THE COMPARATIVE EFFECT OF LIRAGlutide, elAFBRANOR AND obETICHOLIC ACID ON NAFLD ACTIVITY Score AND FIBROSIS STAGE IN A DIET-INDUCED OBESE MOUSE MODEL OF BIOPSY-CONFIRMED NON-ALCOHOLIC STEATOHEPATITIS

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Background: The aim of the study was to compare metabolic, histopathological and molecular effects of liraglutide (GLP-1 receptor agonist), elafibranor (peroxisome proliferator activated receptor (PPAR) α/δ agonist) and obeticholic acid (OCA, farnesoid x receptor agonist) in the liver biopsy-confirmed Gubra-diet-induced obese (DIO) model of nonalcoholic steatohepatitis (DIO-NASH).

Methods: Male C57Bl/6J mice (5 weeks of age) were offered ad libitum access to AMLN diet (40% trans-fat, 22% fructose and 2% cholesterol) for a total of 26 weeks to induce DIO-NASH. Thereafter, a liver pre-biopsy was obtained. Only biopsy-confirmed steatotic and fibrotic animals (steatosis score ≥2; Fibrosis Stage ≥1) were included and stratified into treatment groups: Vehicle (PO, QD), liraglutide (0.2 mg/kg, SC, BID), elafibranor (30 mg/kg, PO, QD), OCA (30 mg/kg, PO, QD). Lean controls receiving vehicle (PO, QD) were included. Animals were treated for 8 weeks. At termination, blood samples were collected for plasma liver enzymes (alanine/aspartate aminotransferases; ALT/AST) and lipids (total cholesterol; TC, triglycerides; TG). Furthermore, liver post-biopsies and tissue samples were obtained for histological, biochemical and RNA sequencing analysis. Primary endpoints included a blinded histological assessment of NAFLD Activity Score (NAS) (steatosis, inflammation, ballooning degeneration) including fibrosis stage.

Results: Liraglutide and elafibranor induced a weight loss of approximately 10%, whereas OCA treatment did not influence body weight. Notably, while liraglutide and OCA reduced liver weight by 37% and 26%, respectively, elafibranor increased liver size. Liraglutide treatment only improved plasma levels of ALT/AST. In addition, liraglutide and OCA reduced plasma TC. All treatments improved terminal hepatosteatosis by reducing liver lipid content. NAS was reduced by OCA (8/10 animals), liraglutide (7/10 animals) and elafibranor (10/10 animals), mainly by reducing steatosis and inflammation components. Only elafibranor significantly reduced liver fibrosis stage (6/10 animals). Transcriptome analysis revealed distinct global gene expression patterns for the different compounds, however, NASH response genes showed very similar effect of all compounds. For all compounds, reduced expression of a number of fibrosis genes showed marked decreases. Furthermore, a wide panel of genes...
related to macrophage recruitment and inflammation, such as CCR2 and MCP-1, were down-regulated by the compounds.

**Conclusion:** Pharmacological intervention with OCA, liraglutide and elafibranor showed different pharmacodynamics in the Gubra DIO-NASH mouse. However, all treatments led to lowered steatosis and improved NASH-associated liver histopathology. Global gene expression analysis supported the similar effect of the three compounds on key NASH endpoints. Notably, only elafibranor improved liver fibrosis stage. Hence, the Gubra DIO-NASH mouse model demonstrating key hallmarks of metabolic NASH and with individual assessment of pre-post biopsy confirmed disease regression is applicable for preclinical pharmacological testing.

**Disclosure:**

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