GS011

Microbial produced ethanol: an underestimated burden on the liver

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Background and aims: Endogenously produced ethanol has been suggested to play a significant role in the development of Non-Alcoholic Fatty Liver Disease (NAFLD). However, its role in human disease has never been demonstrated unequivocally. Here, we report on microbial endogenous ethanol production in individuals with and without NAFLD using four experiments in which we sampled portal vein blood, stimulated endogenous ethanol production, inhibited alcohol dehydrogenase (ADH) and eradicated ethanol producing bacteria via supplementation of broad-spectrum antibiotics.
Method: We designed and performed one prospective clinical study and one intervention study. In the first study, we enrolled 146 individuals from our bariatric surgery (BARIA cohort) and measured ethanol in fasting and 120 minutes after a mixed meal test (MMT). To study the burden of endogenously produced ethanol, we calculated difference between portal and peripheral plasma ethanol levels in a subset of individuals and validated in an external cohort. In the intervention study, 10 individuals with NAFLD and 10 healthy controls were infused with the selective ADH inhibitor 4-methylpyrazole and then underwent an MMT. In the NAFLD group, the MMT with 4-methylpryazol infusion was repeated after a week of supplementation with oral broad-spectrum antibiotics. Fecal shotgun metagenomics was performed in both studies.
Results: Endogenously produced ethanol was present in fasting samples at baseline with a profound increase observed 120 minutes after the MMT. Also, positive correlations were found between post prandial plasma ethanol and several (small intestinal) species belonging to the Lactic Acid Bacteria (LAB).
Portal vein ethanol concentrations increased in a dose dependent manner reaching mean levels in NAFL and NASH groups over 30 mmol/L. In the validation cohort, similar levels were observed. Portal vein ethanol positively correlated with LAB in the small intestine. Finally, in the intervention study, endogenously produced ethanol increased in all participants during the MMT, yet was significantly higher in the NAFLD group. Subsequently, depletion of intestinal microbiota following broad spectrum antibiotics resulted in near complete suppression of detectable ethanol during the MMT with 4-methylpyrazole, underscoring potential causality.
Conclusion: The human gut microbiome can produce large amounts of ethanol, which are clinically relevant in the pathogenesis of NAFLD. LAB abundance correlated significantly with high plasma ethanol concentrations in fasting portal vein blood and in postprandial venous blood.

GS012

A non-calorie restricted low carbohydrate high fat diet improves non-alcoholic fatty liver disease (NAFLD) activity score (NAS) and HbA1c in type 2 diabetes: a six-month randomised controlled trial

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Background and aims: NAFLD affects 55% of people with type 2 diabetes mellitus (T2DM), and glycaemic control predicts the severity of ballooning and fibrosis in NAFLD. Dietary interventions with low carbohydrates improve glycaemic control but the effect on NAFLD activity is unknown. We aimed to investigate the effect of a six-month low-carbohydrate high fat (LCHF) diet on NAFLD assessed by ≥2 points improvement in the NAFLD Activity Score (NAS) with at least 1 point improvement in either lobular inflammation or ballooning without worsening of fibrosis and on glycaemic control.
Method: We conducted a six-month randomised controlled diet trial in 185 people with T2DM. Participants were randomised 2:1 to LCHF or to a diet consisting of high carbohydrates and low fat (HCLF). Both diets were non-calorie-restricted. The LCHF diet consisted of maximum 20 energy percent (E%) carbohydrates, 50–60E% fats and 25–30E% proteins. The HCLF diet consisted of 50–60E% carbohydrates, 20–30E% fats and 20–25E% proteins. We performed liver biopsies and measured HbA1c (mmol/mol) at baseline and after six months. Biopsies were scored in a blinded matter according to the Non-alcoholic Steatohepatitis Clinical Research Network. The participants had ongoing dietitian consultations and compliance was reported continuously through an online food diary platform.
Results: Out of 185 randomised participants, 165 commenced the allocated intervention and were included in the analysis. At baseline the mean age was 56 (SD, 10) years, 58% were female, 88% had NAFLD, median NAS was 3 (1–5) and mean HbA1c was 56 (SD, 10) mmol/mol. After intervention we saw no significant difference between the groups in relation to improvement of ≥2 points in NAS ($p = 0.587$). However, more participants in the LCHF group improved NAS with ≥1 point compared to the HCLF group (70% versus 49%; $P = 0.028$), and fewer in the LCHF group experienced a worsening of NAS compared to the HCLF group (1% versus 23%; $P < 0.001$) (Figure). Participants in the LCHF group improved HbA1c with −9.5 versus −3.4 in the HCLF group ($p < 0.001$) and lost significantly more weight than in the HCLF group (−5.7 kg vs. −1.8 kg; $P < 0.001$). The self-reported macronutrient intake in LCHF versus HCLF throughout the intervention was 13/46E% carbohydrates, 61/29E% fats and 23/21E%.

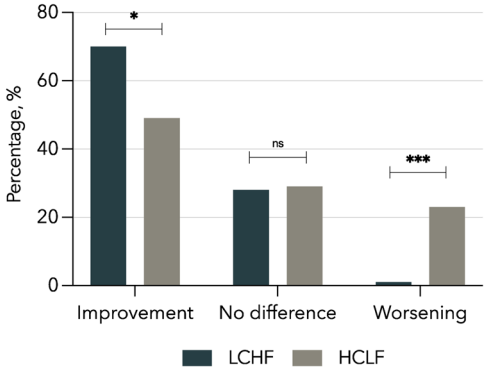

Figure: Change in NAS.

Conclusion: A six-month non-calorie-restricted LCHF diet improves NAS and HbA1c significantly more than a HCLF diet in people with T2DM.

Saturday 25 June

Late-Breaker

LB001

Semaglutide 2.4 mg once weekly improved liver and metabolic parameters, and was well tolerated, in patients with non-alcoholic steatohepatitis-related cirrhosis: a randomised, placebo-controlled phase 2 trial

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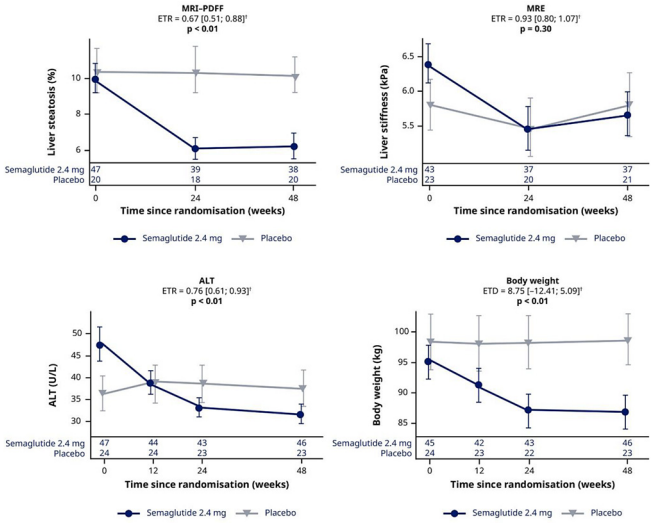
Background and aims: Patients with non-alcoholic steatohepatitis (NASH) and compensated cirrhosis (CC) are at high risk of liver-related and all-cause morbidity and mortality. In a previous placebo-controlled trial in patients with NASH without cirrhosis (NCT02970942), the glucagon-like peptide-1 receptor agonist semaglutide improved NASH resolution and associated metabolic parameters, and was well tolerated. We investigated the efficacy and safety of semaglutide in patients with NASH and CC.
Method: In a phase 2, randomised, placebo-controlled trial, adults with biopsy-confirmed NASH and CC, body mass index (BMI) ≥27 kg/m^2 and glycated haemoglobin (HbA_{1c}) ≤9.5% were randomised 2:1 to semaglutide 2.4 mg or placebo once weekly for 48 weeks. Histological parameters were assessed at baseline and week 48 by a single pathologist. The primary end point was the proportion of patients with ≥1 stage of liver fibrosis improvement without worsening of NASH after 48 weeks. Supportive secondary end points included NASH resolution, changes in: alanine- and aspartate-aminotransferase (ALT, AST); liver stiffness assessed by magnetic resonance elastography (MRE); liver fat content measured by magnetic resonance imaging proton density fat fraction (MRI-PDFF); cardiometabolic parameters; and exploratory biomarkers, and adverse events (AEs).
Results: Patients (N = 71) were mainly female (69%), with a mean age of 59.5 ± 8.0 years, BMI 34.9 ± 5.9 kg/m^2 and non-alcoholic fatty liver disease activity score 4.8 ± 1.0; 75% had type 2 diabetes. Baseline parameters were balanced between treatments. There was no difference between semaglutide and placebo for the primary end point (10.6% vs 29.2%; p = 0.09) or NASH resolution (34.0% vs 20.8%; p = 0.29). Compared with placebo, semaglutide reduced ALT (Figure), AST (estimated treatment ratio [ETR] 0.77; p < 0.01), MRI-PDFF but not MRE score (Figure), and pro-C3 levels (ETR 0.84; p = 0.03), and led to reductions in HbA_{1c} (estimated treatment difference −1.63; p < 0.01 for patients with diabetes) and body weight (Figure). Semaglutide also reduced triglycerides (ETR 0.83; p = 0.01) and very low-density lipoprotein cholesterol (ETR 0.83; p = 0.01). Similar proportions of semaglutide and placebo recipients reported AEs during the trial (89.4% vs 79.2%, of which 12.8% and 8.3%, respectively, were serious). Gastrointestinal AEs occurred in 76.6% of semaglutide recipients (vs 33.3% with placebo). Hepatic and renal function remained stable. There were no decompensating events or deaths.
Conclusion: Although the primary end point was not met, semaglutide 2.4 mg once weekly appeared safe, was well tolerated and improved cardiometabolic parameters and non-invasive markers of liver injury associated with fibrosis progression. Larger trials are needed to investigate whether semaglutide improves liver-related morbidity and mortality in patients with NASH and CC.



Number of observations per treatment group and visit is presented in the lower part of each plot. Error bars show the standard of the mean for observed values and 95% confidence limits for estimated means. †ETD/ETRs with 95% confidence intervals and two-sided p-values are from the same analysis. Missing data were inputed from the observed data in the placebo group using the same analysis of covariance model but without treatment as factor.

ETD = Estimated difference of absolute change from baseline between semaglutide and placebo. ETR = Estimated ratio of relative change from baseline between semaglutide and placebo.

ALT, alanine aminotransferase; ETD, estimated treatment difference; ETR, estimated treatment ratio; MRE, magnetic resonance elastography; MRI-PDFF, magnetic resonance imaging proton density fat fraction.