Diabetic and nondiabetic patients with nonalcoholic fatty liver disease have an impaired incretin effect and fasting hyperglucagonaemia

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Objective. We evaluated whether patients with histologically verified nonalcoholic fatty liver disease (NAFLD) have an impaired incretin effect and hyperglucagonaemia.

Methods. Four groups matched for age, sex and body mass index were studied: (i) 10 patients with normal glucose tolerance and NAFLD; (ii) 10 patients with type 2 diabetes and NAFLD; (iii) eight patients with type 2 diabetes and no liver disease; and (iv) 10 controls. All participants underwent a 50-g oral glucose tolerance test (OGTT) and an isoglycaemic intravenous glucose infusion (IIGI). We determined the incretin effect by relating the beta cell secretory responses during the OGTT and IIGI. Data are presented as medians (interquartile range), and the groups were compared by using the Kruskal–Wallis test.

Results. Controls exhibited a higher incretin effect [55% (43–73%)] compared with the remaining three groups (P < 0.001): 39% (44–71%) in the nondiabetic NAFLD patients, 20% (–5–50%) in NAFLD patients with type 2 diabetes, and 2% (–8–6%) in patients with type 2 diabetes and no liver disease. We found fasting hyperglucagonaemia in NAFLD patients with [7.5 pmol L–1 (6.8–15 pmol L–1)] and without diabetes [7.5 pmol L–1 (5.0–8.0 pmol L–1)]. Fasting glucagon levels were lower but similar in patients with type 2 diabetes and no liver disease [4.5 pmol L–1 (3.0–6.0 pmol L–1)] and controls [3.4 pmol L–1 (1.8–6.0 pmol L–1)]. All groups had similar glucagon-like peptide-1 and glucose-dependent insulinotropic polypeptide responses.

Conclusions. Patients with NAFLD have a reduced incretin effect and fasting hyperglucagonaemia, with the latter occurring independently of glucose (in)tolerance.

Keywords: glucagon, glucagon-like peptide-1, glucose-dependent insulinotropic polypeptide, incretin effect, nonalcoholic fatty liver disease, type 2 diabetes.

Introduction

During the last 30 years, the incidence of nonalcoholic fatty liver disease (NAFLD) has reached epidemic proportions [1]. In Western countries, NAFLD is the most common liver disorder [2] and the third most common reason for liver transplantation [3]. The condition is defined by an intrahepatic fat content exceeding 5% without significant alcohol intake or use of steatogenic medication [4]. The spectrum of NAFLD ranges from simple steatosis to nonalcoholic steatohepatitis (NASH), which may lead to fibrosis and cirrhosis [5]. NAFLD is associated with obesity, insulin resistance and type 2 diabetes, which are features of the metabolic syndrome [6]. Up to 70% of obese patients with type 2 diabetes have NAFLD [7–9].

Glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic polypeptide (GIP) are incretin hormones released from endocrine cells in the gastrointestinal tract after food ingestion. These
hormones amplify insulin secretion after oral glucose [10], a phenomenon known as the incretin effect, which accounts for up to 70% of the insulin response following oral intake of glucose in healthy subjects [11]. Patients with type 2 diabetes exhibit an impaired incretin effect that contributes to their glucose intolerance [12]. Previous studies have found that some obese persons exhibit an impaired incretin effect despite normal glucose tolerance [13, 14], and evidence also exists for NAFLD being associated with impaired GLP-1 and excessive GIP responses [15, 16]. These findings suggest that NAFLD may be associated with an impaired incretin effect despite normal or increased secretion of the incretin hormones [16, 17].

Glucagon is a pancreatic hormone that helps to increase blood glucose levels. Hypoglycaemia and protein intake are the major secretory stimuli for glucagon secretion, which in turn stimulates hepatic glucose production [18]. In addition to insufficient pancreatic beta cell secretion and insulin resistance, the pathophysiology of type 2 diabetes involves elevated fasting and postprandial glucagon levels [14, 19–21]. High levels of fasting glucagon have also been found in nondiabetic patients with NAFLD. However, the investigators used a lean control group, which may have influenced their results [16].

We hypothesized that nondiabetic obese NAFLD patients would have a reduced incretin effect and fasting hyperglucagonaemia. We therefore investigated the influence of NAFLD on the incretin effect and secretion of glucagon in patients with either normal glucose tolerance or type 2 diabetes.

Materials and methods
The study protocol was approved by the scientific-ethical committee of the Capital region of Denmark (H-1-2011-082) and registered with the Danish Data Protection Agency (2011-61-6410) and ClinicalTrials.gov (reg. no. NCT01492283). We conducted the study according to the principles of the Declaration of Helsinki and obtained oral and written informed consent from all participants.

Patients
We included patients from the outpatient clinic at the Department of Medicine, Gentofte Hospital, University of Copenhagen, Hellerup, Denmark, from September 2011 to May 2013. We included patients with normal glucose tolerance and NAFLD, type 2 diabetes and NAFLD, and type 2 diabetes and no liver disease. We used controls from another study by our group [22] that matched with regard to age, sex and body mass index. NAFLD was diagnosed based on histology and graded according to the quantity of hepatic fat infiltration: no NAFLD (<5% fat infiltration), mild (5–33% fat infiltration), moderate (33–66% fat infiltration) and severe (>66% fat infiltration). NASH and fibrosis were graded according to the NAFLD activity score [23]. Type 2 diabetes was diagnosed according to World Health Organization criteria [24]. Normal glucose tolerance was defined as fasting plasma glucose <6.1 mmol L\(^{-1}\) and plasma glucose at 120 min <7.8 mmol L\(^{-1}\) based on a 2-h 75-g oral glucose tolerance test (OGTT). Our exclusion criteria were weekly alcohol consumption of more than seven units for women and 14 for men, treatment with steatogenic drugs within 3 months prior to inclusion, anaemia, inflammatory bowel disease, gut resection (except appendectomy), increased creatinine (>150 \(\mu\)mol L\(^{-1}\)), albuminuria or other chronic diseases. Controls were healthy and had no family history of diabetes, signs of liver disease (based on patient history, biochemical measurements and ultrasound assessment), or other chronic diseases.

Experimental design and biochemical procedures
The inclusion and exclusion criteria were evaluated at a screening visit. On day 1, the subjects ingested 50 g of water-free glucose dissolved in 300 mL of water. Arterialized blood was drawn regularly starting 15 min before ingestion of glucose until 240 min afterwards. On day 2, cannulas were inserted into both antecubital veins. One cannula was used for blood samples and the other for glucose infusion. Sterile 20% (w/v) glucose was infused at a variable rate intended to duplicate the plasma glucose profile from the OGTT [25]. Acetaminophen (Panodil; GlaxoSmithKline A/S, Copenhagen, Denmark) was added to the OGTT to evaluate gastric emptying. Plasma glucose concentrations were measured using Yellow Springs Instrument (YSI) 2300 STAT plus glucose analyser (YSI Inc., Yellow Springs, OH, USA). Concentrations of insulin, C-peptide, GLP-1, GIP and glucagon were measured as previously described [26–29]. Plasma acetaminophen was measured using a routine enzymatic colorimetric assay on the Vitros 5.1. FS analyser (Ortho-Clinical Diagnostics, Johnson & Johnson, Birkerød, Denmark) [30].
Outcome measures

The primary outcome measures were the incretin effect [based on insulin and C-peptide area under the curve (AUC) values] and fasting glucagon. Secondary outcome measures included gastrointestinal-mediated glucose disposal (see following section), GLP-1, GIP and glucagon responses to OGTT compared with isoglycaemic intravenous glucose infusion (IIGI), and gastric emptying during the OGTT.

Calculations and statistical analysis

Fasting levels of glucose, insulin, C-peptide, glucagon, GLP-1 and GIP were summarized using the median of values from −15, −10 and 0 min during OGTT and IIGI. AUC was calculated using the trapezoidal rule, and incremental AUCs were used unless otherwise stated. The incretin effect was calculated based on the AUC for insulin and C-peptide, using the following formula: incretin effect (%) = 100 × (AUCOGTT − AUCIIGI)/AUCOGTT. The time-to-peak and AUC values of paracetamol excursions were used as measures of gastric emptying [31]. Gastrointestinal-mediated glucose disposal (%) was calculated as 100 × (amount of glucose given during OGTT − amount of glucose given during IIGI)/amount of glucose given during OGTT [25]. The homeostatic model assessment for insulin resistance [32] was calculated as a measure of the hepatic insulin resistance. The Matsuda index was used to assess whole body insulin sensitivity [33].

The statistical analyses were performed using the Stata version 14 (Stata Corp, College Station, TX, USA). Data were summarized as medians (interquartile range). Comparisons within groups (measurements on different days) were made using the Wilcoxon test. Comparisons between groups were made with the Kruskal–Wallis non-parametric analysis of variance. We also conducted univariate and multivariate linear regression analyses to evaluate predictors of fasting glucagon. We made the calculations with and without log-transformation. The log-transformation had no influence on the overall conclusion in any of the analyses, and we therefore only report the result of the log-transformed analyses. The predictors included alanine aminotransferase levels, NAFLD activity score and haemoglobin A1c values. The multivariate analysis included all three predictors.

Results

Participant characteristic

We included 38 participants (Table 1). Group 1 included 10 patients with normal glucose tolerance and NAFLD. Group 2 included 10 patients with type 2 diabetes and NAFLD. Group 3 included eight patients with type 2 diabetes and no liver disease. Group 4 included 10 controls. The degree of steatosis in liver biopsies from participants with NAFLD was mild (n = 6), moderate (n = 9) or severe (n = 3). Four patients had NASH and four had fibrosis.

Plasma glucose and gastrointestinal-mediated glucose disposal

As expected, patients with normal glucose tolerance and controls had the lowest fasting plasma glucose (Fig. 1c,d). All groups achieved isoglycaemia during the two experimental days (Fig. 1a–d). During the IIGI, patients with type 2 diabetes needed the largest amount of glucose to achieve isoglycaemia compared to controls. The amount of glucose needed to achieve isoglycaemia was only slightly elevated for patients with NAFLD and normal glucose tolerance [Group 1: 34 g (27–43 g); Group 2: 43 g (39–46 g), Group 3: 40 g (36–46 g), Group 4: 24 g (20–30 g); P < 0.001]. The gastrointestinally mediated glucose disposal was impaired in all patient groups compared to controls [Group 1: 32% (14–46%); Group 2: 13% (39–46%), Group 3: 16% (36–46%), Group 4: 52% (20–30%); P < 0.001].

Insulin, C-peptide and incretin effect

Fasting insulin and C-peptide were higher in patients with NAFLD (Group 1 and Group 2) than in patients with type 2 diabetes and no liver disease (Group 3) and in controls (Group 4). Fasting C-peptide was higher in patients with type 2 diabetes and no liver disease than in controls (Fig. 1e–i). Patients with NAFLD and normal glucose tolerance (Group 1) and controls had an earlier and more rapid increase in plasma insulin and C-peptide in response to both oral and intravenous glucose (Fig. 1e,h,i,l) than patients with type 2 diabetes (Group 2 and Group 3) (Fig. 1f,g,j,k). All patients had a reduced incretin effect compared to controls based on both insulin [Group 1: 55% (44–71%); Group 2: 33% (19–59%), Group 3: 5% (−10% to 13%), Group 4: 70% (55–85%); P < 0.001] and C-peptide [Group 1: 39% (30–48%); Group 2: 20% (−5% to 50%), Group 3: 2% (−8% to 6%), Group 4: 55% (43–73%); P < 0.001].
Glucagon, GLP-1 and GIP

We found fasting hyperglucagonaemia in NAFLD patients with [7.5 pmol L\(^{-1}\) (5.0–8.0 pmol L\(^{-1}\)) and without diabetes [7.5 pmol L\(^{-1}\) (6.8–15 pmol L\(^{-1}\))] (Group 1 and Group 2, respectively). Fasting glucagon levels were similarly low in patients with type 2 diabetes and no liver disease [4.5 pmol L\(^{-1}\) (3.0–6.0 pmol L\(^{-1}\))] and controls [3.4 pmol L\(^{-1}\) (1.8–6.0 pmol L\(^{-1}\))] (Group 2 and Group 4, respectively) (Fig. 2a-d). Patients with normal glucose tolerance and NAFLD exhibited immediate glucagon suppression during the first hour of the OGTT and the IIGI [−83 pmol L\(^{-1}\) (−70 to 16 pmol L\(^{-1}\))] vs. −133 pmol L\(^{-1}\) (−225 to 79 pmol L\(^{-1}\)) × min, \(P = 0.037\) (Fig. 2a). Controls showed similar immediate glucagon suppression during the first hour of the OGTT the IIGI [−38 pmol L\(^{-1}\) (−70 to 10 pmol L\(^{-1}\))] vs. −116 pmol L\(^{-1}\) (−167 to 47 pmol L\(^{-1}\)) × min, \(P = 0.039\) (Fig. 2d). In contrast, patients with type 2 diabetes with or without liver disease had delayed and impaired glucagon suppression during the first hour of the OGTT compared to the IIGI [22 pmol L\(^{-1}\) (−146 to 73 pmol L\(^{-1}\))] vs. −45 pmol L\(^{-1}\) (−116 to 27 pmol L\(^{-1}\)) × min, \(P = 0.027\) and 35 pmol L\(^{-1}\) (−35 to 87 pmol L\(^{-1}\)) vs. −94 pmol L\(^{-1}\) (−115 to 43 pmol L\(^{-1}\)) × min, \(P = 0.039\) (Fig. 2b,c). Impaired glucagon suppression was most pronounced in patients with type 2 diabetes and NAFLD (Fig. 2b). GLP-1 and GIP responses were higher after OGTT than IIGI, with no differences between the groups (Fig. 2e–f).

In the univariate regression analysis, alanine aminotransferase was the only predictor of fasting glucagon (\(P = 0.011\)). The NAFLD activity score (\(P = 0.50\)) and haemoglobin A\(_{1c}\) did not predict

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**Table 1** Patient characteristics

<table>
<thead>
<tr>
<th></th>
<th>NGT and NAFLD Group 1</th>
<th>T2D and NAFLD Group 2</th>
<th>T2D and no liver disease Group 3</th>
<th>Controls Group 4</th>
<th>(P) value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>56 (39–63)</td>
<td>64 (54–65)</td>
<td>59 (50–67)</td>
<td>57 (49–60)</td>
<td>0.47</td>
</tr>
<tr>
<td>Body mass index, kg m(^{-2})</td>
<td>28 (27–34)</td>
<td>30 (28–31)</td>
<td>27 (24–36)</td>
<td>29 (28–29)</td>
<td>0.12</td>
</tr>
<tr>
<td>Waist circumference, cm</td>
<td>103 (97–114)</td>
<td>107 (102–109)</td>
<td>92 (90–106)</td>
<td>104 (99–108)</td>
<td>0.23</td>
</tr>
<tr>
<td>Fasting plasma glucose, mmol L(^{-1})</td>
<td>5.9 (5.3–6.0)</td>
<td>10.1 (6.5–12.4)</td>
<td>7.7 (6.6–8.9)</td>
<td>5.2 (5.0–5.5)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Haemoglobin A(_{1c}) %</td>
<td>5.3 (5.1–5.4)</td>
<td>7.0 (5.9–7.5)</td>
<td>6.2 (5.7–6.8)</td>
<td>5.4 (5.3–5.5)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Haemoglobin A(_{1c}) mmol mol(^{-1})</td>
<td>33 (32–36)</td>
<td>53 (41–60)</td>
<td>44 (42–51)</td>
<td>36 (31–37)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>HOMA(_{IR})</td>
<td>4.6 (3.4–7.7)</td>
<td>8.1 (7.7–11.2)</td>
<td>4.1 (2.5–5.1)</td>
<td>2.6 (1.9–3.2)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Matsuda index</td>
<td>5.5 (3.4–6.1)</td>
<td>4.0 (3.4–4.7)</td>
<td>9.5 (6.5–12.1)</td>
<td>10.1 (7.5–14.5)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Duration of diabetes, months</td>
<td>–</td>
<td>48 (18–78)</td>
<td>52 (11–93)</td>
<td>–</td>
<td>0.92</td>
</tr>
<tr>
<td>Alanine aminotransferase, U L(^{-1})</td>
<td>76 (54–103)</td>
<td>82 (45–108)</td>
<td>29 (24–36)</td>
<td>25 (22–26)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Aspartate aminotransferase, U L(^{-1})</td>
<td>54 (47–75)</td>
<td>59 (38–72)</td>
<td>27 (22–38)</td>
<td>26 (24–28)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Gamma glutamyltransferase, U L(^{-1})</td>
<td>105 (40–236)</td>
<td>93 (67–114)</td>
<td>44 (25–75)</td>
<td>23 (20–38)</td>
<td>0.003</td>
</tr>
<tr>
<td>NAFLD activity score</td>
<td>2 (2–3)</td>
<td>4 (2–4)</td>
<td>–</td>
<td>–</td>
<td>0.04</td>
</tr>
<tr>
<td>Total cholesterol, mmol L(^{-1})</td>
<td>5.6 (4.5–6.7)</td>
<td>4.1 (3.4–4.9)</td>
<td>4.3 (4.1–5.0)</td>
<td>6.3 (5.5–5.7)</td>
<td>0.05</td>
</tr>
<tr>
<td>LDL cholesterol, mmol L(^{-1})</td>
<td>3.5 (2.8–4.3)</td>
<td>2.0 (1.5–3.1)</td>
<td>2.2 (2.0–2.7)</td>
<td>4.2 (3.8–4.6)</td>
<td>&lt;0.003</td>
</tr>
<tr>
<td>HDL cholesterol, mmol L(^{-1})</td>
<td>1.3 (1.1–1.7)</td>
<td>1.2 (1.0–1.6)</td>
<td>1.4 (1.2–1.7)</td>
<td>1.2 (0.9–1.5)</td>
<td>0.71</td>
</tr>
<tr>
<td>Triglycerides, mmol L(^{-1})</td>
<td>1.7 (1.4–2.1)</td>
<td>2.1 (1.5–2.7)</td>
<td>1.2 (0.9–3.0)</td>
<td>1.2 (0.9–1.5)</td>
<td>0.39</td>
</tr>
</tbody>
</table>

Data are expressed as median (interquartile range). \(P\) values are from the Kruskal–Wallis test. HOMA\(_{IR}\), insulin resistance according to the homoeostatic model assessment; NAFLD, nonalcoholic fatty liver disease; NGT, normal glucose tolerance; T2D, type 2 diabetes.
In the multivariate analysis, alanine aminotransferase remained the only significant predictor ($P = 0.013$).

Gastric emptying

Gastric emptying assessed by time-to-peak values of paracetamol excursions during the OGTT was similar between groups ($P = 0.21$). The time-to-peak values were 55 min (48–76 min) in patients with normal glucose tolerance and NAFLD (Group 1), 60 min (40–75 min) in patients with type 2 diabetes and NAFLD (Group 2), 50 min (35–75 min) in patients with type 2 diabetes and no liver disease (Group 3), and 75 min (58–98 min) in controls (Group 4).

Discussion

This study demonstrates that patients with NAFLD have a reduced incretin effect, which seems to be aggravated by type 2 diabetes. The reduced incretin effect does not reflect impaired secretion of GLP-1 and GIP. We also found patients with NAFLD and a normal glucose tolerance and those with NAFLD and type 2 diabetes had fasting hyperglucagonaemia. The patients with normal glucose tolerance and NAFLD had preserved glucagon suppression to both oral and intravenous glucose despite their fasting hyperglucagonaemia.

To ensure the most reliable results, we only included patients with histologically verified NAFLD. The prevalence of NAFLD is high in obese patients with type 2 diabetes. We included patients with normal glucose tolerance and NAFLD, NAFLD and type 2 diabetes, and type 2 diabetes and no liver disease. To avoid problems with different pre-and posthepatic insulin concentrations and to avoid an effect of impaired insulin clearance in patients with NAFLD [34], we used C-peptide profiles to estimate insulin secretion. In theory, impaired or delayed gastric emptying may influence the secretion of incretin hormones [35, 36]. However, we found no difference in gastric emptying between groups, so it is unlikely that this factor influenced our results.

A potential limitation of this study is the small number of participants. We may have overlooked
differences between groups due to the relatively low statistical power. In theory, we may have overlooked low-degree steatosis in controls because we did not perform liver biopsies (for ethical reasons). However, controls were assessed using clinical assessments, ultrasound and blood tests, and clinically significant steatosis is unlikely [37].

Patients with normal glucose tolerance and NAFLD had a higher incretin effect than either group with type 2 diabetes, but lower than controls. Previous studies have found a reduced incretin effect in obese, insulin-resistant patients with normal glucose tolerance [14]. Patients with impaired glucose tolerance also have an impaired incretin effect [13]. These findings suggest that a reduced incretin effect may be an early sign of impaired glucose tolerance associated with insulin resistance and obesity [14, 38]. Our study supports this conclusion by showing that patients with NAFLD and normal glucose tolerance had a reduced incretin effect. This finding suggests that steatosis is associated with an early defect in glucose tolerance and may therefore contribute to the development of type 2 diabetes [39]. We found that the patients with type 2 diabetes and NAFLD had a greater incretin effect and higher insulin and C-peptide responses than patients with type 2 diabetes and no liver disease. The latter may reflect better beta cell function. Beta cell dysfunction has a negative impact on the incretin effect [14, 40, 41] and may therefore explain the difference in the incretin effect between the groups with type 2 diabetes.

We found no differences in secretion of GLP-1 and GIP in any of the groups, which is in line with previous studies in patients with type 2 diabetes [42, 43]. This finding is consistent with patients with type 2 diabetes having reduced beta cell sensitivity to the incretin hormones [12, 20, 24, 38]. Previous studies found blunted GLP-1 secretion in obese patients with NAFLD compared to controls [15, 16]. The discrepancy may partly reflect differences in the selection of the control group and the fact that we included obese individuals as controls. Thus, our findings suggest that
patients with NAFLD and normal glucose tolerance exhibit a form of reduced beta cell sensitivity, which seems similar to patients with type 2 diabetes. Accordingly, the cause of the reduced incretin effect is likely to be due to impaired action of the incretin hormones [12, 19, 20].

In type 2 diabetes, fasting hyperglucagonaemia stimulates hepatic glucose production and thereby contributes to hyperglycaemia [14, 24, 44–46], but some patients with type 2 diabetes do not have fasting hyperglucagonaemia [20, 21, 47]. Fasting hyperglucagonaemia is likely to be an early trait of type 2 diabetes as it also occurs in some obese individuals with or without prediabetes [41, 48]. In this study, patients with NAFLD had fasting hyperglucagonaemia irrespective of type 2 diabetes. This important finding suggests that NAFLD may be involved in the generation of hyperglucagonaemia in type 2 diabetes, which is supported by several animal studies [49, 50]. In mice, hepatic steatosis reduces the number of glucagon receptors in hepatocytes and impairs hepatic glucose production in response to glucagon infusion [49]. Interestingly, glucagon receptor knock-out mice display alpha cell hyperplasia [51]. A study by Longuet et al. [52] indicates that a ‘circulation factor’ produced after disruption of hepatic glucagon signalling causes alpha cell proliferation and hypersecretion of glucagon. Taken together, we hypothesize that hepatic steatosis may downregulate glucagon receptors, which in turn causes the liver to generate more of the circulating factor suggested by Longuet et al. and consequently leads to hyperglucagonaemia. Impaired elimination of glucagon by the steatotic liver could provide an alternative explanation; however, the liver does not seem to play a major role in glucagon elimination [53]. Although the prevalence of NAFLD is high in obese patients with type 2 diabetes, some patients have a normal liver. NAFLD may therefore explain the inconsistent finding of fasting hyperglucagonaemia in patients with type 2 diabetes.

To evaluate all gastrointestinal factors with an effect on glucose disposal, we calculated the gastrointestinal-mediated glucose disposal, which includes the incretin effect. The gastrointestinal-mediated glucose disposal also depends on differences in glucagon secretion during the OGTT and IIGI [24]. All patients had impaired incretin effect. Accordingly, we found impaired gastrointestinal-mediated glucose disposal compared with controls. Interestingly, the two groups with type 2 diabetes had similar gastrointestinal-mediated glucose disposal, but different incretin effects. The higher incretin effect in type 2 diabetes and NAFLD may therefore be counteracted by the more abnormal glucagon response demonstrated in these patients, resulting in similar gastrointestinal-mediated glucose disposal. Thus, our findings suggest that abnormal glucagon responses associated with NAFLD may aggravate glucose intolerance in existing type 2 diabetes. This might also explain why patients with type 2 diabetes and NAFLD are often challenging to treat and need high insulin doses.

In conclusion, we demonstrate that patients with NAFLD despite having normal glucose tolerance are characterized by reduced incretin effect, fasting hyperglucagonaemia and impaired handling of ingested glucose. This study emphasizes the role of NAFLD in metabolic dysregulation and also suggests an important role for the liver in the regulation of glucagon secretion. Future treatments of NAFLD may include strategies to reduce hyperglucagonaemia.

Conflict of interest statement
None.

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Incretin effect and glucagon in NAFLD

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