A novel small molecule modulating the mitochondrial NEET proteins improves inflammation and fibrosis in kidneys of NASH mice

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INTRODUCTION

Non-alcoholic steatohepatitis (NASH) is a disease characterized by excessive fat accumulation, inflammation, and ballooning degeneration of hepatocytes, with or without fibrosis in the liver. It is now reported that NASH not only affects the liver but is also associated with chronic kidney disease (CKD). However, the morphological appearance of NASH kidneys has been poorly characterized. These observations highlight the need for a treatment that targets both conditions. Here, we assessed the effect of a novel chemistry that regulates the function of 3 mitochondrial proteins called the NEET proteins, previously reported to be important in metabolic diseases, on a diet-induced NASH model in mice.

OBJECTIVES

Characterize a model of NASH-induced CKD and study the impact of dEF3122, a novel small molecule modulating the mitochondrial NEET proteins, on kidney lesions in a mouse model of NASH.

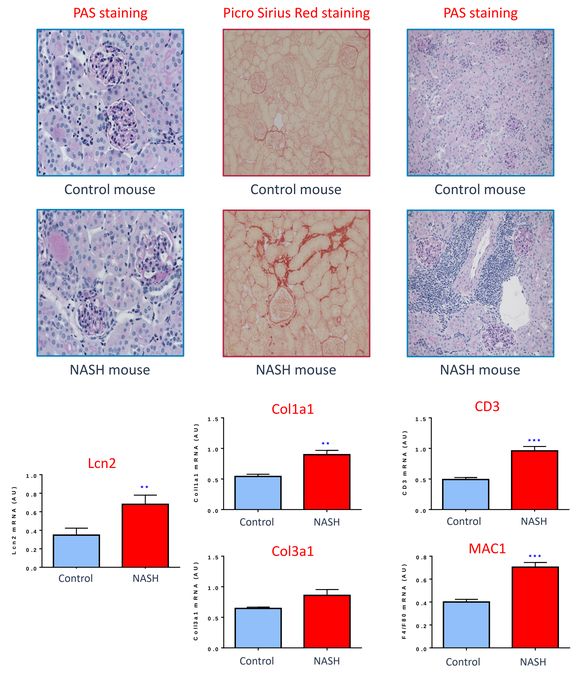
METHODS

Five-week-old male C57BL/6JRj mice were fed either chow diet or AMLN diet (40% total fat kcal of which 18.5% were trans-fat kcal, 20% fructose, 2% cholesterol). This diet was maintained for 30 weeks prior to study initiation. Three weeks prior to study initiation, liver biopsies were taken and animals with steatosis grade <2 and fibrosis stage <1 were deselected from the study. Prior to first administration of test article, stratified randomization of mice into treatment groups was performed according to collagen 1a1 (IHC) morphometry from the week -3 biopsies. Each treatment group consistent of 12 mice. For a total of 8 weeks, compound dEF3122 was administered orally, twice daily at a concentration of 7mg/kg (low dose, EFL) or 20mg/kg (high dose, EFH). At the end of the treatment animals were sacrificed, the livers and kidneys were collected, and sections were stained with H&E, PAS and picrosirius red (PSR) to analyze their morphology. Histopathological analysis was performed by a pathologist blinded to the study. Infiltration was determined using anti-CD3 and anti-F4/F80 antibodies. Quantification was performed using Image J. Lesion markers were analyzed by quantitative RT-PCR.

RESULTS

NASH induces renal lesions

Figure 1: NASH mice develop renal lesions



NASH mice presented severe renal lesions such as glomerulosclerosis, tubular casts and lipid accumulation, and interstitial fibrosis and mononuclear cell infiltration.

NEET Modulator improves renal lesions in NASH mice

Figure 2: Renal fibrosis

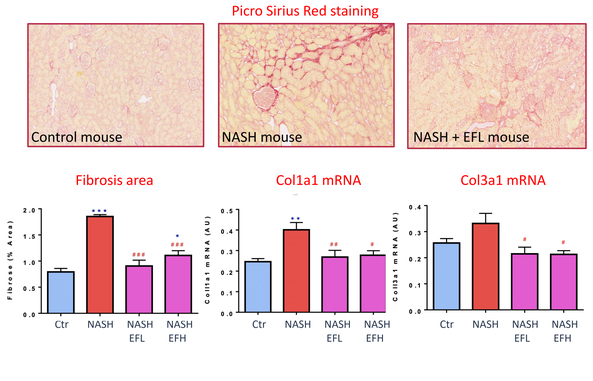


Figure 3: Renal lymphocyte infiltration

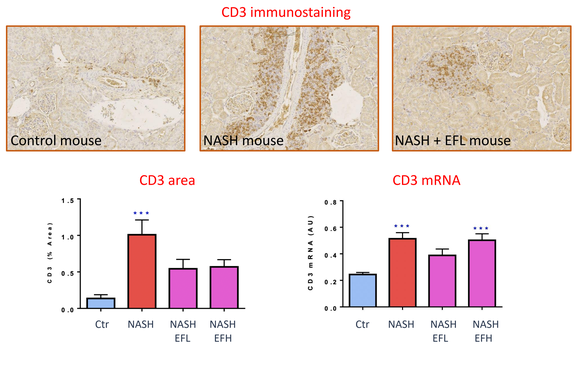
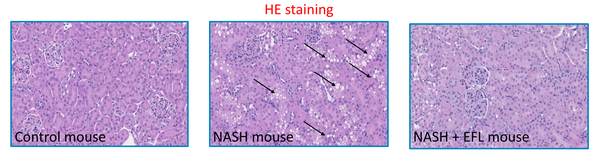
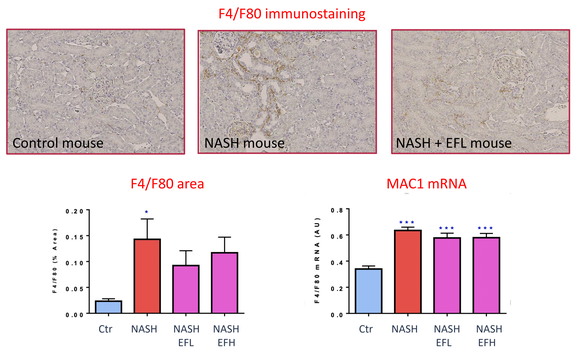


Figure 5: Tubular lipid accumulation



dEF3122, a modulator of mitochondrial NEET proteins, significantly improved periportal liver fibrosis and inflammation (data not shown). More importantly, the same compound protected also kidneys from NASH-induced renal lesions. In particular, fibrosis and mononuclear cell infiltration were decreased, although the difference reached the significance for fibrosis, exclusively. Tubular lipid accumulation (arrows in HE staining) was also reduced by dEF3122 administration.

Figure 4: Renal macrophage infiltration



CONCLUSIONS

We have shown that NASH mice develop renal lesions, recapitulating the phenotype observed in humans. More importantly, we have identified a novel treatment that, by regulating the activity of a family of proteins of critical importance in mitochondrial biology (NEET proteins), protects mice from the development of both liver and kidney lesions. This could offer new perspectives in the treatment of NASH and other diseases with an important fibrotic/inflammatory component.

DISCLOSURES

JV : Stock Shareholder at Obseva SA and Hoffman La Roche AG, Board member at Obseva SA, Inatherys SAS and Step Pharma SAS.

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