



## Original Research Article

# PREVALENCE OF NON-ALCOHOLIC FATTY LIVER DISEASE AND ITS ASSOCIATED RISK FACTORS AMONG PEOPLE VISITING A TERTIARY CARE CENTRE IN CHENNAI: A CROSS SECTIONAL STUDY

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### ABSTRACT

**Background:** Non-alcoholic fatty liver disease (NAFLD) is characterized by an abnormal accumulation of fat in the liver in the absence of secondary causes. It is a hepatic manifestation of metabolic syndrome. There is a rising trend in the prevalence of the disease in India. NAFLD is itself a precursor of cirrhosis, hepatocellular carcinoma and chronic liver failure. The aim is to assess the prevalence of NAFLD and its associated risk factors among individuals attending the outpatient clinic at a tertiary care centre in Chennai.

**Materials and Methods:** A hospital cross-sectional study was carried out in Government Medical College, Omandurar Government Estate, Chennai over 3 months (February - May 2023). A sample size of 111 participants were included using convenience sampling method. An abdominal ultrasound was carried out to diagnose the patients with NAFLD using standardized echogenicity criteria. Bivariate analysis (Chi-square test) and multiple logistic regression was used to identify significant associations.

**Results:** During the study period of 1 year 111 BMEs, both bone marrow aspirations and BMB were performed for various indications. The mean age was 40.6 years of these, the Male: Female ratio was 1.3:1. In this study comprising 111 patients, the majority were in the 21–40 years age group (42 cases, 37.8%), followed by the 41–60 years group (28 cases, 25.2%), and the 2–20 years group (24 cases, 21.6%). The least representation was from the 61–80 years age group (17 cases, 15.3%). A slight male predominance was observed overall, with 62 males (55.9%) and 49 females (44.1%). Assessment of bone marrow cellularity revealed that normocellular marrow was most common (36%), followed by hypercellular (28%), hypocellular (16%), and diluted samples (13%).

**Conclusion:** The overall prevalence of NAFLD was 32.43% (95% CI). We identified age ( $p = 0.009$ ), dietary pattern (non-vegetarian diet,  $p = 0.007$ ) and hypertension ( $p=0.003$ ) as factors significantly associated with NAFLD. Among NAFLD-positive cases, 91.7 % had normal serum transaminase levels and 44.1 % were anemic.

**Keywords:** Non-Alcoholic Fatty Liver Disease (NAFLD), Prevalence, Risk Factors, Obesity, Type 2 Diabetes Mellitus, Metabolic Syndrome, Dyslipidemia, Cross-Sectional Study.

### INTRODUCTION

A leading global cause of chronic liver disease and affecting one in every four individuals, non-alcoholic

fatty liver disease (NAFLD) is characterized by hepatic steatosis in the absence of secondary causes of accumulation of fat in the liver, such as significant alcohol consumption, viral hepatitis, autoimmune

disorders, steatogenic medications etc. It encompasses a wide spectrum of diseases ranging from benign hepatic steatosis to more aggressive non-alcoholic steatohepatitis, which could then progress to liver fibrosis and in the worst cases, even cirrhosis.<sup>[1]</sup>

In 2022, Riazi et al, in a meta-analysis reported that there has been a global trend in the increasing prevalence of this disease, with a recent worldwide prevalence estimate of 32%.<sup>[2]</sup> Another systematic review and meta-analysis that included several population-based studies in the Asia-Pacific region has identified an overall prevalence of 29.6%.<sup>[3]</sup>

In India specifically, a systematic review conducted in 2021 estimated NAFLD prevalence of 28.1% in community based studies and 40.8 % in hospital-based studies.<sup>[4]</sup> However, this meta-analysis could not assess the demographic details of the participants, such as the number of males/females affected, their age groups, BMI categories, associated comorbidities (PCOS, metabolic syndrome) etc. which are all important predictors of NAFLD. There was also a paucity of studies done in Tamil Nadu alone.

Although the gold standard for the diagnosis of liver fibrosis is a biopsy, it remains an invasive procedure and unless an accurate measurement of the stage of fibrosis is required, it is not an ideal technique for screening. Newer advanced modalities such as magnetic resonance elastography (MRE) and transient elastography (TE); which are non-invasive techniques for detection of liver fibrosis and are just as efficient as carrying out a biopsy; are being used in the US for screening.<sup>[5,6]</sup> However, they are not economically feasible due to high equipment costs and requirement of more specialized training to operate. Thus, ultrasound is a cheaper alternative.

Documenting the prevalence of disease and identifying its associated risk factors will help in obtaining a better understanding of the natural history of NAFLD. This will help in promoting awareness about the development of the disease to the general public and therefore encourage behavioral change. This will also contribute to strategies that will help in detecting, treating, monitoring and preventing this disease.

## MATERIALS AND METHODS

**Study design, study setting and study period:** An institutional based cross-sectional study was carried out to assess the prevalence of non-alcoholic fatty liver disease among people residing in Chennai. The data was collected over a period of 3 months from February 2023 to May 2023.

**Study population:** The study population includes all the people who are above the age of 18 years both men and women, non-alcoholics as per the AUDIT score (less than 8), residing in Chennai for more than 6 months and visiting the outpatient services at Government Medical College and Hospital,

Omandurar Government Estate, Chennai. **The following patients were excluded:**

1. Individuals aged less than 18 years.
2. Individuals who have an AUDIT score of more than 8.<sup>[7]</sup>
3. Individuals who have already been diagnosed with NAFLD.
4. Individuals who are already on medication for some chronic liver conditions.
5. Individuals who are already seropositive for hepatitis.

**Sampling technique and Sample size:** Based on the study “Prevalence of Non-alcoholic fatty liver disease in an adult population in a rural community of Haryana, India” where  $p = 30.7\%$ . Assuming 95% confidence level and margin of error as 10%, the estimated sample size using the formula  $N = Z^2 (1 - \alpha/2) \times pq / d^2$  was 82.<sup>[8]</sup> Convenient sampling technique was employed.

**Study tool:** Structured, pretested validated questionnaire prepared in English translated to Tamil by expert scholar and again retranslated to English to ensure Linguistic validity.

### Methodology

Individuals attending the outpatient clinic at Government Medical College and Hospital, Omandurar Government Estate, satisfying the inclusion criteria, and consenting to participate in this study were included. Written informed consent was obtained from the participants prior to enrolment. Participants then faced an interviewer administered questionnaire under full privacy and confidentiality. The first section of the questionnaire collected the participant's particulars/information, demographic details and their symptomatology, if any, along with the medical history, alcohol consumption (AUDIT) and questions regarding the risk factors assessed.

The participant then underwent abdominal ultrasound for the identification of fatty liver based on standard criteria that encompassed ultrasound appearance of the intrahepatic vessels, liver parenchyma and diaphragm, liver brightness and the contrast between the liver and kidney. NAFLD is considered absent when the echotexture of the liver was normal. Participants were identified as having liver steatosis when there was an increase in liver echogenicity.<sup>[9]</sup> Those who were identified with NAFLD underwent blood investigations for assessing haemoglobin levels, liver function tests and renal function tests. Health education regarding healthy diet and life style changes were provided to all the participants. Those diagnosed with NAFLD were referred to General Medicine Department for further management.

An operational definition that was used in this study – “Any grade of fatty liver in the absence of alcohol intake or a person who has an AUDIT score of less than 8”.<sup>[5-9]</sup>

**Data Analysis:** The data thus assimilated is maintained in a spreadsheet and is statistically analysed using SPSS Version 22 software. The p-value of <0.05 was taken as statistically significant. Continuous variables are presented in the form of

descriptive statistics (mean and standard deviation) and categorical variables in the form of frequency distributions and percentages. Bivariate analysis using Chi square test was done to assess the association between 'NAFLD' (dependent variable) and various risk factors. Multiple logistic regression analysis was done on the factors which were found to be significant in bivariate analysis and adjusted Odds ratio with 95% CI were calculated.

**Ethical Consideration:** Nil. The study was approved by Institutional Ethical Committee.

## RESULTS

A total of 350 participants were approached of which only 200 fit in the eligibility criteria. Of these, only 111 participants wished to participate. Among them, 64.9% of people were female and 35.1% of people were male.

**Table 1: Diagnostic findings of correlation between bone marrow aspirate and bone marrow biopsy (n=111).**

Factors	Categories	Number of people (n=111)	Percentage (%)
Gender distribution	Females	72	64.9
	Males	39	35.1
Age in years	18-28	18	16.2
	29-38	26	23.4
	39-48	26	23.4
	49-58	17	15.3
	More than 58	24	21.6
Location	Rural	32	28.8
	Urban	79	71.2

The distribution of the comorbidities and other assessed factors is given in Table 2. Diabetes was present in 36.9% and hypothyroidism in 17.1% of the sample.

**Table 2: Distribution of comorbidities and factors assessed among participants**

Factors assessed	Categories	Number of participants (n=111)	Percentage (%)
Physical activities*	Yes	11	9.9
	No	100	90.1
Diabetes mellitus <sup>1</sup>	Yes	41	36.9
	No	70	63.1
Hypertension <sup>1</sup>	Yes	32	28.8
	No	79	71.2
Thyroid disorders <sup>1</sup>	Hyperthyroid	1	0.9
	Hypothyroid	19	17.1
	Euthyroid	91	82

<sup>1</sup> Based on the medical history as said by the participant.

\*Exercise was taken as yes to those individuals who gave history of doing moderate intensity aerobic physical activity for at least 150 minutes per week.<sup>[10]</sup>

The overall prevalence of NAFLD among the study population is 32.43% with 95% confidence interval. The association between NAFLD and the associated factors is given in [Table 3 and Table 4].

**Table 3: NAFLD Association with demographic characteristics**

Factors	Categories	Fatty liver	No fatty liver	Chi square	P-value
Gender	Female	23 (63.9%)	49 (65.3%)	0.22	0.881
	Male	13 (36.1%)	26 (34.7%)		
Age in years	18-28	0 (0%)	18 (24%)	13.57	0.009
	29-38	10 (27.8%)	16 (21.3%)		
	39-48	12 (33.3%)	14 (18.7%)		
	49-58	8 (22.2%)	9 (12%)		
	More than 58	6 (16.7%)	18 (24%)		
Diet	Mixed	18 (50%)	54 (72%)	9.846	0.007
	Predominantly Non vegetarian	10 (27.8%)	5 (6.7%)		
	Vegetarian	8 (22.2%)	16 (21.3%)		

**Table 4: NAFLD association with Hypertension and Exercise.**

Factors	Division	Fatty liver	No fatty liver	Chi square	P-value	Prevalence/Odds ratio
Hypertension	Yes	17 (47.2%)	15 (20%)	8.785	0.003	0.279 (0.118-0.664)
	No	19 (52.8%)	60 (80%)			
Exercise	Yes	1 (2.8%)	10 (13.3%)	3.036	0.081	5.38 (0.662-43.809)
	No	35 (97.2%)	65 (86.7%)			

Among the assessed actors, age, diet and hypertension demonstrated significant association with NAFLD. Among the 36 participants who have

NAFLD, routine blood investigations showed 44.1% of the participants had anaemia, while majority had normal renal and liver function tests.

**Table 5: Laboratory parameters among participants with NAFLD**

Blood investigation	Categories	Frequency
Hb#	Anaemia	16
	Not anaemic	20
RFT+	Elevated	3
	Normal	33
AST or ALT^	Elevated	3
	Normal	33

#Anaemia cut off is taken as Hemoglobin less than 12g/dL for uniformity.<sup>[11]</sup>

+Blood urea or Serum creatinine value of more than 40 mg/dL or 1.2 mg/dL, respectively.

^AST/ALT values more than 40 IU/L taken as elevated.

## DISCUSSION

This study has found a NAFLD prevalence of 32.43 % among outpatients visiting the tertiary care centre in Chennai which is consistent with other recent Indian data. Shalimar et.al (2022) had reported a pooled prevalence of 38.6% in India, which is similar to or even greater than estimates from Western Countries.<sup>[4]</sup>

Our study did note a higher prevalence compared to the 30.7% reported from a similar study carried out in a rural Haryana community; which suggests that urban areas are more likely to carry a comparatively higher disease burden. This could be due to multiple factors: sedentary lifestyle, less physical activity and increased consumption of processed foods and high-fat diets.<sup>[8]</sup>

**Age:** A statistically significant association was found between age and the presence of NAFLD ( $p= 0.009$ ). Higher proportions were noted in the age groups of 39-48 years (33.3%) and 49-58 years (22.2%). Therefore, our study reinforces that increasing age is associated with NAFLD severity. With advancing age, there is reduced physical activity and muscle mass, progressive insulin resistance, accumulation of visceral fat, more inflammation, and hormonal changes: all contributing to increased hepatic fat accumulation. These findings are consistent with a study that took into account histological evidence to compare severity of NAFLD between elderly and younger patients. They had found that there was a higher prevalence of NASH 74% v/s 56%.<sup>[12]</sup>

**Diet:** In our study, we found a significant association between non-vegetarian diet and the disease ( $p = 0.007$ ). Diet plays an important role in both the pathophysiology as well as the management of NAFLD. Systematic reviews of both observational and interventional studies highlight the importance of overall dietary patterns and adherence to a healthy, more balanced and nutritious diet being inversely associated with the development of hepatic steatosis. Meta-analyses of randomized controlled trials further show that healthier cuisines such as the Mediterranean diet emphasizing fresh, whole foods and healthy fats would improve key clinical markers,

including lipid profiles, and liver enzymes, therefore indicating its therapeutic potential in management of NAFLD. Western-style diets, consisting of more refined carbohydrates and saturated fats, are more likely to increase the risk. The type of fat also matters; mono-unsaturated and poly-unsaturated fatty acids or “healthy fats” help in decreasing liver fat, while consumption of saturated fats have been implicated in increasing the severity of NAFLD.<sup>[13,14,15]</sup>

**Hypertension:** Our study showed a statistically significant association between hypertension and NAFLD ( $p= 0.003$ , 95% CI: 0.118-0.664, OR= 0.279). The strongest evidence indicating this was a study conducted by Li et al. whose key finding suggested that hypertension did increase the risk of NAFLD with a hazard ratio of approximately 1.63.<sup>[15]</sup> Another study by Huang et al also concluded that hypertension is an independent risk factor for NAFLD, increasing its odds by approximately 1.45 times.<sup>[16]</sup> Hypertension was also reported as a significant independent predictor of NAFLD in adult Indian population by Singhai et al.<sup>[17]</sup>

Our study did not demonstrate a statistically significant association between physical activity and NAFLD ( $p= 0.081$ ). However, there is robust international evidence to confirm that regular physical activity of  $\geq 150$  minutes per week of moderate-intensity aerobic activity can improve insulin sensitivity, reduce visceral fat and inhibit hepatic lipogenesis. Thus larger studies with sufficient number of physically active participants are required to adequately assess this relationship.<sup>[18]</sup> 44.4 % ( $n=16$ ) of the NAFLD participants had anemia (Hb<12g/dL was taken as a working definition). Although no statistical testing of this association was done, this finding does warrant further research. Many studies have suggested that NAFLD is associated with altered hemoglobin levels, hepcidin dysregulation, abnormal serum ferritin values, impaired iron utilization etc. These papers suggest both iron deficiency as well as iron overload being important mechanisms in the pathophysiology of the disease.<sup>[19,20,21]</sup>

However, it may be important to note that 64.9 % of this sample was female, and given the already high prevalence of iron deficiency anemia in Indian women, it could also be a factor independent of NAFLD. Thus, we require larger prospective studies that control for gender, dietary intake of iron, laboratory parameters such as serum ferritin, so as to further test this association.

This study also confirmed the well-established fact of serum transaminases being unreliable markers for screening of NAFLD as 91.7% of NAFLD positive participants had AST and ALT values within normal physiological parameters. This finding further emphasizes the need for an ultrasound-based screening rather than biochemical marker based approaches.

**Strengths and Limitations:** Using ultrasound-based techniques that incorporate standardized echogenicity criteria for diagnosis of NAFLD is a major strength of the study. Others include, the use of a validated questionnaire that was available in both Tamil and English and the application of the AUDIT score to screen for alcohol use. This study has thus contributed to the limited literature of prevalence data of NAFLD in South Indian settings.

With that being said, there are limitations to be noted as well. First, the small sample size (n=111) and the use of convenience sampling techniques does limit the generalizability of our findings to the much broader Indian population. Second, anthropometric measurements were not taken into account, which are some of the most important modifiable NAFLD risk factors and therefore represent a major data gap. Third, although ultrasound is a good screening tool, the gold standard method for diagnosis of NAFLD is by histological confirmation; which is not economically feasible. Fourth, there is potential information bias as the comorbidities such as diabetes, hypertension and thyroid disorders were identified through self-report alone; without any laboratory confirmation.

## CONCLUSION

A significant burden of NAFLD was identified, with 32.4 % of the study population being diagnosed with the disease. In our study, people aged 39 years and above, non-vegetarians and hypertensives were found to be statistically significant associated risk factors. Most of our NAFLD-positive patients had normal liver function tests reinforcing that biochemical markers alone would be insufficient for screening; ultrasound- based techniques are also essential. We also identified a high prevalence of anaemia among the positive cases; warranting further research through adequately powered studies.

Thus, we propose urgent action in the following areas (1) To do routine screening using ultrasound in high-risk individuals (2) to advise lifestyle modifications and behavioural changes to prevent the development of disease (3) Integration into existing non-communicable disease prevention strategies.

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