Risk Factors Associated With Non-Alcoholic Fatty Liver Disease in Indians: A Case–Control Study

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Background/Aims: **NAFLD has today emerged as the leading cause of liver disorder. There is scanty data on risk factors associated with NAFLD emanating from India. The present study was conducted to identify the risk factors associated with NAFLD.** ***Methods:*** **464 consecutive NAFLD patients and 181 control patients were subjected to detailed questionnaire regarding their lifestyle and dietary risk factors. Anthropometric measurements were obtained and biochemical assays were done. Comparison of different variables was made between NAFLD patients and controls using principal component analysis (PCA).** ***Results:*** **NAFLD patients had higher BMI [26.25 ± 3.80 vs 21.46 ± 3.08 kg/m^2, *P* = 0.000], waist-hip ratio [0.96 ± 0.12 vs 0.90 ± 0.08, *P* = 0.000] and waist-height ratio [0.57 ± 0.09 vs 0.50 ± 0.06, *P* = 0.000] compared to controls. Fasting blood sugar [101.88 ± 31.57 vs 90.87 ± 10.74 mg/dl] and triglyceride levels [196.16 ± 102.66 vs 133.20 ± 58.37 mg/dl] were significantly higher in NAFLD group. HOMA-IR was also higher in NAFLD group [2.53 ± 2.57 vs 1.16 ± 0.58, *P* = 0.000]. Majority (90.2%) of NAFLD patients were sedentary. Family history of metabolic syndrome (MS) was positively correlated with NAFLD. Dietary risk factors associated with NAFLD were non-vegetarian diet [35% vs 23%, *P* = 0.002], fried food [35% vs 9%, *P* = 0.000], spicy foods [51% vs 15%, *P* = 0.001] and tea [55% vs 39%, *P* = 0.001]. Diabetes, hypertension, snoring and sleep apnoea syndrome were common factors in NAFLD. On multivariate PCA, waist/height ratio and BMI were significantly higher in the NAFLD patients.** ***Conclusion:*** **The risk factors associated with NAFLD are sedentary lifestyle, obesity family history of MS, consumption of meat/fish, spicy foods, fried foods and tea. Other risk factors associated with NAFLD included snoring and MS. (J Clin Exp Hepatol 2015;5:295–302)**

NAFLD

Non-alcoholic fatty liver disease (NAFLD) is a distinct clinico-pathologic entity characterized histologically by a spectrum ranging from simple steatosis to steatohepatitis (NASH), cirrhosis and even hepatocellular carcinoma (HCC).[1,2] NAFLD (steatosis of the liver) is highly prevalent in Western countries.[1] With the introduction of westernized lifestyle and the increasing frequency of obesity in the Asia-Pacific region, the prevalence of NAFLD has increased over the past two decades. Studies from different regions of India have shown that NAFLD is very common in Indians.[3–5] Risk factors implicated in the development of NAFLD are obesity and metabolic syndrome (MS).[6,7] However, it is obvious that NAFLD is multifactorial and identifying the various risk factors associated with NAFLD in our population could help us to intervene in order to prevent its progression to more severe forms of the disease.

The few studies identifying the risk factors associated with NAFLD have emanated from the West, where the profiles of NAFLD patients appear to be different.[8–10] The observed differences in Indian NAFLD patients include a lower frequency of MS,[9,11] lesser degree of adiposity and histological milder disease at presentation.[8] A study that compared Indians with matched Caucasians, Hispanics, Blacks and Eastern Asians showed 2- to 3-fold increase in insulin resistance, and 2-fold increase in hepatic triglyceride content in the Indian subgroup.[12]

The aim of the present study was to compare the dietary pattern, and the anthropometric, metabolic and biochemical parameters between NAFLD patients and controls with the objective to find dietary and lifestyle risk factors associated with NAFLD in our population.

Keywords: fatty liver, diet, lifestyle, anthropometry, metabolic syndrome
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Abbreviations: ALT: Alanine Transaminase; AST: Aspartate Transaminase; BMI: Body Mass Index; FBG: fasting blood glucose; HCC: hepatocellular carcinoma; HC: hip circumference; HDL: high-density lipoprotein; HOMA-B: beta-cell function; HOMA: Homeostatic Model Assessment; IR: insulin resistance; MS: Metabolic syndrome; NAFLD: Non-alcoholic fatty liver disease; NASH: non-alcoholic steatohepatitis; PCA: Principal Component Analysis; SD: standard deviation; WC: waist circumference

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METHODS

This was an observational, analytical, single-centre study of cases and controls attending a Gastroenterology clinic at Cuttack, Orissa from October 2012 to October 2013. The study was approved by the Kalinga Gastroenterology Foundation (KGF) Ethical Committee. Only those subjects who provided informed consent for the study were included.

Four hundred and sixty-four consecutive NAFLD patients and 181 controls were analyzed in this study.

Patients

These were NAFLD patients (n = 464). The diagnosis of NAFLD was made on the basis of ultrasonography and histological confirmation whenever possible. Subjects were considered as cases if they presented with fatty liver defined according to the standard criteria accepted by the American Gastroenterology Association: An increase in hepatic echogenicity taking renal echogenicity as a reference, the presence of enhancement and lack of differentiation in periportal intensity and the vascular wall due to great hyper-echogenicity of the parenchyma.[13]

Liver biopsy was done in 25 patients who gave consent for biopsy. Liver biopsy was done using Bard Biopsy Gun 16 G, through intercostal approach. Grading and staging of liver histopathology was done as per the classification proposed by Kleiner et al. using necro-inflammatory activity for grading of NASH.

Controls

Consecutive age-matched patients who had a normal abdominal ultrasonography, which was performed for various causes, such as abdominal pain and dyspepsia, served as controls.

Exclusion criteria

Patients and controls consuming alcohol >20 g/day, having other known liver diseases (hepatitis viruses A to E, autoimmune disease and Wilson's disease) and those on medications known to induce fatty liver or insulin sensitization, such as estrogens, amiodarone, methotrexate, tamoxifen, glitazones and metformin were excluded.

All patients and controls were questioned about their socio-economic background, occupation, family history of MS and liver diseases, diet pattern and eating habits, smoking and drinking habits, exercise, snoring and symptoms of sleep apnea syndrome in a face-to-face interview. Socio-economic assessment was based on the educational status, occupation and income of the patient as suggested by the modified Kuppuswamy socio-economic scale.[14] The patients were categorized into upper, middle and lower socio-economic status. The average daily tea or coffee use was categorized into 1–3 cups and more than 3 cups/day. Subjects were asked about the consumption of vegetarian food and frequency of consumption of meat, fruits and aerated drinks. Fried foods were defined as those foods that were prepared by submerging them in hot oil either by using a deep fryer or a chip pan, without using water. For all dietary items, the frequency was categorized as never (reference), 1–2 times per week (infrequent), and >3 times per week (frequent). Subjects were asked about whether they took spices in their diet; they were asked to classify the intensity of spicy diet on a subjective basis as mild, moderate or severe. Moderate exercise for $\geq$30 min each time $\geq$4 times weekly was considered to be regular exercise. A questionnaire addressing all these questions was filled up.

The anthropometric assessment included measurements of weight, stature, and waist circumference (WC) and hip circumference (HC). Body mass index (BMI) was calculated as weight (kg)/stature (m^2). The WC and HC were measured at the level midway between the lowest rib and the iliac crest and at the level of the great trochanter, respectively.

The measurements of fasting glucose, triglycerides, cholesterol and high-density lipoprotein (HDL) cholesterol and liver function tests were performed by standard laboratory methods. Serum insulin level was estimated by electro-chemiluminescence using standard kit (Roche-Diagnostics, USA) with autoanalyser Elecsys 2010 (Roche-Hitachi, Japan). IR was calculated using the homeostatic model assessment (HOMA) method using a mathematical model derived from FBG and plasma insulin. The value of HOMA was calculated by the following equation: (fasting insulin (μU/ml) × FBG (mg/dl))/405, and depicted as HOMA-IR value.[15]

In order to identify the various risk factors associated with NAFLD, comparison of dietary pattern and lifestyle, anthropometry indices and biochemical and metabolic parameters was done between the cases and healthy controls.

Statistics

Normally distributed continuous variables were expressed as mean ± SD, and the continuous variables with skewed distribution were expressed as median (range). Student's t-test for unpaired data was used to compare groups when variables are normally distributed; otherwise, the Mann–Whitney test was used. Chi-test was used to compare differences in categorical variables. Logistic regression analysis was used for multivariate modelling. Statistical significance was assumed for $P \leq 0.05$.

Power calculation for sample size determination to yield statistically significant results was performed using G*power version 3.1.3.[16] The sample size required for both cases and controls was 55 (=N) each in order to attain a power of 0.95 with effect size 0.7 for α = 0.05. In the current study, we analyzed data from 460 individuals and 181 controls.