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Original Research Article

A study of correlation between clinical profile of patients with non-alcoholic fatty liver disease and individual components of metabolic syndrome at a tertiary care centre in Uttarakhand region: an observational cross-sectional study

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ABSTRACT

Background: Research indicates that the occurrence of non-alcoholic fatty liver disease (NAFLD) in India is reported to range from approximately 9% to 32%. This study aims to examine the clinical characteristics of subjects diagnosed with NAFLD and investigate the association between NAFLD and individual components of metabolic syndrome.
Methods: This was a cross-sectional, hospital-based study, conducted over a period of 2 months from May 2017 to June 2017 in patients attending medicine OPD or admitted in IPD in AIIMS Rishikesh. Study population included sonologicaly diagnosed cases of NAFLD of age group 18 to 70 years. Patients with a history of jaundice, positive viral markers for hepatitis C or hepatitis B, pregnant women, individuals who consume alcohol, and patients with a history of hepatotoxic drug consumption were excluded.
Results: A total of 40 NAFLD cases, as confirmed ultrasonographically, were included in the study. Out of all the patients diagnosed with NAFLD, 26 (65%) were found to have metabolic syndrome based on the NCEP ATP III criteria. Upon investigating the correlation between NAFLD and individual components of metabolic syndrome, a significant association ($p<0.05$) was observed with BMI, waist circumference, systolic and diastolic blood pressure, as well as serum triglyceride levels.
Conclusions: Uttarakhand being hilly region mainly, it is expected that lifestyle will be healthy due to cultural and environmental factors, but rapid urbanisation has led to change in lifestyle which needs to be addressed in the population of this Himalayan state.

Keywords: Lifestyle modification, Metabolic syndrome, Non-alcoholic fatty liver disease

INTRODUCTION

According to the definition, non-alcoholic fatty liver disease (NAFLD) is characterized by the collection of excessive fats in the liver, accounting for >510% of the liver weight, in the absence of notable alcohol intake. The amount of alcohol intake considered noteworthy varies, but as per Indian guidelines, a threshold of 16g/day for males and 8g/day for females can be taken as a reference.[1] NAFLD encompasses a range of liver conditions that include simple hepatic steatosis (accumulation of fat in the liver), non-alcoholic steatohepatitis (NASH), and progressive liver fibrosis, which can eventually lead to liver cirrhosis. One of the

main cause of the morbidity and mortality in patients suffering from liver damage is NAFLD.[2] Research indicates that the occurrence of NAFLD in India is reported to range from approximately 9% to 32%.[3] It has been found to be associated with various metabolic abnormalities and thus have implications far beyond liver.[4] When many metabolic disorders clusters together or if there is progression of any metabolic syndrome, it can lead to NAFLD. Thus, NAFLD is linked with various metabolic syndrome variables like obesity, type II diabetes mellitus, dyslipidaemia, and hypertension. These metabolic disorders increase the risk of cardiac complications such a coronary artery disease (CAD), structural heart diseases (such as ventricle wall abnormalities and cardiac failure), cardiac valve diseases and arrhythmias.[5] There are many western studies that have shown the significant relation between the metabolic syndrome and non-alcoholic fatty liver disease but only few reports are reported in India. The common mechanism among various pathophysiological mechanisms linked to the pathogenesis of NAFLD and metabolic syndrome is the insulin resistance present in these patients.[6] Given that NAFLD has the potential to progress to cirrhosis and liver failure, and the conventional belief that liver disease confers protection against cardiovascular disease, conducting fundamental medical research in this area holds considerable importance and no much of the information is available about this topic. Published literature on NAFLD from India, especially in Uttarakhand region, is sparse. Ethnic and racial factors have found to be influencing liver fat accumulation which makes this study more significant in this part of the country.[7] In conclusion, although there is a lot of work done, yet studies are required to highlight various features of NAFLD in India. The objective of this study was to investigate the clinical characteristics of NAFLD patients stratified by severity as diagnosed by ultrasonography, and using the modified NCEP ATP III criteria to determine the presence of metabolic syndrome in NAFLD patients and significance of the association between the two.

The aim of this study were to evaluate the clinical characteristics of patients with NAFLD based on the degree of fatty liver as determined by USG, and to find whether metabolic syndrome and NFLD are notably related to each other, using NCEP ATP III criteria.

METHODS

A cross-sectional study was conducted in a tertiary care hospital setup, which is a type of observational and analytical study. Clearance was taken for the study from the institutional ethics committee (IEC). The study population included the patients of NAFLD, who have been diagnosed by abdominal ultrasonography, attending OPD or admitted in IPD of Department of Medicine. This cross-sectional, hospital-based study was conducted over a period of 2 months from May 2017 to June 2017 in 40 patients attending medicine OPD or IPD which included patients greater than 18 years of age, detected to have NAFLD on USG abdomen. All the patients who were younger than 18 years or older than 70 years were not included. Pregnant females; patients who had jaundice or were detected positive for hepatitis C or hepatitis B; patients consuming antiviral agents, steroids, calcium channel blockers, synthetic estrogen, and heparin; patients consuming significant amount of alcohol men-16gms/day, women-8gms/day were also not included in the study.

An informed written consent (stating all the details about the nature and purpose of the study in a way that was understandable by the subject) was signed by each subject recruited in the study. Full confidentiality of the patient was maintained and assured no personal information of the patient will be cited at any point of time during or after the completion of the study. All the subjects were evaluated for metabolic syndrome according to latest NCEP ATP III guidelines. It recommended that the metabolic syndrome is to be diagnosed using 5 variables (≥ three of them) for diagnosing, namely abdominal girth, serum triglyceride level, serum high-density lipoprotein (HDL) cholesterol level, blood pressure, and fasting plasma glucose level. Patients diagnosed with NAFLD were staged according to severity using the ultrasonography technique. The staging of NAFLD is done as follows: Grade 1: mild hyperechoeic intensities present with liver appearing brighter than renal cortex with normal diaphragm and vessel findings. Grade 2: Moderately hyperechoeic lesions present on ultrasound. Intrahepatic blood vessels and diaphragm showing slight impairment. Grade 3: Marked hyperechoeic lesions is present on ultrasound. Intrahepatic blood vessel, diaphragm are not visualised.

A questionnaire/case record sheet was made and filled by asking questions by the principal investigator himself. The case record sheet included particulars, chief complaints, anthropometric values like height, weight, waist circumference, investigation values and any medical history related to NAFLD. History about various aspects of liver disease was taken from each subject including history of jaundice, hepatitis B and hepatitis C. Drug history and alcohol history was also taken. Anthropometric measurements were done on each subject. Subjects were weighed in minimal clothing, using a digital balance. The height was measured, with stadiometer and expressed in cm. Body mass index (BMI) was calculated from the height and weight as BMI = weight (kg)/height^2 (m). Abdominal girth around waist was measured at the narrowest point between the lowest rib. Seated right arm blood pressure was taken by a trained nurse after a rest of 5 min. Overnight fasting blood sample was collected by phlebotomist after informed consent. The blood samples were analysed for (i) fasting blood glucose (ii) lipid profile including serum HDL and serum triglycerides. Results ofall the tests were intimated to the subjects. Metabolic syndrome was diagnosed according to these guidelines given by NCEP:

elevated waist circumference measured by measuring tape, for men ≥102 cm (≥40in) and for women ≥88 cm (≥35in); triglycerides ≥150 mg/dL, HDL cholesterol estimated by CHOD-PAP method, for men <40 mg/dl and for women <50 mg/dL; raised blood pressure ≥130/85 mmHg as measured by properly calibrated sphygmomanometer or use of antihypertensives, measured in the right arm in seated position; elevated fasting glucose ≥ 110 mg/dL or already on medication for hyperglycemia, estimated by GOD-POD method. Then, each component of metabolic syndrome was correlated with allthe cases of different severity of NAFLD included in the study. Instruments used were weighing machine, measuring tape, stadiometer, stethoscope, sphygmomanometer. Quality ofthe study was taken care of and all the guidelines given in University Code of Good Practise in Research (UCOGPR) were followed to ensure that. All the parameters were measured and analysed by a single person eliminating the chance of observer's bias. Proper documentation of the procedures and methods was done. Laboratory tests were done at well calibrated standard labs of AIIMS, Rishikesh having good quality facilities and equipment and well trained and competent staff thereby ensuring accuracy and precision of the test results used inthe study.

Data was recorded in Microsoft excel sheet and bio-statistical analysis was done to know significant association of independent variable with dependent variable using SPSS software (v23) by applying different statistical tools like ANOVA and Pearson correlations.

RESULTS

The study enrolled 40 patients diagnosed with NAFLD using ultrasound. Among them, 32 cases had grade I non-alcoholic fatty liver, while 8 cases had grade II non-alcoholic fatty liver with mean age of 46.95±7.39 years, and the male-to-female ratio 1:1.

Table 1: Clinical profile of subjects with NAFLD.

	Mean	SD	N
Age (years)	46.95	10.95	40
BMI (kg/m^2)	28.740	3.1973	40
Waist circumference (cms)	103.300	6.6860	40
BP (systolic) (mm Hg)	131.65	17.510	40
BP (diastolic) (mm Hg)	81.85	11.012	40
S. triglyceride (mg/dl)	162.10	39.520	40
S. HDL (mg/dl)	39.80	6.806	40
Fasting blood glucose (mg/dl)	116.95	25.771	40

Table 1 contains clinical and laboratory data for all patients included in study. Mostly, patients were asymptomatic in relation to fatty liver and came with non-specific symptoms and fatty liver was mostly an accidental finding. Average BMI was 28.74±3.19 kg/m^2. 65% (26 of 40 patients) presented with metabolic syndrome which fulfilled the NCEP ATP III guidelines. In Grade I fatty liver patients, 20 (62.5%) had metabolic syndrome while in Grade 2 fatty liver patients, 6 (75%) had metabolic syndrome (Table 2).

Table 2: Distribution of prevalence of metabolic syndrome in different grades of fatty liver.

Metabolic syndrome	Grade 1 NAFLD (n=32) (%)	Grade 2 NAFLD (n=8) (%)
Present	20 (62.5)	6 (75)
Absent	12 (37.5)	2 (25)

Table 3: Comparison of means of individual components of metabolic syndrome in patients of NAFLD with or without metabolic syndrome.

		N	Mean	Std. deviation
BMI	0*	14	25.829	3.1012
	1*	26	30.308	1.9089
	Total	40	28.740	3.1973
WC (cms)	0	14	100.357	9.7496
	1	26	104.885	3.5842
	Total	40	103.300	6.6860
BPS	0	14	121.00	11.910
	1	26	137.38	17.516
	Total	40	131.65	17.510
BPD	0	14	74.00	7.845
	1	26	86.08	10.202
	Total	40	81.85	11.012
S. triglyceride	0	14	133.14	23.471
	1	26	177.69	37.783
	Total	40	162.10	39.520
S. HDL	0	14	40.86	10.022
	1	26	39.23	4.366
	Total	40	39.80	6.806
FBG	0	14	114.29	27.717
	1	26	118.38	25.109
	Total	40	116.95	25.771

*0= metabolic syndrome absent
*1= metabolic syndrome present

The association of fatty liver grading with individual components of metabolic syndrome was found in case of systolic blood pressure and fasting blood glucose only (Table 4). When correlating NAFLD with or without metabolic syndrome with individual components of metabolic syndrome, significant association [p value less than 0.05] was found with BMI, abdominal girth, systolic and diastolic blood pressure, and serum triglyceride but not with fasting blood glucose or serum HDL (Table 5).

There were 32 (80%) patients had increased waist circumference. 24(60%) had high blood pressure (hypertension). The fraction of patients with hypertension

is directly proportional to the grade of NAFLD (Table 6). Fasting blood glucose was found to be abnormal in 18 (45%) subjects. 43.75% and 50% of Grade 1 and Grade 2 fatty liver patients had impaired fasting glucose (Table 6). 34 (85%) subjects had low HDL levels while 24 (60%) had hypertriglyceridemia (Table 6). All the variables followed the same trend with fatty liver grade i.e., as the fatty liver becomes more severe, all these variables become more impaired. On comparing metabolic syndrome variables in NAFLD patients having and not having metabolic syndrome, all variables were significant statistically, but HDL and fasting blood glucose didn't show any significance (Table 3, 5).

Table 4: Correlation of fatty liver grading with individual component of metabolic syndrome.

		BMI	WC	BPS	BPD	S. triglyceride	S.HDL	FBG
FLG	Pearson correlation	-0.26	0.299	0.386	0.248	-0.286	0.108	0.325
	Sig. (2 tailed)	0.873	0.061	0.014	0.122	0.073	0.508	0.041
	N	40	40	40	40	40	40	40

*Correlation is significant at the 0.05 level (2-tailed).

Table 5: Correlation of NAFLD with and without metabolic syndrome to individual components of metabolic syndrome.

				Sum of Squares	df	Mean Square	F	Sig.
BMI	Between groups	(Combined)		182.569	1	182.569	32.100	0.000
		Linear term	Unweighted	182.569	1	182.569	32.100	0.000
			Weighted	182.569	1	182.569	32.100	0.000
	Within groups			216.127	38	5.688		
	Total			398.696	39			
WC (cms)	Between groups	(Combined)		186.532	1	186.532	4.553	0.039
		Linear term	Unweighted	186.532	1	186.532	4.553	0.039
			Weighted	186.532	1	186.532	4.553	0.039
	Within groups			1556.868	38	40.970		
	Total			1743.400	39			
BPS	Between groups	(Combined)		2442.946	1	2442.946	9.757	0.003
		Linear term	Unweighted	2442.946	1	2442.946	9.757	0.003
			Weighted	2442.946	1	2442.946	9.757	0.003
	Within groups			9514.154	38	250.372		
	Total			11957.100	39			
BPD	Between groups	(Combined)		1327.254	1	1327.254	14.826	0.000
		Linear term	Unweighted	1327.254	1	1327.254	14.826	0.000
			Weighted	1327.254	1	1327.254	14.826	0.000
	Within groups			3401.846	38	89.522		
	Total			4729.100	39			
S. triglyceride	Between groups	(Combined)		18060.347	1	18060.347	16.016	0.000
		Linear term	Unweighted	18060.347	1	18060.347	16.016	0.000
			Weighted	18060.347	1	18060.347	16.016	0.000
	Within groups			42851.253	38	1127.665		
	Total			60911.600	39			
S. HDL	Between groups	(Combined)		24.070	1	24.070	0.513	0.478
		Linear term	Unweighted	24.070	1	24.070	0.513	0.478
			Weighted	24.070	1	24.070	0.513	0.478
	Within groups			1782.330	38	46.903		
	Total			1806.400	39			
FBG	Between groups	(Combined)		152.889	1	152.889	0.226	0.638
		Linear term	Unweighted	152.889	1	152.889	0.226	0.638
			Weighted	152.889	1	152.889	0.226	0.638
	Within groups			25749.011	38	677.606		
	Total			25901.900	39			

Table 6: Distribution of grades of NAFLD with and without metabolic syndrome.

Variables	Grade 1 NAFLD (n=32)		Grade 2 NAFLD (n=8)	
	MS yes	MS no	MS yes	MS no
Central obesity WC>88 cms(F) >102 cms(M)	18 (56.25)	6 (18.75)	6 (75)	2 (25)
Impaired fasting glucose FBG>110 mg/Dl	12 (37.5)	2 (6.25)	2 (25)	2 (25)
Hypertensive BP>130/85 mm of Hg	12 (37.5)	6 (18.75)	6 (75)	0
Low HDL <40 mg/dl(M), <50 mg/dl(F)	20 (62.5)	10 (31.25)	2 (25)	2 (25)
Hypertriglyceridemia S. triglyceride >150 mg/dl	26 (81.25)	2 (6.25)	4 (50)	0

DISCUSSION

In recent times, NAFLD has emerged as a prevalent and significant cause of chronic liver disease worldwide, despite previously being considered of lesser importance. NAFLD can cause hepatocellular carcinoma. NAFLD is also an independent risk factor leading to cardiovascular disease (CVD). NAFLD therefore possesses many complications apart from only affecting the liver. NAFLD has gained significant attention during the global epidemic of morbid obesity and metabolic syndrome. Some researchers now posit that NAFLD could be regarded as an additional component of the metabolic syndrome.[8] The pathology behind NAFLD has not been clearly understood yet. However, one mechanism is the excessive collection of lipids in the liver. Excess lipids lead to inflammation and increased formation of inflammatory cytokines such as TNF alpha, interleukins, leptins, etc which can lead to insulin resistance and can also cause increased oxidation of fats which can progress to hepatitis and even fibrosis in later stages.[9] The existing published data on clinical profile ofpatients with NAFLD is sparse but presently, a lot of studies are coming up with variable results pointing in one direction. The conclusion is made on the (i) the fact that NAFLD is a recently recognized condition, (ii) it is presumed that the NAFLD is a benign condition with indolent course, (iii) due to high number of viral hepatitis cases in India, the priority of this condition is less. About 20% and 68% NAFLD patients were overweight and obese in a study according to Asia-Pacific criteria.[10] Studies also show the prevalence of hypertension (10%-12%) in cases of NAFLD.[11] Since, the metabolic syndrome is becoming an emerging problem in India, the risk for this disease is yet unknown but estimated to be increased in patients with the metabolic syndrome. A study found 43.2% prevalence of metabolic syndrome in Indian population and its prevalence is likely increasing.[12] One study found that approximately 64.6% of patients having NAFLD were also suffering from metabolic syndrome according to criteria for waist circumference.[13] Same findings are also reported in other studies.[14] These studies also showed a direct relationship between grades of fatty liver and increase in prevalence of individual components of metabolic syndrome e.g. in a study, as the grade of fatty liver disease increased, the percentage of hypertensive patients also increased dramatically. A similar trend was seen with other components too like hypertriglyceridemia was seen in 25%, 77.2% and 85.7% of patients with Grade I, II and III NAFLD. The various parameters are significantly derranged in higher grades of NAFLD and is greater in patients of NAFLD who are suffering from metabolic syndrome than those who are not**.** Thus, it can be inferred that the increasing severity of fatty liver disease is directly proportional to the various components of metabolic syndrome.

The corresponding rates for general and obese patients of NASH are 3-4% and 15-20%, respectively.[15,16] One study found that almost 50% of patients suffering from fatty liver disease were also suffering from metabolic syndrome and about 90% had about one criteria for metabolic syndrome.[17] Among other Indian centers, metabolic syndrome was present in 21-68% of NAFLD patients, and at least one of the above components of the metabolic syndrome is present in almost all the patients.[18,19] Even LDL cholesterol and glycated haemoglobin levels were high in patients with NAFLD. The levelsof glycated haemoglobin and LDL cholesterol were significantly altered in patients with NAFLD.[20] Considering and evaluating the already published data in this field and analyzing the relationships between various studies, it can be concluded that NAFLD and metabolic syndrome are correlated at the basic level and their association needs to be assessed. In the present study we aim to corroborate this finding in our settings.[21]

The metabolic syndrome is emerging as an epidemic in developing countries like India. Recent data shows strong association between the entities of metabolic syndrome and NAFLD. Among the patients diagnosed with NAFLD, 65% (26 cases) met the criteria for metabolic syndrome according to the NCEP ATP III criteria.[22] According to earlier investigations conducted by Duseja et al and Bajaj et al, the prevalence of increased waist circumference was reported to be 58.7% and 47.1% respectively.[11,23] Duseja et al reported a 50% prevalence of metabolic syndrome among NAFLD patients, while Uchil et al found a similar association with a prevalence of 47.1%.[11,14] Furthermore, 92.3% of NAFLD patients with metabolic syndrome had increased waist circumference (male >102 cm, female >88 cm), which was significant with $p<0.05$ (Table 5). However, the current study shows a higher percentage of cases with increased waist circumference compared to these

previous findings. Type 2 diabetes mellitus, which is a significant component of metabolic syndrome, often coexists with obesity and NAFLD. Both NAFLD and fibrosis have a strong association with diabetes mellitus. The average plasma glucose level (fasting) of NAFLD patients with metabolic syndrome was 116.95±25.771 mg/dl (Table 1). In this study, 12 (30%) patients had diabetes (fasting plasma glucose greater than 126 mg/dl), which is greater than the Kaushal et al study (9%).[18] Additionally, 14 (35%) cases were prediabetic (fasting blood glucose greater than 110 mg/dl), which is lesser as compared with the 63% reported by Deb et al.[13] Duseja et al reported that 72.4% and Bajaj et al reported that 28% of patients were prediabetic in their studies and had statistical significance when compared to NAFLD without metabolic syndrome (Table 5).[11,23] Similar findings with respect to blood pressure greater than 130/85 was found in our study (45%) and that conducted by Bajaj et al (23.1%) and Deb et al (43.6%).[23,13] The average mean BP was 131.65±17.5/81.85±11.01 mmHg. Among patients with NAFLD and metabolic syndrome, 22 (84.6%) had triglyceride levels >150 mg/dl which is notably higher than study conducted by Uchil et al which showed 43.6% and Bajaj et al which showed 23.1% but similar to Deb et al.[14,23,13] 22 (84.6%) patients showed HDL of less than 40 mg/dl in males and less than 50 mg/dl in female with an average of 39.8±6.8 mg/dl which is notably greater as compared to what is described as 66.7% by Bajaj et al and 54.8% by Deb et al.[23,13] There are significant differences in the derangement of various parameters between cases of NAFLD with and in the absence of metabolic syndrome. Also, Grade II has more deranged parameters as compared to the components than Grade 1 (Table 6). Hence, it can be concluded that there is a direct correlation between the severity of NAFLD and the presence of metabolic syndrome.

This study has some limitations. We could not include any Grade III NAFLD subject due to limitation of time and paucity of Grade III subjects i.e., a period of 2 months can be considered insufficient for such a study. We can extend the study and include more diverse patients in the study to get more valuable results.

CONCLUSION

This study established a strong correlation between NAFLD and metabolic syndrome. The results of the current study demonstrate a higher occurrence of metabolic syndrome components in individuals with NAFLD. The severity of fatty liver disease was found to be positively correlated with the extent of impairment in various components of metabolic syndrome. As the NAFLD is mostly asymptomatic and its association with metabolic syndrome shows that it increases the risk of cardiac complications such a coronary artery disease, structural heart diseases (such as ventricle wall abnormalities, cardiac failure), cardiac valve diseases and arrhythmias, thus its early detection has become necessary. Early detection can help the clinician to suggest lifestyle modification and dietary changes to the patient to prevent progression of the severity of the disease and subsequently prevent diabetic and cardiovascular complications of the disease and thereby reducing liver related morbidity and mortality. Extended research in this area will help the patients to get a multi-disciplinary approach and thereby look for holistic measures of prevention and better treatment.

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