Metabolic, biochemical, histopathological, and transcriptomic effects of semaglutide and lanifibranor treatment in the GAN diet-induced obese and biopsy-confirmed mouse model of NASH

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Background & Aim

Semaglutide (GLP-1 analogue) and lanifibranor (pan-PPAR agonist) have recently been reported to promote NASH resolution (semaglutide, lanifibranor) and improvement in fibrosis stage (lanifibranor) in late-stage clinical trials for NASH. The present study aimed to evaluate the metabolic, biochemical, histopathological and transcriptomic effects of semaglutide and lanifibranor treatment in the GAN (Gubra-Amylin NASH) diet-induced obese (DIO) mouse model of fibrosing NASH.

Study outline

Group #	Animal	Gender	Strain	Number of animals	Treatment	Administration route	Dosing Frequency	Dosing volume	Dosing concentration
1	LEAN-CHOW	Male	C57BL/6JRj	10	Vehicle	SC/PO	QD	5 ml/kg	-
2	DIO-NASH	Male	C57BL/6JRj	16	Vehicle	SC	QD	5 ml/kg	-
3	DIO-NASH	Male	C57BL/6JRj	14	Semaglutide	SC	QD	5 ml/kg	30 nmol/kg
4	DIO-NASH	Male	C57BL/6JRj	16	Vehicle	PO	QD	5 ml/kg	-
5	DIO-NASH	Male	C57BL/6JRj	14	Lanifibranor	PO	QD	5 ml/kg	30 mg/kg

Improvement in metabolic and biochemical parameters

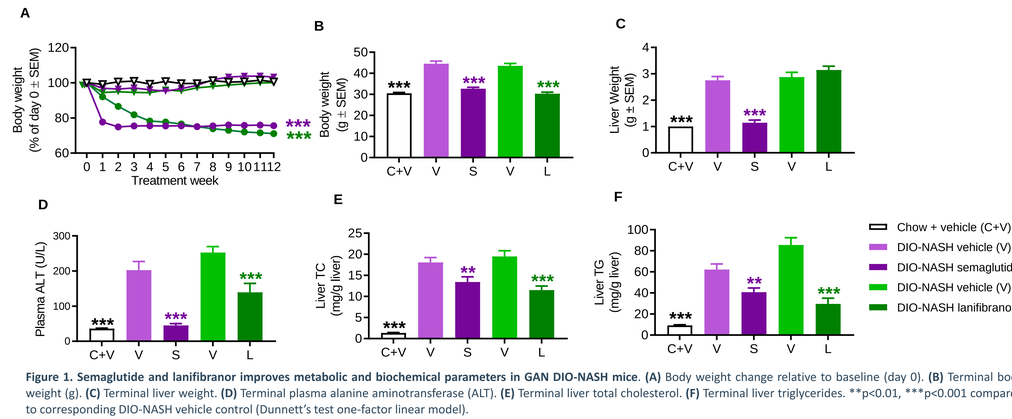
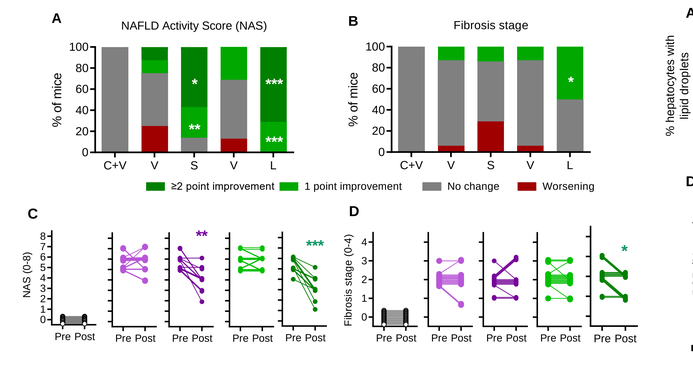


Figure 1. Semaglutide and lanifibranor improves metabolic and biochemical parameters in GAN DIO-NASH mice. **(A)** Body weight change relative to baseline (day 0). **(B)** Terminal body weight (g). **(C)** Terminal liver weight. **(D)** Terminal plasma alanine aminotransferase (ALT). **(E)** Terminal liver total cholesterol. **(F)** Terminal liver triglycerides. **p<0.01, ***p<0.001 compared to corresponding DIO-NASH vehicle control (Dunnett's test one-factor linear model).

Improvement in NAFLD Activity Score and Fibrosis Stage



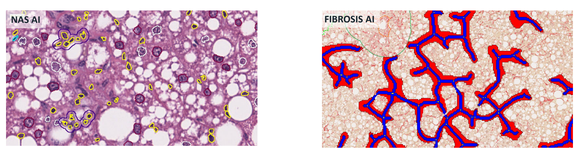
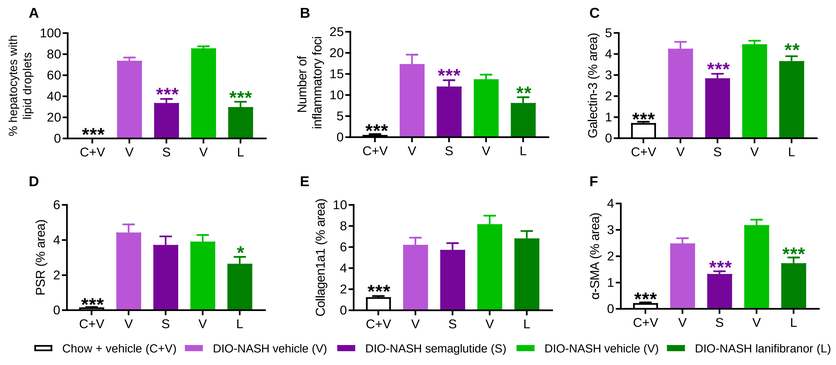


Figure 2. Semaglutide and lanifibranor improves liver histopathological scores in GAN DIO-NASH mice. Histopathological scores were determined by Gubra Histopathological Objective Scoring Technique (GHOST) deep learning-based image analysis. **(A)** NAFLD Activity Score (NAS). **(B)** Fibrosis stage. **(C)** Comparison of individual pre-post NAS. **(D)** Comparison of individual pre-post Fibrosis stage. *p<0.05, **p<0.01, ***p<0.001 to corresponding DIO-NASH vehicle group (One-sided Fisher's exact test with Bonferroni correction). Bottom panels: Representative HE and PSR photomicrographs used for GHOST evaluation.

Improvement in quantitative histology of steatosis, inflammation and fibrosis



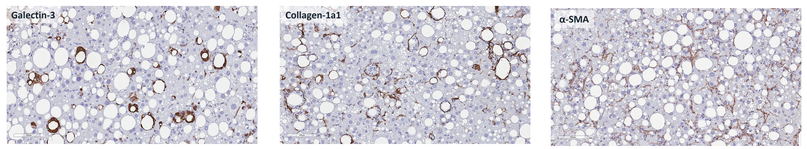


Figure 3. Semaglutide and lanifibranor improves quantitative liver histological markers in GAN DIO-NASH mice. Histomorphometric assessments were performed by GHOST deep learning-based image analysis on scoring-associated variables (panels A-B) and conventional IHC image analysis (panels C-F). **(A)** % hepatocytes with lipid droplets. **(B)** Number of inflammatory foci. **(C)** % area of galectin-3. **(D)** % area of PSR. **(E)** % area of collagen-1a1. **(F)** % area of alpha-smooth muscle actin (α-SMA). Mean ± SEM. **p<0.01, ***p<0.001 to corresponding DIO-NASH vehicle group (Dunnett's test one-factor linear model). Bottom panels: Representative galectin-3, collagen 1a1 and α-SMA photomicrographs (scale bar, 100 µm).

Improvement in transcriptomic profile for fibrosis

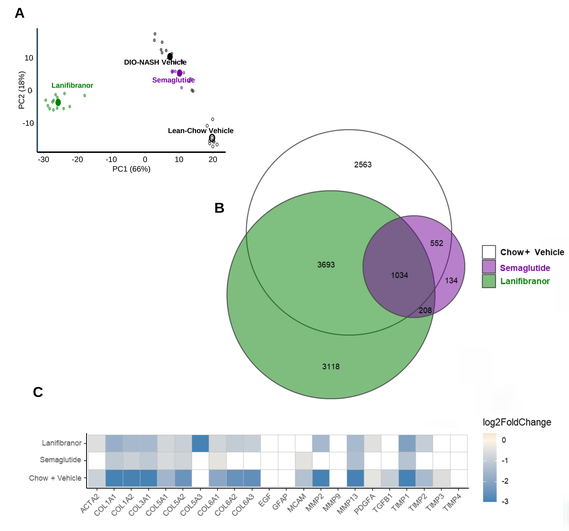


Figure 4. Semaglutide and lanifibranor suppress fibrosis-associated genes in GAN DIO-NASH mice. **(A)** Principal component analysis (PCA) of samples based on top 500 most variable gene expression levels. **(B)** Venn diagram depicting shared and separate differentially expressed genes in treatment groups. **(C)** Regulation of hepatic extracellular matrix (ECM) candidate genes. Blue colour gradients indicate significantly (p<0.05) down-regulated gene expression compared to DIO-NASH vehicle mice. White boxes indicate genes not regulated (p>0.05) compared to DIO-NASH vehicle mice.

CONCLUSION

- Semaglutide and lanifibranor reduce body weight, plasma liver ALT and liver lipids.
- Semaglutide and lanifibranor demonstrate ≥2-point significant improvement in NAFLD Activity Score.
- Semaglutide and lanifibranor reduce quantitative histological markers of steatosis, inflammation and stellate cell activation.
- Lanifibranor demonstrate 1-point significant improvement in Fibrosis Stage.
- Lanifibranor reduce quantitative histological marker of fibrosis.
- Semaglutide and lanifibranor demonstrate transcriptomic effects on fibrosis-associated genes.
- These findings are in agreement with clinical findings, further highlighting clinical translatability of the GAN DIO-NASH mouse model.