

Original Article

Gamma-glutamyltransferase (GGT) is a Predictor of NAFLD Activity Score for Diagnosing Non-Alcoholic Steatohepatitis (NASH)

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Abstract

Nonalcoholic fatty liver disease (NAFLD) encompasses a spectrum of conditions ranging from simple steatosis to steatohepatitis, advanced fibrosis and end stage liver disease. Despite the high prevalence and severity of hepatic illness, NAFLD remains underdiagnosed, because of few symptoms, lack of accurate laboratory markers. The study was aimed at to evaluate a bio-chemical score for diagnosing non-alcoholic steatohepatitis. A hospital based cross-sectional observational study was carried out for a period of two years from July 2013 to June 2015 in the Department of Hepatology, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh. The study was conducted among 50 patients of NAFLD attending at department of Hepatology and underwent for biochemical investigations and liver biopsy with NAFLD Activity Score (NAS). All data were presented as mean \pm SD and analyzed by SPSS (version 16). Qualitative data were analyzed by Chi-square test and quantitative data were analyzed by student's t-test. Performance of the test were assessed by sensitivity and specificity test. Statistically significant result were considered when p value $<$ 0.05. Total patients were divided into two groups 25 were NASH and 25 were non-NASH. Mean GGT was found 73.6 ± 48.6 U/L in NASH group and 49.9 ± 25.4 U/L in non-NASH group. There was significant difference in the NAFLD activity score

for diagnosing NASH between elevated and normal GGT (P value 0.035). Higher GGT values correlated with higher specificity. The Gamma-glutamyltransferase (GGT) has been proposed as a noninvasive and available marker for assessment of NASH.

Keywords: Nonalcoholic fatty liver disease, Gamma-Glutamyltransferase, NAFLD activity score, Non-alcoholic Steatohepatitis.

INTRODUCTION

Nonalcoholic fatty liver disease (NAFLD) is a metabolic disorder characterized by excessive triglyceride accumulation in hepatocytes.¹ NAFLD has a multifactorial etiology and a combination of environmental, genetic and metabolic factors play a role in the development of advanced disease. NAFLD consists of a wide spectrum of conditions, ranging from simple steatosis to nonalcoholic steatohepatitis (NASH) which can progress to cirrhosis and hepatocellular carcinoma (HCC).² The prevalence of NAFLD increases with increasing age. Obesity, diabetes mellitus (DM), insulin resistance are predisposing factors for NAFLD. Although NAFLD is more common in subject with obesity and diabetes mellitus (DM), it also occurs in lean and non-diabetic subject.³⁻⁵ The fatty liver may be diagnosed if the liver echogenicity exceeds that of renal cortex and spleen and there is attenuation of the ultrasound wave, loss of definition of the diaphragm, and poor delineation of the intrahepatic architecture.⁶ Although liver biopsy remains the 'gold standard', there are practical limitations, including costs and risk.

AST is a hepatic transaminase that plays a role in diagnosis of steatohepatitis. Up to 3.6% of people in the United States have asymptomatic increase in AST⁷. In Asian studies, AST is considered as an independent marker for severity of hepatic fibrosis if it is at least twice as much as the maximum normal value.⁸

The AST/ALT ratio is approximately 0.8 in normal subjects. The AST is greater than the ALT in alcoholic

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hepatitis and a ratio greater than 2:1 is highly suggestive of this disorder. A ratio >1.0 may also suggest the presence of cirrhosis in patients with chronic viral hepatitis.⁹

ALT is a marker of hepatic steatosis or hepatitis¹⁰ and NASH has been associated with slight elevation of liver enzymes¹¹. Patients typically present with asymptomatic serum aminotransferase elevations of 2-3 times the normal¹². This was also explored by Pulzi et al 2011¹³, where majority had mild elevation but less than 5 times upper normal limit and exists in all degree of NAFLD. But Alam et al 2013 showed serum alanine aminotransferase levels were not able to predict NASH.¹⁴

Excess deposition of fat in the liver is associated with an elevated serum Gamma-Glutamyltransferase and insulin resistance.¹⁵ An increased Gamma-Glutamyl transferase level is a risk factor for advanced fibrosis in non-alcoholic fatty liver disease.¹⁶

The Gamma-Glutamyltransferase (GGT) has been proposed as a noninvasive and available marker for assessment of NASH.

METHODS AND MATERIALS

This was an observational, cross-sectional study. Patients of NAFLD attending at outpatient and inpatient department of Hepatology, Bangabandhu Sheikh Mujib Medical University, Dhaka, Bangladesh from July 2013 to June 2015 were included in this study. Twenty five NASH and twenty five Non-NASH patients confirmed by liver biopsy were included in this study. Nonalcoholic fatty liver disease activity score was constructed according to Kleiner et al. 2005¹⁷ with steatosis (0-3), lobular inflammation (0-3), hepatocellular ballooning (0-2) and a separate fibrosis staging (0-4). The proposed non-alcoholic fatty liver disease activity score was the sum of steatosis, lobular inflammation, and hepatocellular ballooning. Non-alcoholic fatty liver disease activity score is a good scoring system. Non-alcoholic fatty liver disease activity score of greater than or equal to 5 correlated with diagnosing non-alcoholic steatohepatitis and the biopsy with scoring of 1 to 4 was diagnosed as simple steatosis fatty liver. Patient's inclusion criteria were the ultrasonographical evidence of fatty liver and patients of 18 to 60 years of age. Exclusion criteria were the significant alcohol intake (>30 g/day in case of male; >20 g/day in case of female)¹⁸, viral hepatitis

(hepatitis B virus, hepatitis C virus), Wilson's disease, autoimmune liver diseases, hereditary hemochromatosis, primary biliary cirrhosis, cirrhosis of liver, pregnancy, comorbid conditions (chronic obstructive airway disease, chronic kidney disease, cardiac failure), hypothyroidism, consumption of drugs causing fatty change in the liver (steroid, oral contraceptive pill, tamoxifen, amiodarone, diltiazem, protease inhibitor). In the American Association for the Study of Liver Diseases Practice Guideline 2018, significant alcohol consumption can be defined as >21 standard drinks per week in men and >14 standard drinks per week in women over 2 years period preceding baseline liver histology. The liver biopsy was done by Trucut liver biopsy needle 14 F, 15 cm. The tissue was processed at the Department of Pathology, by standard protocol in automatic tissue processor (BAVIMED 2050, BAVIMED Laborgeneratebau GmbH, Birkeau, Germany). The processed tissue was then properly embedded on the melted paraffin for making blocks and sections. The sections were stained with hematoxylin and eosin for microscopic examination.

After receiving the liver biopsy report, they were grouped as non-alcoholic steatohepatitis and simple steatosis. Consecutive 25 non-alcoholic steatohepatitis patients and 25 simple steatosis patients confirmed by liver biopsy were included in this study.

Statistical Analysis

All data were presented as mean \pm SD and were analyzed by SPSS (version 16). The qualitative data were analyzed by Chi-squared test and the quantitative data were analyzed by student's t-test. Performance of the test were assessed by sensitivity and specificity test. Statistically significant result were considered when p value <0.05.

RESULTS

Table-I shows that contains Fifty (50) patients were included in this study. Twenty five were NASH and twenty five were non-NASH. Overall, Thirty four (68%) had normal G-GT. G-GT in NASH group were 73.6 ± 48.6 IU/L and in Non-NASH group were 49.9 ± 25.4 IU/L. In Non-NASH group 10% of elevated G-GT had no NASH. There was significant difference in the NAFLD activity score for diagnosing NASH between elevated and normal G-GT (P value 0.035). Higher G-GT values correlated with higher specificity.

Table -I: Clinical and laboratory characteristics of study patients in two group (n=50)

Variables	Group-I NASH (n=25)		Group-II Simple Steatosis (Non-NASH) (n=25)		P Value
	Mean	±SD	Mean	±SD	
Age (years)	41.8	±10.7	39.7	±7.5	0.425 ^{ns}
Weight (kg)	65.6	±8.6	63.3	±9.7	0.444 ^{ns}
Height (cm)	159.2	±9.1	157.7	±8.3	0.545 ^{ns}
BMI (kg/m ²)	26.0	±3.9	25.5	±4.0	0.656 ^{ns}
Waist circumference (cm)	97.9	±9.0	93.9	±9.8	0.139 ^{ns}
Systolic blood pressure (mm of Hg)	129.8	±16.9	128.6	±12.2	0.774 ^{ns}
Diastolic blood pressure (mm of Hg)	80.2	±7.8	81.0	±6.1	0.688 ^{ns}
Platelet count (x10 ⁹ /L)	303.1	±68.7	327.8	±66.8	0.203 ^{ns}
FBS (mmol/L)	6.6	±2.8	5.9	±2.2	0.330 ^{ns}
2HABF (mmol/L)	10.0	±4.2	9.1	±4.7	0.478 ^{ns}
Total cholesterol (mg/dl)	210.0	±48.7	199.9	±38.4	0.419 ^{ns}
LDL (mg/dl)	126.0	±40.5	119.6	±36.7	0.561 ^{ns}
HDL (mg/dl)	40.7	±9.1	36.6	±8.9	0.113 ^{ns}
TG (mg/dl)	209.0	±95.9	222.8	±116.2	0.649 ^{ns}
AST (U/L)	55.2	±30.1	33.6	±20.0	0.004 ^s
ALT (U/L)	97.0	±51.5	55.5	±28.6	0.001 ^s
AST/ALT	0.6	±0.2	0.7	±0.3	0.171 ^{ns}
HOMA-IR	2.4	±1.9	2.3	±1.6	0.841 ^{ns}
GGT (U/L)	73.6	±48.6	49.9	±25.4	0.035 ^s
Serum ferritin(µgm/L)	139.4	±124.5	103.5	±69.9	0.214 ^{ns}

Group I =Nonalcoholic steatohepatitis (NASH) (NAS ≥5-8)

Group II =Non-NASH fatty liver (Simple Steatosis) (NAS 0-4)

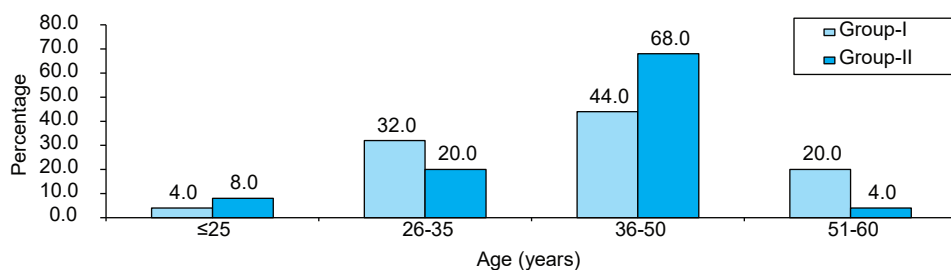


Fig-1: Bar diagram shows age distribution of the study patients.

Figure 1 shows the age distribution of the study patients, here 11(44.0%) patients were in age group 36-50 years in NASH group (Group -I) and 17(68.0%) patient were in age group 36-50 years in Non-NASH group (Group-II). The mean age was found 41.8 ± 10.7 years in NASH group (Group- I) and 39.7 ± 7.5 years in Non-NASH group (Group- II).

Gamma-Glutamyltransferase (G-GT) of the study patients

Table-II shows Mean GGT was found 73.6±48.6 U/L in NASH or group- I and 49.9±25.4 U/L in Non-NASH or group- II. The mean G-GT was statistically significant (p<0.05) between two group.

Table-II : Distribution of the study patients according to GGT (n=50)

GGT (U/L)	Group-I (n=25)		Group-II (n=25)		P value
	n	%	n	%	
Male ≤85 U/L, female ≤55 U/L	14	56.0	20	80.0	
Male >85 U/L, female >55 U/L	11	44.0	5	20.0	
Mean±SD	73.6	±48.6	49.9	±25.4	0.035 ^s
Min-max	24.0	-209.0	12.0	-121.0	s= significant

DISCUSSION

Non Alcoholic Fatty Liver Disease (NAFLD) is a clinico-pathological entity where fat (predominantly Triglyceride) accumulates in liver without significant alcohol ingestion (male>30g/day, Female >20 g/day) or ingestion of certain drugs observed.¹⁹ It encompasses a spectrum of conditions ranging from simple steatosis to nonalcoholic steatohepatitis (NASH), fibrosis and end stage liver disease.²⁰

This study, a hospital based study where most of the patients were from low socioeconomic status. In this study female predominated 30 (60%) out of study population of 50 cases. Among them 16 (32%) were in NASH and 14(28%) non NASH fatty liver (NNFL). Similar female preponderance (57%) was observed in the study conducted in department of Hepatology, BSMMU.¹⁴. This female preponderance in this study may be due to social conservative attitude which bounded most of females to stay at home for household work leading to sedentary life style and also due to intake of carbohydrate predominant food material.

Mean age of patients were 40.8 (±9.2) years. Majority patients 11(44%) belong to 36 to 50 years range in NASH group. 17(68%) patients of NNFL group belong to 36-50 years range. Similar mean age (40.8 ±10.2 years) was observed in the study conducted in department of Hepatology, BSMMU.¹⁴ Mean GGT in NASH group was 73.6± 48.6 U/L, whereas 49.9 ± 25.4 U/L in NNFL group. Mean Gama-GT differed significantly in NASH patients (p value- 0.035). Gama- GT as a marker of disease severity and diagnosis of NASH was explored.¹³. This value correlates with previous data^{14, 21}, where GGT had predictive value for NASH.

ETHICAL ISSUE

Ethical clearance for the study was taken from the Institutional Review Board of the Bangabandhu Sheikh Mujib Medical University prior to the commencement of this study. Approval paper was given by 75th Institutional Review Board, Bangabandhu Sheikh Mujib Medical

University, meeting held on 30th November 2014 (No. BSMMU/2014/13573).

CONFLICT OF INTEREST

No conflict of interest.

CONCLUSIONS

Gamma-glutamyltransferase (GGT) level has the predictive value for diagnosing NASH in NAFLD patients. We propose the use of GGT in NAFLD patients for the detection of NASH from Non- NASH.

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