal models adequately mimicking this condition and that display both the metabolic and histological features of human NASH are scarce. We herein investigate whether Ldlr−/−.Leiden mice on a high fat diet represent a suitable NASH model.

Method: Ldlr−/−.Leiden mice were fed high-fat diets (no added cholesterol) containing lard or milk fat for 28 weeks. Effects on body weight, plasma and liver biochemical variables, liver histology, adipose tissue (inflammation) and atherosclerosis (aortic root) were assessed. Additionally, disease induction at earlier timepoints in the milk-fat group were investigated by taking a liver biopsy at t = 12 weeks and sacrifice at t = 22 weeks. The response to treatment (week 18–28) with 10 mg/kg/d FXR agonist obeticholic acid (OCA) on NASH and fibrosis was also evaluated.

Results: Both high-fat diets induced obesity, hyperlipidemia, hyperinsulinemia, and increased ALT and AST levels. Mice on both diets developed progressive macro- and micro-vascular steatosis, hepatic inflammation and fibrosis. OCA treatment significantly reduced hepatic inflammation and fibrosis in both models. Lard-fat diet group had more severe hyperinsulinemia and adipose tissue inflammation, while milk-fat diet group had more severe hepatic inflammation with advanced bridging fibrosis (F3) in all mice after 28 weeks. Another longitudinal study with the milk-fat diet revealed that after 22 weeks on the diet fibrosis was significantly induced, but primarily in F1-F2 stage with occasionally bridging fibrosis. In addition, milk-fat diet induced severe atherosclerotic lesions (primarily type IV and V based on AHA classification) in the aortic root area after 22 weeks.

Conclusion: Ldlr−/−.Leiden mice fed high-fat diets recapitulate features of the metabolic syndrome and NASH with progressive liver fibrosis and simultaneous atherosclerosis development. By adaptation of the fat content of the diet, either insulin resistance and adipose tissue inflammation (lard-based diet) or hepatic inflammation and fibrosis (milk-fat diet) can be emphasized. This represents a novel translational animal model of fibrosing NASH in association with atherosclerosis that can be used to investigate the effects of new drugs, alone (or drugs in combination).