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ASSESSING LIPID PROFILES AND LIVER FUNCTION IN ALCOHOLIC AND NON-ALCOHOLIC FATTY LIVER DISEASE: A COMPARATIVE PERSPECTIVE

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Abstract:
Background: The aim of the present study was to compare the lipid profile and liver function tests in individuals diagnosed with alcoholic fatty liver disease (AFLD) and non-alcoholic fatty liver disease (NAFLD).
Methods: This was a cross-sectional study conducted on 50 AFLD and 50 NAFLD patients. We measured lipid parameters and liver function tests using standard methods on Biosystem BA-400 chemistry analyzer. In addition, we have also calculated De Ritis ratio in both AFLD and NAFLD patients.
Results: Patients with AFLD showed significantly higher levels of total cholesterol (TC), triglycerides (TG), low-density lipoprotein cholesterol (LDL-C), and very low-density lipoprotein cholesterol (VLDL-C) compared to those with NAFLD. In contrast, HDL-C levels were notably lower in AFLD patients than in those with NAFLD. Additionally, AFLD patients had elevated serum levels of aspartate aminotransferase (AST), total bilirubin, and the AST/ALT ratio compared to individuals with NAFLD.
Conclusion: Atherogenic dyslipidemia and elevated AST and AST/ALT (De Ritis) ratio were more prominent in AFLD patients than in NAFLD, suggesting the De Ritis ratio as a potential marker to differentiate between the two conditions.

Keywords: Alcoholic fatty liver disease, non-alcoholic fatty liver disease, lipid profile

Introduction:

Alcoholic fatty liver disease (AFLD) is a type of liver disease that is complicated by alcohol consumption, which has a crucial role in the regulation of lipid metabolism. AFLD, even if considered less dangerous than alcoholic steatohepatitis, may increase liver morbidity and mortality. Non-alcoholic fatty liver disease (NAFLD) is the most established liver disorder that can increase morbidity and mortality due to metabolic-chronic disease, even modified by genetic and epigenetic factors. [1] NAFLD, a prevalent liver condition in developed nations, impacts approximately 30% of the population. [2] A global study found the prevalence to be around 25% [3], while in India, it is

estimated to affect 9% to 32% of the overall population, with higher rates among those dealing with obesity and diabetes. [4]

Nonalcoholic fatty liver disease is identified by a significant accumulation (ranging from 5% to 10%) of fats in the liver tissue, despite the absence of substantial long-term alcohol intake. [5] The most common ailments associated with hepatic steatosis, which is defined by the buildup of fat in the liver, are AFLD, caused by excessive alcohol intake, and NAFLD, brought on by obesity and insulin resistance. [6] AFLD and NAFLD demonstrate similar pathological developments, ranging from basic liver fat accumulation to inflammation, liver scarring, and liver cancer. Both AFLD and NAFLD frequently accompany complications outside of the liver, such as heart disease and cancer. The prognosis and likelihood of survival for individuals with AFLD and NAFLD are impacted by a variety of factors related to the diseases. [7]

Multiple research studies have shown that people with NAFLD have abnormalities in their lipid levels and liver enzymes [8-11]. In contrast, there has been limited research to investigate the involvement of lipid profiles and liver enzymes in AFLD [12, 13]. There is a lack of studies comparing lipid markers and liver enzymes in patients diagnosed with AFLD and NAFLD. Therefore, this study aims to compare the lipid profile and liver function tests in individuals diagnosed with AFLD and NAFLD.

Materials and Methods:

Study design and study participants:

This cross-sectional study was carried out at the Department of Biochemistry, Shyam Shah Medical College, REWA, MP, India over the course of one year. A total of 100 participants were included in the present study, comprising 50 individuals with non-alcoholic fatty liver disease (NAFLD) and 50 patients diagnosed with alcoholic fatty liver disease (AFLD) based on ultrasonography (USG). Patients with alcoholic and non-alcoholic fatty liver disease were recruited from the outpatient department of the medicine ward at Shyam Shah Medical College and associated Hospital in Rewa, Madhya Pradesh, India. The age of these patients ranged from 18 to 60 years old. The criteria for including patients in the study for AFLD were those who had a history of drinking alcohol above 210gm/week for males and 140gm/week for females over the past two years and had an ultrasound that showed fatty liver. NAFLD patients were included if they had no history of alcohol consumption but had an ultrasound that showed fatty liver. Patients with a history of hepatitis, diabetes, thyroid disorders, heart disease, or were taking medications that could affect Heart Rate Variability (HRV) were excluded from the study.

The study was approved by the institutional ethical committee with IEC/MC/2020/460. This study was conducted in accordance with the provisions of the Declaration of Helsinki. Following a thorough explanation of the research to every participant, they were required to give written consent. In order to protect the confidentiality of the participants' information, coding and computer recording techniques were utilized.

Anthropometric measurements:

Participants' height and weight were measured using the appropriate equipment, with minimal clothing and bare feet. Weight was measured with calibrated electronic scales, while height was determined with a portable stadiometer accurate to the nearest centimeter. BMI was calculated by dividing weight in kilograms by height in meters squared. All measurements were done by the same trained person. After a 10-minute rest, systolic and diastolic blood pressures were assessed using a mercury sphygmomanometer as per medical protocols.

Biochemical measurements:

In a sterile environment, around 5 milliliters of venous blood was taken from each patient during fasting and placed in plain tubes for the purpose of analyzing liver function and lipid levels. The plain tubes with the blood samples were then spun at 3000 revolutions per minute for 10-15 minutes to isolate the serum. Standard methods were utilized on the Biosystem BA-400 chemistry analyzer to

analyze the lipid parameters and liver function tests. Additionally, the levels of low-density lipoprotein and very low-density lipoprotein cholesterol were determined using Friedewald' s equation [14].

Statistical analysis:

The data analysis was conducted using IBM SPSS Statistics 20 (Armonk, NY, USA). The findings were displayed as mean values along with their respective standard deviations (SD). In order to compare the statistical differences in the studied parameters between NAFLD and AFLD, the student independent sample t-test was employed. A p-value lower than 0.05 was deemed to be significant.

Results:

The average age of patients with NAFLD was 43.64 years, while those with AFLD had an average age of 44.60 years. This age difference, however, was not statistically significant. The mean BMI for NAFLD patients was 27.15 kg/m², compared to 25.45 kg/m² for AFLD patients. BMI was notably higher in the NAFLD group than in the AFLD group. NAFLD patients had a SBP of 129.52 mmHg and a DBP of 83.34 mmHg. For AFLD patients, the mean systolic and diastolic pressures were 126.32 mmHg and 82.88 mmHg, respectively. Statistical analysis revealed that the differences in blood pressure between the two groups were not significant. In **Figure 1 (a)**, a comparison is presented showing the fasting blood sugar (FBS) and lipid profile of patients with NAFLD and AFLD. Patients diagnosed with AFLD displayed notably elevated levels of total cholesterol (TC), triglycerides (TG), low-density lipoprotein cholesterol (LDL-C), and very low-density lipoprotein cholesterol (VLDL-C) in comparison to those diagnosed with NAFLD ($p<0.05$ for each). Conversely, AFLD patients demonstrated a marked decrease in high-density lipoprotein cholesterol (HDL-C) levels compared to NAFLD patients ($p<0.05$). **Figure 1 (b)** represents comparison of ASL and ALT between NAFLD and AFLD. AST was significantly higher in AFLD comparison to NAFLD. Total bilirubin was significantly high in AFLD comparison to NAFLD (**Figure 1 (c)**). **Figure 1 (d)** shows no significant difference in total protein between AFLD and NAFLD. In addition, we have calculated AST/ALT ratio, which was found to be significantly higher in AFLD compared to NAFLD (1.26±0.29 vs 0.87±0.16, $p<0.05$).

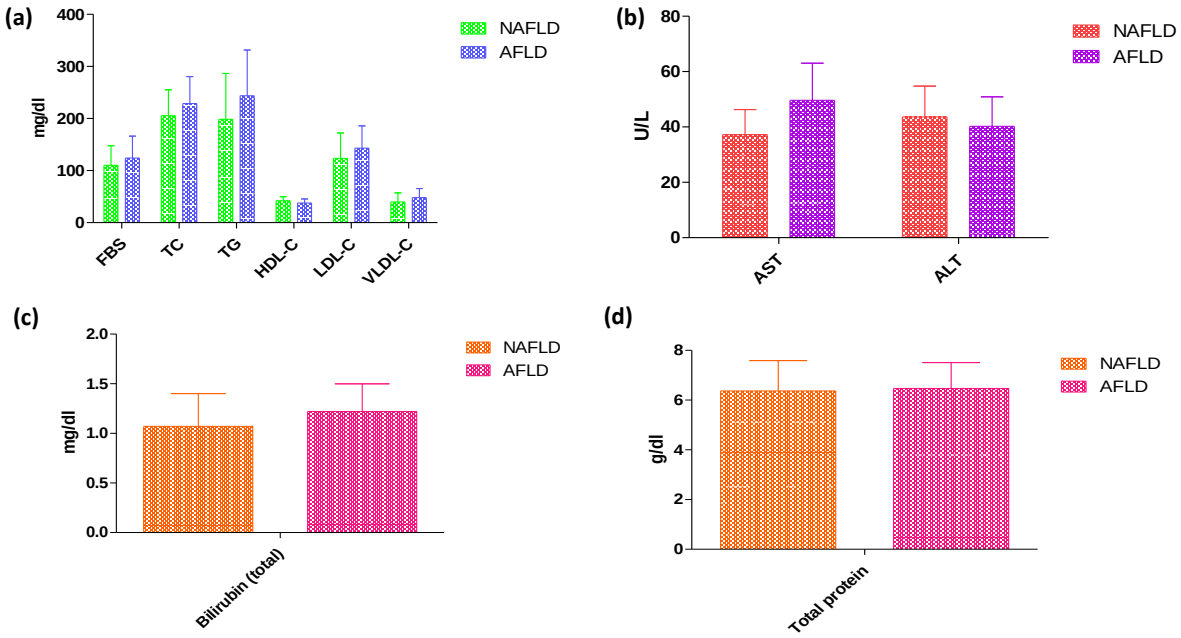

Figure 1: Comparison of biochemical parameters between NAFLD and AFLD. (a) FBS and lipid parameters. (b) AST and ALT levels. (c) bilirubin levels. (d) total protein levels.

Discussion:

ALD and NAFLD are significant global health and socioeconomic challenges. Despite having similar pathological progressions, from simple hepatic steatosis to steatohepatitis and liver cirrhosis [15], ALD and NAFLD differ in several aspects, including clinical features and patient outcomes. NAFLD is becoming increasingly prevalent as the predominant chronic liver ailment in the Western world. It is linked with insulin resistance and often presents alongside characteristics of the metabolic syndrome. A significant comorbidity commonly observed in NAFLD patients is dyslipidemia, which involves elevated triglyceride levels, decreased high-density lipoprotein cholesterol (HDL-c), and increased levels of very low-density lipoprotein (VLDL) and low-density lipoprotein cholesterol (LDL-c). [16, 17] Recent studies suggest that certain lipid profile traits may be linked to the severity of nonalcoholic fatty liver disease (NAFLD) and the progression of nonalcoholic steatohepatitis (NASH) and liver fibrosis [18, 19]. Alcohol-induced lipo-toxicity is a growing area of interest as it plays a significant role in the development of alcohol-related steatohepatitis. The process of detoxifying alcohol in the liver results in the production of harmful metabolites and hepatic steatosis. It is widely known that alcohol causes the accumulation of free fatty acids in the liver by inhibiting mitochondrial beta-oxidation. However, research suggests that the relationship between alcohol and hepatic lipid metabolism is more intricate. Alcohol increases the liver's uptake of free fatty acids, affects different types of fatty acids differently (short-chain vs. long-chain and saturated vs. unsaturated), and may also impact other lipid classes such as lysophosphatidylcholines, which can contribute to lipo-toxicity. [20]

Upon comparing lipid parameters between AFLD and NAFLD, we observed significantly higher levels of TG, TC, LDL-C, and VLDL-C, along with significantly lower levels of HDL-C in AFLD compared to NAFLD. This suggests a more severe dyslipidemic pattern in AFLD patients. Our findings align with those of Shahi et al. [12], who reported that individuals with AFLD exhibited a lipid abnormality spectrum characterized by elevated TC, LDL-C, TG, and VLDL-C levels compared to those with NAFLD. Similarly, Israelsen et al. [21] found that HDL levels were lower in NAFLD patients compared to those with alcoholic liver disease. However, a study by Ahn et al. [22] reported mean total cholesterol levels of 183.1 ± 36.9 in AFLD and 186.3 ± 33.2 in NAFLD, showing a smaller difference between the two groups. In this study, AFLD patients exhibited higher serum levels of AST and total bilirubin compared to NAFLD patients. However, there was no significant difference in serum ALT levels or total protein levels between the two groups. Ramesh et al [13] reported higher AST and ALT both in AFLD compared to NAFLD. Similarly, Majhi et al [23] found the mean levels of AST and ALT to be 124.8 IU/L and 54.21 IU/L, respectively. Increased serum concentrations of aminotransferases are considered sensitive indicators of liver cell injury and are useful in identifying hepatocellular diseases such as hepatitis, alcoholic liver disease, and cirrhosis. [24] The De Ritis ratio, defined as the ratio between aspartate aminotransferase (AST) and alanine aminotransferase (ALT), was found to be higher in AFLD compared to NAFLD in our study, which is similar to the study conducted by Ramesh et al. [13] This elevated ratio reflects the reduced serum activity of ALT in patients with alcoholic liver disease, which may be attributed to an alcohol-related deficiency of pyridoxal 5'-phosphate. Additionally, the increase in the De Ritis ratio could result from mitochondrial damage, cell necrosis, and increased cell membrane permeability, leading to elevated serum AST levels, particularly in individuals with high alcohol consumption. [25]

Nevertheless, our study is limited by several factors. Initially, the research was carried out in a hospital environment, leading to an inherent bias in patient selection. Additionally, the study took place over a relatively brief period. Furthermore, the absence of a control group hinders our ability to examine the potential occurrence of NAFLD. Moreover, ultrasonography was used to detect NAFLD, while liver biopsy, though considered the gold standard for diagnosing fatty liver disease, was not employed due to its invasive nature, potential complications, and high cost, making it impractical for routine use in the general population. Lastly, the generalizability of the study is limited by the small sample size and the restriction of the research to a single hospital center.

Conclusion:

There was a notable presence of atherogenic dyslipidemia and elevated AST and AST/ALT ratio (De Ritis ratio) in AFLD patients compared to those with NAFLD. The De Ritis ratio is higher in AFLD, making it a potential marker for distinguishing between NAFLD and AFLD.

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