A translational mouse model for NASH and advanced fibrosis in association with atherosclerosis

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INTRODUCTION

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/fibrosis are scarce.

AIM

To investigate whether Ldlr-/-.Leiden mice on a high fat diet represent a suitable and rapid NASH/fibrosis model to study severe stages of fibrosis in the context of obesity and associated insulin resistance and CVD.

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. The response to treatment (week 18-28) with 10 mg/kg/d FXR agonist obeticholic acid (OCA) on NASH and fibrosis was also evaluated.
- 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.

Diet name:	High fat diet (HFD)	Fast food diet (FFD)
Fat	45% kcal Lard	41% kcal Milk fat
Protein	20% kcal Casein	14% kcal Casein
Carbohydrate	35% kcal a.o. Sucrose	44% kcal a.o. Fructose

RESULTS

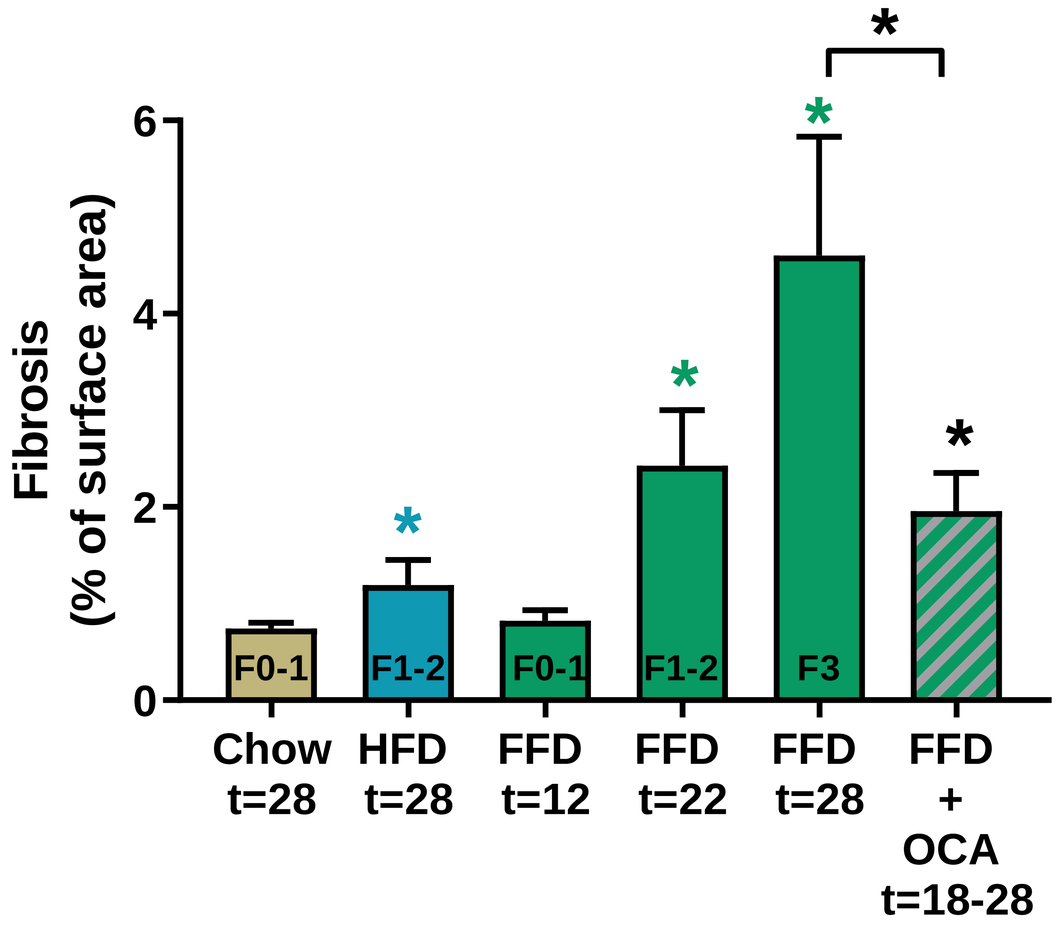
Ldlr-/-.Leiden mice on high fat diets develop NASH with progressive fibrosis

At t=28 weeks:	Chow	HFD	FFD
Body weight (g)	38.3 ± 1.5	52.3 ± 1.1*	48.7 ± 1.5*
Glucose (mM)	7.7 ± 0.4	7.7 ± 0.2	6.5 ± 0.3*
Insulin (ng/mL)	2.9 ± 0.6	14.7 ± 4.2*	3.9 ± 0.4*
ALT pooled/group (U/L)	46.6	290.0	364.0
Macrovesicular steatosis (% of liver surface area)	3.5 ± 2.4	30.1 ± 1.3*	40.9 ± 3.9*
Microvesicular steatosis (% of liver surface area)	5.5 ± 3.7	32.5 ± 1.9*	28.6 ± 2.8*
Hepatic inflammation (# of infl. foci/mm²)	0.4 ± 0.1	7.9 ± 3.9*	15.0 ± 4.2*
eWAT inflammation (# of CLS/mm²)	0.2 ± 0.2	0.8 ± 0.2*	0.5 ± 0.1*

Values are means ± SEM | * p<0.05 *vs.* chow

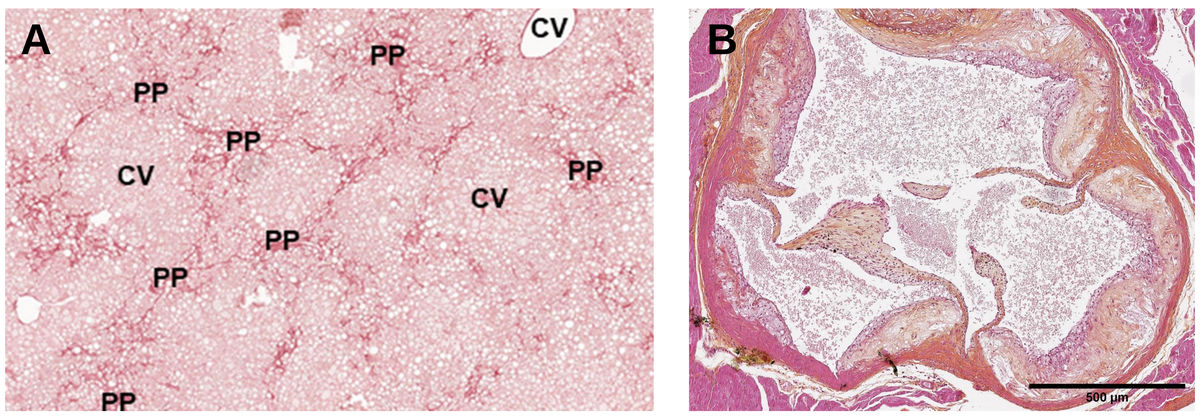
Both high fat diets induce obesity, hyperinsulinemia, increased ALT levels, steatosis and inflammation in liver and WAT.

Values are means ± SEM | * p<0.05 *vs.* chow



Both high fat diets induce hepatic fibrosis but FFD has stronger effects.
While insulin resistance and adipose tissue inflammation is more pronounced with HFD, hepatic inflammation and fibrosis is more pronounced with FFD.
OCA treatment (from 18 to 28 weeks) significantly reduces fibrosis.

Ldlr-/-.Leiden mice on high fat diets develop hepatic fibrosis with simultaneous atherosclerosis development



A. Ldlr-/-.Leiden mice develop bridging fibrosis, all mice show F3 stage after 28 weeks on FFD. CV: central vein; PP: periportal area.
B. Ldlr-/-.Leiden mice develop severe atherosclerotic lesions (type IV & V) in the aortic root area after 22 weeks on FFD.

CONCLUSIONS

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 NASH/fibrosis in association with atherosclerosis that can be used to investigate the effects of new drugs or drug combinations.

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