FACTORS ASSOCIATED WITH HEPATIC STEATOSIS IN ADULT GREEK SUBJECTS WITH TYPE 2 DIABETES MELLITUS (#0005)

Angeliki Meritsi[1], Emanuel Manesis[2], Marina Noutsou[1], Panagiotis Koussis[3], Stamatia Rapti[4], Dimitra Latsou[5], Ioannis Tsorlalis[1], Eustathios Tsitsopoulos[4], Hariklia Moshoyianni[6], Spilios Manolakopoulos[7], Dimitrios Pektasidis [8], Anastasia Thanopoulou[1]

1 Diabetic Center, 2nd Department of Internal Medicine, Medical School, National and Kapodistrian University of Athens, Greece, 2 Liver Unit, Euroclinic, Athens, Greece, 3 MRI-CT Department, Biomedicine SA, Athens, Greece, 4 Laboratory of Molecular Genetics, Biomedicine SA, Athens, Greece, 5 Department of Social and Educational Policy, University of Peloponnese, Corinth, Greece, 6 Biomedicine SA, Athens, Greece, 7 Liver and Gastrointestinal Unit, 2nd Department of Internal Medicine, Medical School, National and Kapodistrian University of Athens, Greece, 8 2nd Department of Internal Medicine, Medical School, National and Kapodistrian University of Athens, Greece

ABSTRACT

BACKGROUND: NAFLD is dramatically increasing in parallel with the pandemic of Type 2 Diabetes Mellitus.

OBJECTIVES: To investigate factors affecting hepatic steatosis in adult Greek individuals with established Type 2 Diabetes Mellitus.

METHODS: We investigated 140 consecutive subjects with Type 2 Diabetes attending the Diabetic Outpatient Clinic at an Academic Hospital in Athens, Greece. All of them had demographic, clinical and biochemical data recorded. HS was estimated by Magnetic Resonance Imaging determined by Proton Density Fat Fraction Software (MRI-PDFF) and defined as the percentage of total liver fat divided by the liver volume. Hepatic Steatosis of >5% was considered abnormal. The PNPLA3 (I148M) variant was evaluated as a genetic factor by standard molecular techniques. FIBROMAX™ was also calculated.

RESULTS: Of the 140 participants, median age was 61.7, 46% were females, diabetes duration was 10 years and HbA1c was 6.7%. The median value of Hepatic Steatosis was 7.8. The PNPLA3 rs 738409 CC/CG/GG genotype frequencies were 54.2%, 35%, and 10.8% respectively. In multivariate analysis, PNPLA3 rs 738409 (β= 0.425, p=0.001), waist circumference (β=2.448, p=0.001) and female sex (β=0.419, p=0.002) had a positive effect on hepatic steatosis, while duration of diabetes (β= -0.179, p=0.011) had a negative effect.

CONCLUSION: Hepatic Steatosis in Type 2 Diabetes is the sum of interplay of both genetic and epigenetic factors exerting a positive or negative effect, the most prominent among them being abdominal obesity and PNPLA3 molecular variability.

INTRODUCTION

Non alcoholic fatty liver disease (NAFLD) has attracted the particular interest of medical research over the last two decades, as it affects almost a quarter of the world's population. Type 2 Diabetes seems to be an independent risk factor for the development of NAFLD. Almost 70% people with Type 2 Diabetes are estimated to have NAFLD and 20-30% to have non-alcoholic steatohepatitis (NASH). As with most complex traits, the phenotypic patterns and severity of NAFLD appear to be the outcome of multiple interactions between genetic, epigenetic and environmental factors.

In this study we aimed to investigate factors associated with HS as measured by proton density fat fraction in magnetic resonance imaging (MRI PDFF) in Greek subjects with Type 2 Diabetes unselected for NAFLD.

RESULTS / TABLES

In a univariate analysis HS was directly correlated with ActiTest (r 0.507, p<0.001),NashTest2 (r 0.532, p<0.001),VLDL cholesterol (r 0.219, p=0.016) and triglycerides (r 0.262, p=0.004). HS was also directly correlated with BMI (r 0.379, p<0.001), waist circumference (r 0.290, p<0.001), metabolic syndrome (r 0.199, p=0.029), AST (r 0.377, p<0.001), ALT (r 0.594, p<0.001),GGT (r 0.449, p<0.001) and the G-allele of PNPLA3 rs 738409 (CC vs CG/GG p=0.001) and inversely but significantly correlated with age (r -0.286, p=0.002) and duration of diabetes (r -0.277, p=0.002). Furthermore, a statistically significant difference was found between sex and HS; the median of HS for male sex was 9.1 and for female sex 7.2 (p=0.008). No association was found with FibroTest (p=0.878), HDL (p=0.099), LDL (p=0.614), total cholesterole (p=0.390), HbA1c (p=0.781), ApoA1 (p=0.861) and uric acid (p=0.575).

SUBJECTS BASIC CHARACTERISTICS

Variable	Median (IQR) or N %
Age (years)	61.7 (56.91-65.83)
Sex (Female)	46%
BMI	30.5 (27.30-33.47)
T2DM duration	10 (5.00-16.75)
HbA1c (%)	6.7 (6.20-7.23)
PNPLA3 polymorphisms CC (%) CG (%) GG (%)	 54.2% 35% 10.8%

MULTIVARIATE LINEAR REGRESSION FOR HEPATIC STEATOSIS BY MRI-PDFF

Variable	beta coefficient	95% CI	P value	Correlations		
				Zero-order	Partial	Part
PNPLA3 rs 738409 (CG/GG)	0.425	0.182-0.667	0.001	0.328	0.308	0.267
Waist Circumference (cm)	2.448	1.334-3.563	0.001	0.267	0.235	0.218
Female sex	0.419	0.164-0.674	0.002	0.266	0.310	0.269
Duration of Diabetes (years)	(-) 0.179	(-)0.316- (-) 0.042	0.011	(-)0.311	(-)0.281	(-)0.242

DISCUSSION

Our findings confirm the association, among lipid profile parameters examined, of specifically serum triglycerides and VLD cholesterole with HS. A predominant association between women- but not with men- and HS was identified. This may be explained based on the fact that estrogen levels are different in men and women, with estrogen levels playing a critical role in lipid metabolism. Waist circumference showed superiority over BMI in predicting fatty liver. This may be due to existing evidence that insulin resistance in NAFLD is more related to waist girth than BMI, and fatty liver is a liver manifestation of metabolic syndrome.

In multivariate analysis, duration of diabetes was also identified as independent predictor of HS. One possible explanation is that the significantly increased level of hyperinsulinemia in early Type 2 Diabetes drives uptake of free fatty acids by hepatocytes.

Findings from our study confirm an association between PNPLA3 rs 738409 variant with liver fat content. The specific polymorphism could be useful to identify people needed specific strategies to prevent and detect liver fibrosis and its complications.

Finally, we used FibromaxTM as diagnostic tools for steatosis (SteatoTest), necrosis and inflammation (ActiTest- NashTest 2) and Fibrosis (FibroTest). All these biomarkers although validated for subjects with NAFLD they included a limited percentage of subjects with diabetes and seem to underperform when applied to this population.

SUMMARY

Our study confirms the clinical and lipid profile of subjects with Type 2 Diabetes at risk of developing NAFLD and suggests that PNPLA3 rs 738409 is a major determinant of liver steatosis that might be a useful screening tool for them.

CONCLUSION

Hepatic Steatosis in Type 2 Diabetes is the sum of interplay of various factors exerting a direct or an inverse association, the most prominent among them being abdominal obesity and PNPLA3 molecular variability.