Observational Study

Analysis of prevalence, risk factors & co-morbid associations between non alcoholic fatty liver disease and type 2 diabetes mellitus in a tertiary care hospital of Eastern India

Manab Nandy[1], Satyaki Chakraborty[2], Asish Kumar Basu[3]

Non Alcoholic Fatty Liver Disease (NAFLD) is one of the commonest causes of elevated liver enzymes in India nowadays. It is well documented that NAFLD prevalence is higher among patients with features of insulin resistance, obesity and dyslipidaemia. This association can promt one to consider NAFLD as a hepatic manifestation of Metabolic Syndrome. On the other hand India is now viewed as one of the largest home of Diabetes in the world . To analyse the close association between Type 2 Diabetes Mellitus (DM) and NAFLD a total of 100 patients were selected in this cross sectional, observational study. This study found significant link between obesity, dyslipidemia, diabetes, and NAFLD, thus establishing some modifiable risk factors and clinical parameters, which should be focused for management by clinicians early during the course of the disease. Significant data for proving an association between the complications of diabetes and NAFLD would highlight a subgroup of Type 2 DM patients requiring earlier intensive therapy and management of risk factors.

[*J Indian Med Assoc* 2018; **116:** 23-6]

Key words **: Non alcoholic fatty liver disease, Diabetes mellitus, Co-morbid associations**

NAFLD refers to the broad range of liver pathology ranging from mild steatosis to non-alcoholic steatohepatitis (NASH) in the absence of significant alcohol consumption. Today it is the fourth most common indication for liver transplantation and is one of the commonest causes for elevation of liver enzymes[1,2]. Prevalence of the NAFLD is estimated to be around 9-32% in the general Indian population, with a higher incidence rate amongst obese and diabetic patients[3]. It is well documented that NAFLD prevalence is higher among patients with features of insulin resistance, obesity and dyslipidaemia. This association can prompt one to consider NAFLD as a hepatic manifestation of Metabolic Syndrome[3].

In contrast, Diabetes has been well documented enough for the general population to be aware of its consequences[4]. China and India lead the world with the largest number of diabetes subjects[5]. The threat of end organ complications, nephropathy, retinopathy and neuropathy looms large over the population of uncontrolled diabetics. Furthermore, 15-25% of patients with NAFLD, progress to cirrhosis and its complications over 10-20 years[5].

Department of Pharmacology, Medical College Kolkata 700073
[1]MBBS, DCH, MD, Associate Professor and Corresponding author
[2]MBBS. Intern, Calcutta National Medical College, Kolkata 700014
[3]MD, DM, Professor & Head, Department of Endocrinology, Medical College, Kolkata 700073

Due to the close association between Type 2 DM and NAFLD, the analysis of risk factors is of utmost importance in identifying individuals susceptible to the co-morbid complications arising from either disease. Significant data for proving an association between the complications of diabetes and NAFLD would highlight a subgroup of Type 2 DM patients requiring earlier intensive therapy and management of risk factors.

Aims and Objectives

The aim of this study is to assess the risk factors associated with development of NAFLD in the diabetic study population. It will also aim to identify any co-relation between NAFLD and end organ changes associated with Diabetes such as nephropathy, neuropathy and retinopathy.

Materials and Methods

A total of 100 patients participated in this cross sectional, observational study. After obtaining a certificate of approval from the Institutional Ethics Committee, Patients with Type 2 DM from the Internal Medicine ward of a Tertiary Medical College of Eastern India, were identified by review of their records. The sample population was chosen on the basis of simple random sampling. Each subject was given an 'Informed Consent' along with an 'Information Sheet', specifying the procedures undertaken in this project, in the language of their choice. The choice

of subjects however was dependent on the following criteria:

Inclusion Criteria : Patients having history of Type 2 DM or suffering directly from its complications.

Exclusion Criteria : Patients suffering from Type 1 DM or Maturity Onset Diabetes of Young (MODY) or any form of Secondary Diabetes, Gestational DM, Patients who are on medication that include steroids or drugs that are known to cause fatty liver such as amiodarone, aspirin, methotrexate, anti-viral drugs (nucleoside analogs), tamoxifen, etc.

Patients consuming alcohol greater than 20 ml/day (or >21 drinks per week) and women consuming alcohol greater than 15 ml/day (or >14 drinks per week) as this automatically excludes diagnosis for NAFLD[4].

After sample selection, a screening procedure was implemented to sort the patients into two groups, A and B. The screening was based on evidence of fatty liver on hepatic ultrasonography. The diabetics not showing evidence of steatosis on ultrasonography were grouped in A, and those showing evidence of the same were grouped in B.

Patients in either group were subjected to laboratory Investigations like Liver function tests, HbA1c, urine albumin and creatinine, and lipid profile.

All of the above data was 'cross tabulated' using Microsoft Excel. Data on subjects of either group was tabulated on separate Excel sheets.

The data after cross tabulation was analyzed using 'SPSS version 22'. A logistic regression model was developed to evaluate predictors of NAFLD. Differences between normally distributed variables were assessed using 'unpaired t test'. Categorical variables were assessed using Chi-Square (χ^2) test. Relative Risk (RR) and Odds Ratios (OR) were calculated for hypothesized risk factors. Assuming a confidence interval of 95%, a p-value of less than 0.05 was considered significant.

Observations and Results

A total of 100 patients (60 males and 40 females) suffering from Type 2 DM were enrolled in the study. The mean age of the patients was 55.83± 7.81 (mean± SD). Out of this a cohort of 52 patients were identified as having NAFLD based on the findings on hepatic ultrasonography (Table 1).

Participants with NAFLD had greater mean age (59.07±7.57 years), duration of diabetes (6.48±1.74) and body mass index (27.53±3.05). This particular cohort of patients had higher levels of HbA1c (7.50±1.12), ALT (56.09±13), AST (54.40±6.20) and triglycerides (205.42±34.90). The prevalence of NAFLD was found to be greater in males than females. This is illustrated in Fig 1.

Table 1— *Compares the demographical, clinical and laboratory characteristics of patients with NAFLD with those without NAFLD*

Variable N	Without NAFLD 48	WITH NAFLD 52	P-value
Male (%)	28(58.4)	32(61.5)	
Female (%)	20(41.6)	20(38.5)	
Age (years)	53.39±7.39	59.07±7.57	NS
Length of DM(years)	4.93±2.07	6.48±1.74	p<0.05
B.M.I (kg/m2)	24.66±2.54	27.53±3.05	P<0.05
ALT (IU/L)	18.10±8	56.09±13	p<0.05
AST (IU/L)	19.23±6.74	54.40±6.20	P<0.05
HbA1c(%)	6.46±1.16	7.50±1.12	p<0.05
Triglycerides (mg/dl)	132.18±40.69	206.71±92.90	p<0.05
Cholesterol (mg/dl)	188±30.50	210±54.40	NS
LDL (mg/dl)	126.00±26.20	142.43±48	NS
HDL (mg/dl)	45.20±8.10	38.42±10.00	NS

Out of the 52 patients found to have NAFLD, 12(23%) had normal BMI, 24 (46%) were overweight and 16 (31%) were obese as illustrated in Fig 2.

BMI of 30 or more was considered obese.

Dyslipidemia was determined according to triglyceride, cholesterol and HDL/LDL levels and reference values.The findings are illustrated in Fig 3. In 61.5% (32) of the NAFLD subjects had dyslipidemia compared to 39.6% (19) of the non-NAFLD subjects : χ^2 value=9.75; RR= 1.53, OR= 2.44, p=0.03 (p<0.05, 95% CI).

Incidence of microvascular complications in either group was assessed. The findings are illustrated in Figs 4-7.

In 55.7% (29) of the patients having NAFLD were diagnosed to also have diabetic nephropathy (DN) compared to 35% (17) of the subjects without NAFLD: χ^2 value=4.16,

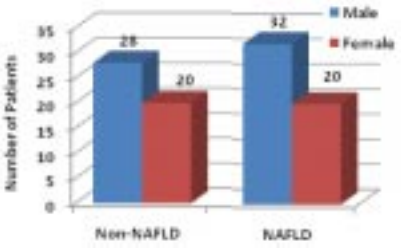

Fig 1— Showing incidence of NAFLD between male and female

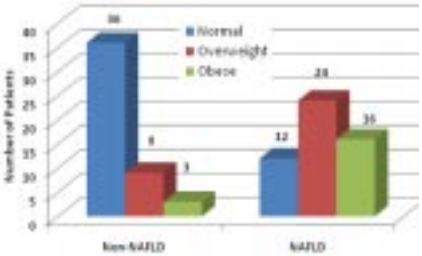

Fig 2 — Showing incidence of NAFLD among overweight and obese patients

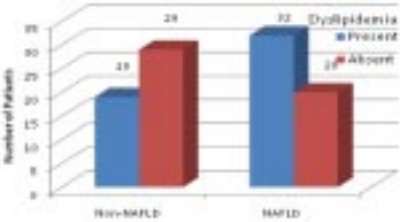

Fig 3 — Comparison of prevalence of dyslipidemia in non-NAFLD and NAFLD group

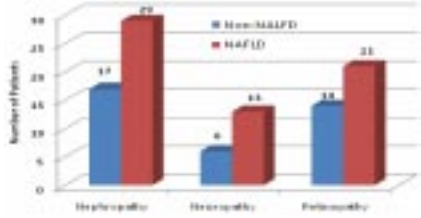

Fig 4 — Incidence of microvascular complications of diabetes mellitus in non-NAFLD and NAFLD group

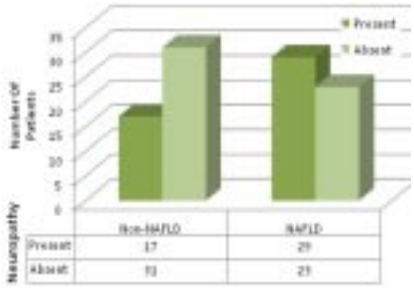

Fig 5 — Comparison of incidence of nephropathy in non-NAFLD and NAFLD group

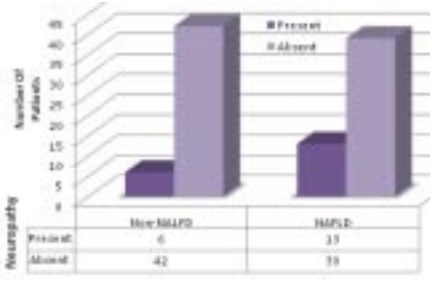

Fig 6 – Comparison of incidence of neuropathy in non-NAFLD and NAFLD group

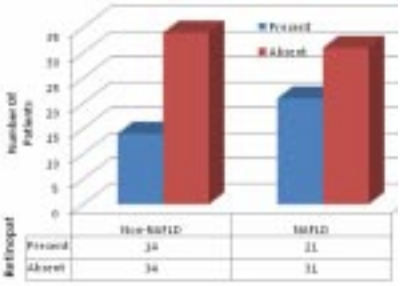

Fig 7 — Comparison of incidence of retinopathy in non-NAFLD and NAFLD group

p=0.041 (p<0.05, 95% CI), OR=2.29, RR= 1.48 (CI= 0.97-2.2).

In 25% (13) of the patients having NAFLD were diagnosed to also have diabetic neuropathy (DPN) compared to 12.5% (6) of the subjects without NAFLD: χ^2 value=2.53, p=0.11 (p>0.05, 95% CI).

In 40.4 % (21)of the patients having NAFLD were diagnosed to also have retinopathy (DR) compared to 29.2% (14) of the subjects without NAFLD: χ^2 value= 1.38, p value= 0.240 (p>0.05, 95% CI).

Discussion

Multiple studies have shown a close association between NAFLD and Type 2 DM. Cusi Kenneth *et al* (2009) pointed out that 60-70% of diabetics show evidence of NAFLD[7]. Sanjay Katra *et al* found the overall prevalence of NAFLD in Type 2 DM patients was to be 56% in Indian population[3]. Gupte P *et al* (2004) and Kamani P *et al* (2007) determined the prevalence to be 12.5% and 20% respectively[8,9]. A number of studies around the world have identified the possible risk factors leading to the development of NAFLD. Type 2 Diabetes Mellitus itself is now considered an independent risk factor for the development of NAFLD[3,4,6].

Study conducted by Hosseinpanah *et al* (2007) found that diabetic patients with NAFLD were more likely to have greater BMI[10].Wen-Shan *et al* (2013) determined the same to be and also came to the conclusion that diabetics with NAFLD have higher levels of liver transaminases as well as lower high density lipoproteins (HDL) level compared to diabetics without NAFLD[11]. This study however used hepatic ultrasonography to detect the presence of steatosis although it has been well established that liver biopsy remains the gold standard for diagnosis of NAFLD[12]. There have been studies linking NAFLD with greater incidences of chronic kidney disease and even identifying NAFLD in the presence of Type 2 Diabetes as a significant cardiometabolic risk factor[13]. In a much more recent study the same author found that diabetic patients suffering from NAFLD had a greater incidence of developing diabetic retinopathy[14].

The prevalence of NAFLD among the diabetic subjects, 52%, is in line with the overall prevalence, 54.5% as postulated by Mohan *et al*[15]. The prevalence of NAFLD in males were greater compared to females, which contraindicates a major pan-Indian study carried out by Kalra *et al* where the researchers had concluded that the female prevalence of NAFLD (60%) was higher than that of males (54.3%)[3].

However the male and female prevalence of 54.3% and 55% respectively is line with many previous studies on co-occurrence of NAFLD and Type 2 DM[9,16].This study revealed that older patients were more likely to develop NAFLD but without statistical significance. Both insulin resistance and obesity are key features of metabolic syndrome and 30% of NAFLD subjects have metabolic syndrome[17].

Greater duration of diabetes and higher HbA1c levels were found to be good predictors of NAFLD prevalence and the presence of microvascular complications. This was similar with the findings by Banerjee *et al* who found that longer duration of diabetes and poor glycemic control were associated with higher rates of progression to a severe form NAFLD, non-alcoholic steatohepatitis (NASH)[18].

This fact is further reinstated by the strong correlation between dyslipidemia, an important predictor of cardiovascular risk, and fatty liver disease. The role of liver enzyme elevation in NAFLD has been widely debated. It has been widely suggested that liver enzyme elevation correlation is related to the degree of steatosis as evidenced on biopsy[8,9,12].

As illustrated in Fig 4, higher rates of microvascular complications were found in the subjects identified as having NAFLD. However, not all the findings were statistically significant. The null hypothesis (H_0) therefore cannot be rejected. The positive correlation of diabetic nephropathy with NAFLD was found to have a p value of 0.041 (p<0.05) however the relative risk of 1.48 lies between a confidence of interval of 0.97-2.20 (includes 1.00 in the reference range), hence eliminating its statistical significance as possible risk factor. Diabetic neuropathy and retinopathy were also found to have a p value of greater than 0.05 and insignificant OR and RR (confidence interval

includes 1.00). The above findings are similar with a study carried out in the Chinese population by Wen Shan et al[11].

Conclusion

This study found significant links between obesity, dyslipidemia, diabetes duration, glycemic control and NAFLD, thus establishing some modifiable risk factors and clinical parameters. Along with medical interventions, these includes dietary modifications and lifestyle changes involving increased exercise. Liver enzyme abnormalities plus Type 2 DM leads to greater risk of cardiovascular and renal disease[13]. Therefore management of NAFLD progression is not just essential for preventing hepatic complications but also important for prevention of cardiovascular disease and renal impairment.

Funding : This project is funded and approved by Indian Council of Medical Research as Short Term Students' Project.

Conflict of Interest : None declared.

References

1 Angulo P — Nonalcoholic Fatty Liver Disease and Liver Transplantation. *Liver Transpl* 2006; **12:** 523-34.
2 Ruhl CE, Everhart JE — Determinants of the association of overweight with elevated serum alanine aminotransferase activity in the United States. *Gastroenterology* 2003; **124:** 71-9.
3 Kalra S, Vithalani M, Gulati G, Kulkarni CM, Kadam Y, Pallivathukkal J, Das B — Study of Prevalence of Nonalcoholic Fatty Liver Disease (NAFLD) in Type 2 Diabetes Patients in India (SPRINT). *J Assoc Physicians India* 2013; **61:** 448-53.
4 Smyth S Heron — Diabetes and obesity: the twin epidemics. *Nature Medicine* 2002; **12:** 75-80.
5 V Mohan V, Sandeep S, Deepa R, Shah B ,Varghese C — Epidemiology of type 2 diabetes: Indian scenario. *Indian Journal of Medical Research* 2007; 217-30.
6 Hui JM, Kench JG, Chitturi S, Suda A, Farrell GC, Byth K, *et al* — Long term outcomes of cirrhosis in non-alcoholic steatohepatitis compared with hepatitis C. *Hepatology* 2003; **38:** 420-7.
7 Cusi, Kenneth — NAFLD in Type 2 Diabetes Mellitus. *Curr Opin Endocrinol Diabetes Obes* 2009; **16:** 141-9.
8 Gupte P, Amarapurkar D, Agal S, Baijal R, Kulshrestha P, Pramanik S, *et al* — Non-alcoholic steatohepatitis in type 2 diabetes mellitus. *J Gastroenterol Hepatol* 2004; 1**9:** 854-8.
9 Amarapurkar D1, Kamani P, Patel N, Gupte P, Kumar P, Agal S, *et al* —Prevalence of non-alcoholic fatty liver disease: population based study. *Ann Hepatol* 2007; **6:** 161-3.
10 Hosseinpanah F, Rambod M, Sadeghi L — Predictors of Non-Alcoholic Fatty Liver Disease in Type 2 Diabetes. *Int J Endocrinol Metab* 2007; **2:** 61-9.
11 Wen-Shan Lv, Rui-Xia Sun, Yan-Yan Gao, Jun-Ping Wen, Rong-Fang Pan, Li Li, Jing Wang — NAFLD and Microvascular Complications in Type 2 Diabetes Mellitus. *World J Gastroenterology* 2013; **19:** 3134-42.
12 Saadeh S, Younossi ZM, Remer EM — The Utility of Radiological Imaging in NAFLD. *Gastroenterology* 2002; **123:** 745-50.
13 G Targhar, M Chonchol, G Zoppini, E Bonora — Risk of chronic kidney disease in patients with non-alcoholic fatty liver disease: Is there a link? *Journal of Hepatology* 2011; **54:** 1020-9.
14 G Targhar, G Lippi, L Bertolini, S Rodella — NAFLD is independently associated with increased prevalence of Chronic Kidney Disease proliferative/laser treated retinopathy in Type 2 DM patients. *Diabetologia* 2008; **51:** 444-50.
15 Mohan V, Farooq S, Deepa M, Ravikumar R, Picthumoni CS — Prevalence of non-alcoholic fatty liver disease in urban South Indians in relation to different grades of glucose intolerance and metabolic syndrome. *Diabetes res Clin Pract* 2009; **84:** 84-91.
16 Uchil D, Pipalia D, Chawla M, Patel R, Maniar S, Juneja A — Non-Alcoholic Fatty Liver Diesease(NALFD)- the hepatic component of metabolic syndrome. *J Assoc Physicians India* 2009; **57:** 201-4.
17 Browning JD, Szczepaniak LS, Dobbins R, Nuremberg P, Horton JD, Cohen JC — Prevalence of hepatic steatosis in an urban population in the United States: impact of ethnicity. *Hepatology* 2004; **40:** 1387-95.
18 Banerjee S, Ghosh US, Dutta S — Clinicopathological profile of hepatic involvement in type-2 diabetes mellitus and its significance. *J Assoc Physicians India* 2008; **56:** 593-9.
19 Popovic L, Zamaklar M, Lalik K, Vasolic O — Analysis of effect of type 2 diabetes duration on beta cell secretory function and insulin resistance. *Srp Arh Celok Lek* 2006; **134:** 219-23.