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Effect of exenatide therapy on hepatic fat quantity and hepatic biomarkers in type 2 diabetic patients

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Dear Editor,

According to the data from the Edinburgh Type 2 Diabetes Study up to 43% of type 2 diabetes mellitus (T2DM) patients have non-alcoholic fatty liver disease (NAFLD) which is associated with greater burden of diabetic complications in T2DM population (1). Liver biopsy, the gold standard for NAFLD diagnosis is not routinely performed because it is an invasive procedure with a certain degree of sampling error (2). Instead, alternative diagnostic methods have been developed and fatty liver index (FLI) as a measure of liver fat content that correlates with abdominal ultrasound and is calculated using an equation including body mass index (BMI), waist circumference, triglycerides (TG) and GGT is one of them (3). Glucagon-like peptide-1 (GLP-1) receptor agonist exenatide is a glucose lowering agent who possesses proven efficacy in achieving and maintaining glycaemic control, weight loss, and improvement in metabolic parameters (4). The decrease in glycated haemoglobin (HbA1c) levels in T2DM patients treated with exenatide was shown to correlate with hepatic fat reduction (5). Recent studies reported the presence of GLP-1 receptor on human hepatocytes and the possible direct exenatide modulation of intrahepatic lipid metabolism and insulin signalling with a consequent improvement in hepatic steatosis (6).

The aim of our study was to evaluate the effect of exenatide therapy, alone or in combination with oral hypoglycaemic agents (OHA), compared to OHA on hepatic fat quantity measured by FLI.

Out of 125 T2DM patients included in the study, 61 (48.8%) were male, mean age was 57±8 years, BMI 38.57±5.35 kg/m2, waist circumference 119.5±14.5 cm, diabetes duration 12±7 years, and HbA1c level 8.57±1.49%. Eighty-seven recieved exenatide therapy: exenatide alone 17 (19.5%), exenatide and metformin 38 (43.7%), exenatide and sulphonylurea 7 (8%), exenatide and metformin and sulphonylurea 25 (28.7%), and 38 metformin or/and sulphonylurea, i.e. metformin 15 (39.5 %), sulphonylurea 2 (5.3%), metformin + sulphonylurea 21 (55.3%) patients. Exenatide was used in dosage of 10 μg twice daily. All subjects were studied in the morning between 08:00 and 09:30 hours after an overnight fast at the study entry and after 6 months. Basic anthropometric measurements were performed on all study subjects. Fasting venous blood samples were collected for the determination of

complete blood count and biochemistry panel, lipid profile status, HbA1c and liver biochemistry. FLI was calculated according to the formula: FLI = ($e^{0.953*\log e (\text{triglycerides}) + 0.139*\text{BMI} + 0.718*\log e (\text{ggt}) + 0.053*\text{waist circumference} - 15.745}$) / ($1 + e^{0.953*\log e (\text{triglycerides}) + 0.139*\text{BMI} + 0.718*\log e (\text{ggt}) + 0.053*\text{waist circumference} - 15.745}$) * 100. Glucose, cholesterol and triglycerides in serum were measured by an enzymatic colorimetric method. Complete blood count was determined on an automatic blood counter (Advia 120, Siemens Diagnostic Solutions, USA). Aspartate aminotransferase (AST), alanine aminotransferase (ALT), γ-glutamyltransferase (GGT) and alkaline phosphatase (ALK) were measured using standard laboratory methods.

The detailed baseline and 6 months after characteristics as well as their changes are given in table 1. The exenatide treated group showed significantly higher change in BMI, waist circumference, ALK, ALT and FLI while lower HbA1c change after 6 months treatment compared to OHA group.

In this open label parallel-group uncontrolled 6 months study we assessed changes in NAFLD markers, FLI, BMI, waist circumference, HbA1c and TG levels in two T2DM groups of patients, one treated with exenatide and the other treated with metformin or/and sulphonylurea. The data from our study suggests that the addition of exenatide to current OHA therapy results in reduction of NAFLD marker levels and intrahepatic fat quantity calculated by FLI. In both exenatide and OHA group a decrease in BMI and HbA1c value was noticed. Similar effect of GLP-1 agonists (exenatide and liraglutide) was observed by Cuthnerson et al. (2012) (5). They reported a significant intrahepatic liver fat and biomarker reduction followed by 6 months GLP-1 agonists therapy in 25 T2DM patients. Data from the study of Klonoffa et al (2008) (7) suggest that exenatide treatment in T2DM patients leads to significant ALT and intrahepatic fat reduction in correlation to insulin resistance decreasment. Sathyanarayana et al (2011) (9) performed an uncontrolled observational study with exenatide and pioglitazone and highlighted the benefit effect of exenatide to intrahepatic fat content independently of BMI reduction. This data clearly suggest that exenatide therapy could reduce or even reverse hepatic fat accumulation. The question arises whether exenatide has direct effect on intrahepatic liver fat reduction or it is mediated by weight loss and improved glucose control.

The reduction in hepatic fat content in exenatide group could be partially explained by reduced caloric intake which is one of the main therapeutic contributions of this drug (4). That is in concordance with decrease in TG levels in the exenatide group and the increase of TG levels in the OHA group. That is also consistent with the BMI and waist circumference reduction which was greater in the exenatide group. However, patient food intake was not controlled, and both groups did have decrease in BMI as well as waist circumference so the effect of reduced caloric intake could be argued.

There are several studies explaining the molecular pleiotropic effect of exenatide on human hepatocytes and the hepatic fat reduction. Although the data from Samson et al (2009) (10) suggest that exenatide might reduce hepatic fat increasing adiponectin mediated activation of AMP-activated protein kinase and mitochondrial fat oxidation enhancement, recent studies suggest that a direct effect of exenatide on hepatic lipogenesis and lipid oxidation cannot be ruled out. GLP-1 treatment in mice resulted in a significant reduction in mRNA expression of stearoyl-CoA desaturase 1 and genes associated with fatty acid synthesis (10). Additionally, the presence of GLP-1 receptor on human hepatocytes has been confirmed as well as activation of signal transduction cascades that lead to decreasment in hepatic steatosis by modulation of insulin signalling pathway after exenatide binding (6). Consistent with this, exenatide plays a direct important role in hepatic fat metabolism but the responsible molecular mechanism needs further research.

The present study is limited by the unavailability of performing liver biopsy, the "gold standard" procedure for diagnosis of NAFLD.

In conclusion, introduction of exenatide therapy to T2DM patients seems to have positive pleiotropic effect in liver, especially in hepatic fat metabolism, although so far it has been mostly studied for its effect in weight reduction, glucose dependent insulin secretion with consequent glucoregulation and positive cardiovascular effects. However, whether the detection of decrease in hepatic fat content in T2DM patients treated with exenatide is due to GLP-1 based therapy itself or the ΔFLI is mostly due to improvement in abdominal fat accumulation needs to be assessed in further follow-up studies.

Author Disclosure Statement

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References:

1. Williamson RM, Glancy S, Perry E et al. Prevalence and Risk Factors for Hepatic Steatosis and Nonalcoholic Fatty Liver Disease in People with Type 2 Diabetes: the Edinburgh Type 2 Diabetes Study, Diabetes Care 2011, 34:1139-1144

2. Ratziu V, Charlotte F, Heurtier A et al. Sampling variability of liver biopsy in nonalcoholic fatty liver disease. Gastroenterology. 2005 Jun;128(7):1898-906.

3. Zelber-Sagi S, Webb M, Assy N. Comparison of fatty liver index with noninvasive methods for steatosis detection and quantification, World J Gastroenterol 2013 January 7; 19(1): 57-64

4. Holst JJ, Vilsbøll T, Deacon CF. The incretin system and its role in type 2 diabetes mellitus Mol Cell Endocrinol. 2009 Jan 15;297(1-2):127-36.

5. Cuthbertson DJ, Irwin A, Gardner CJ et al. Improved Glycaemia Correlates with Liver Fat Reduction in Obese, Type 2 Diabetes, Patients Given Glucagon-Like Peptide-1 (GLP-1) Receptor Agonists, PLoS One. 2012;7(12):e50117

6. Gupta NA, Mells J, Dunham RM et al. Glucagon-like Peptide-1 Receptor (GLP-1R) is present on human hepatocytes and has a direct role in decreasing hepatic steatosis *in vitro* by modulating elements of the insulin signaling pathway, Hepatology 2010, 51(5): 1584–1592

7. Klonoffa DC, Buseb JB, Nielsenc LL et al. Exenatide effects on diabetes, obesity, cardiovascular risk factors and hepatic biomarkers in patients with type 2 diabetes treated for at least 3 years, Current Medical Research and Opinion 2008, 24(1):275-286

8. Buse JB, Klonoff DC, Nielsen LL et al. Metabolic effects of two years of exenatide treatment on diabetes, obesity, and hepatic biomarkers in patients with type 2 diabetes: an interim analysis of data from the open-label, uncontrolled extension of three double-blind, placebo-controlled trials. Clin Ther 2007, 29: 139–153.

9. Sathyanarayana P, Jogi M, Muthupillai R et al. 2011 Effects of Combined Exenatide and Pioglitazone Therapy on Hepatic Fat Content in Type 2 Diabetes. Obesity (Silver Spring)

10. Samson SL, Gonzalez EV, Yechoor V et al. Gene therapy for diabetes: metabolic effects of helper-dependent adenoviral exendin 4 expression in a diet-induced obesity mouse model. Mol Ther 2008;16:1805–1812

Table 1. The comparison between baseline and 6 months after characteristics as well as their changes in the exenatide and OHA group.

Variable	EXENATIDE GROUP (n=87)	OHAGROUP (n=38)	p
Age (years)	57±7	58±10	NS
Gender (m/f)	41/46	20/18	NS
Duration of diabetes (years)	10 ±6	14±6	0.001
BMI baseline (kg/m^2)	38.95±5.32	37.73±5.41	NS
BMI after 6 months (kg/m^2)	35.80±5.56	35.42±4.42	NS
Δ BMI (kg/m^2)	-2.76±2.74	1.21±2.35	0.005
Waist circumference baseline (cm)	119.93±13.08	118.58±16.96	NS
Waist circumference after 6 months (cm)	109.89±16.64	119.26±17.47	NS
Δ waist circumference (cm)	-7.47±12.23	-1.89±5.88	0.035
HbA1c baseline (%)	8.62±1.47	8.67±1.52	NS
HbA1c after 6 months (%)	7.59±1.36	8.65±1.63	0.033
Δ HbA1c (%)	-0.88±1.46	-0.19±1.86	NS
Triglycerides baseline (mmol/L)	2.02 (0.86-10.89)	2.06(0.92-7.72)	NS
Triglycerides after 6 months (mmol/L)	1.76(0.70-6.32)	2.08(0.95-4.42)	NS

Δtriglycerides (mmol/L)	-0.11(-10.2-2.1)	-0.14(-3.1-2.3)	NS
AST baseline (U/L)	24(12-76)	21(13-61)	NS
AST after 6 months (U/L)	22(13-61)	24(11-59)	NS
ΔAST (U/L)	-3(-42-41)	-2(-11-34)	0.05
ALT baseline (U/L)	34(10-89)	24(8-72)	0.015
ALT after 6 months (U/L)	26(13-88)	26.5(11-72)	NS
ΔALT (U/L)	-4(-50-46)	0(-37-26)	0.045
GGT baseline(U/L)	39.50(12-225)	28.50(11-149)	0.027
GGT after 6 months (U/L)	33(12-103)	31(15-173)	NS
ΔGGT (U/L)	-6(-126-26)	0(-34-116)	NS
ALP baseline	85(30-139)	84(39-126)	NS
ALP after 6 months (U/L)	76.5(22-172)	81(15-168)	0.041
ΔALP (U/L)	-4(-57-47)	1(-58-77)	NS
FLI baseline	39.24±27.57	31.65±29.64	NS
FLI after 6 months	19.9±18.65	30.59±24.83	0.046
ΔFLI	-25.95±23.15	-11.01±25.48	0.003