Original Article

A Multivariate Logistic Model to Predict Grades of Non-Alcoholic Fatty Liver Disease in the Pakistani Population

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Abstract

Introduction: Non-alcoholic fatty liver disease has a high global burden. The patients are usually diagnosed with abdominal ultrasound and are then graded based on the extent of the disease.

Objective: Our aim is to make a multivariate logistic model that can efficiently predict grades of NAFLD.

Methodology: An analytical cross-sectional study was conducted at the Radiology departments of Allied hospitals of Rawalpindi Medical University on a sample of 138 patients. Data was collected using a proforma. Demographic data and data of all the lab values were collected. Informed consent was taken from all the participants.

Results: The mean age of the patients was 44.14±13.193 years of which 52.2%(n=72) were males. 24% of the patients (n=33) were diabetics. Upon correlation analysis, ALT, AST, age, duration of diabetes, and BMI were found to be significantly correlated ($p<0.05$) with grades of BMI. All the correlated variables were included in the univariate and multivariate logistic regression analysis. On multivariate logistic regression analysis, BMI (AOR=5.66, $p<0.05$), Age (AOR=1.98, $p<0.05$), ALT (AOR=1.92, $p<0.05$), and AST (AOR=1.16, $p<0.05$) were found to be significant predictors of grades of NAFLD.

Conclusion: We have successfully identified the predictors of the grades of NAFLD in the Pakistani population and have proposed a model based on these factors. However, our model still needs to be clinically validated.

Keywords: Non-alcoholic Fatty Liver Disease, BMI, Diabetes Mellitus, ALT, AST

Introduction

Non-alcoholic fatty liver disease (NAFLD) is a significant global burden, with an estimated incidence of 25% worldwide (1). However, certain regions, such as Pakistan, have reported even higher prevalence rates, reaching up to 74% (2). The progressive nature of NAFLD can lead to severe complications, including nonalcoholic steatohepatitis (NASH) and hepatocellular carcinoma, which can be fatal (1).

Traditionally, the evaluation of NAFLD severity has relied on invasive procedures like liver biopsy. However, these methods have limitations. For instance, a liver biopsy may not accurately grade the extent of fibrosis and fails to differentiate between NAFLD and NASH (3). Moreover, the requirement for specialized equipment limited to hospital settings makes routine monitoring challenging for patients. Sampling errors and contraindications in certain individuals further restrict the widespread use of liver biopsy (4).

To overcome these limitations, there is a pressing need for novel non-invasive procedures that are more patient-friendly. Our study aimed to address this gap by developing a statistical model capable of predicting the grades and severity of NAFLD. By utilizing this model, the reliance on invasive procedures like liver biopsy can be significantly reduced, allowing for more accessible and efficient monitoring of the disease.

Since NAFLD is graded. into I, II, and III based on the extent of liver damage and fat accumulation implementing our predictive model at an earlier stage, such as Grade 2, could yield substantial benefits (5). Early intervention can help prevent the progression of NAFLD and associated complications, such as the development of diabetes mellitus. By identifying patients at higher risk and initiating appropriate medical interventions, adverse outcomes can be mitigated, leading to improved patient outcomes and quality of life.

In conclusion, the high incidence of NAFLD worldwide, particularly in regions like Pakistan, underscores the urgent need for effective and non-invasive methods to assess and monitor disease severity. Our study aimed to develop a statistical model that can accurately predict the grades and severity of NAFLD. By reducing the dependence on invasive procedures and enabling timely interventions, this model holds great potential to improve patient care and outcomes. Further research and validation are essential to establish the efficacy and reliability of our proposed model in real-world clinical settings.

Materials and Methods

A convenient non-probability sampling technique was used to carry out an analytical cross-sectional study on a group of 138 outpatients visiting the radiology department in allied hospitals of Rawalpindi Medical University. The subjects were made aware of their participation in this study which was conducted over a period of 3 months starting from July 1st, 2022 keeping in mind the inclusion and exclusion criteria. Informed consent was taken from the patients and data were collected using questionnaires. Anthropometric data (age, sex, weight, and BMI) were obtained. Clinical assay values like Alanine Transaminase (ALT) and Aspartate Transaminase (AST) were also recorded. The inclusion criterion was based on patients aged 18 or above who were diagnosed with Non-Alcoholic Fatty Liver Disease (NAFLD) stage I, II, or III. Patients who refused to give participation consent or who were established as being chronic alcohol abusers were

removed from the study. Patients who tested positive for any viral liver diseases were also not included. Based on remarkable anthropometric parameters and clinical assay profiles, the stage of NAFLD was recognized. SPSS Version 28 was used in this study for data analysis. The descriptive statistics (Mean and Standard Deviation) were used to summarize the collected data in all study subjects showing continuous variability. An association between diabetes and NAFLD within diagnosed patients of Rawalpindi hospitals was established using multivariate Logistic Regression. A significance level of 5% was used in all statistical analyses. The manuscript follows the STROBE guidelines for observational studies.

Results

Of the total sample size of 138 participants, with a mean age of 44.14±13.193 years, 72 (52.2%) subjects were males and 66 (47.8%) were females. 33 (24%) patients suffered from diabetes mellitus whereas 105 (76%) were non-diabetic. The majority of the patients diagnosed with NAFLD were of grade I category 66 (47.85). Grade II and grade III accounted for 53 (38.4%) and 19 (13.85%) subjects, respectively. The descriptive statistics of the study sample have been summarized in **Error! Reference source not found.** and **Error! Reference source not found.**

Table-I Demographic and clinical characteristics of the study sample

Variables	Prevalence n (%)
Mean age of patients (Mean ± SD)	44.14 +/- 13.193 years
Male patients	72 (52.2%)
Female patients	66 (47.8%)
Diabetic patients	33 (24%)

Table-II Prevalence of grades of NAFLD in the study sample

Grades of NAFLD	Prevalence (%)
Grade I	66 (47.8%)
Grade II	53 (38.4%)
Grade III	19 (13.8%)

Data are presented as frequency (%). NAFLD, non-alcoholic fatty liver disease.

To determine which variables were associated with grades of fatty liver and should be put in the multivariate model, a correlational analysis between grades of fatty liver and age, ALT, AST, ALP levels, duration of diabetes mellitus, and other variables was done. All correlations with a p-value <0.05 were considered significant. Development and progression of NAFLD correlated with age, ALT, AST, ALP levels, duration of diabetes, BMI, comorbidities, and smoking. To elucidate this; Kendall's tau correlation was used, which indicated a strong

positive correlation between grades of fatty liver and age (τ_b**=0.596, p=0.000**) in the study sample as shown in **Error! Reference source not found.**

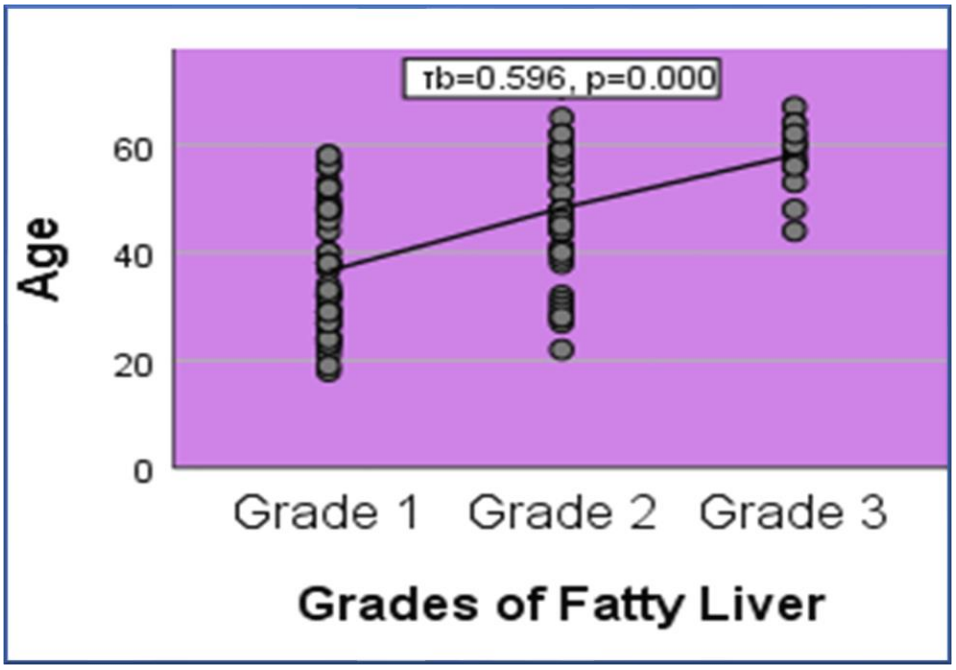

Figure 1: Scatter plot for Kendall's tau correlation between grades of fatty liver and age

Similarly; Spearman's rank correlation of grades of fatty liver with ALT (**r_s=0.646, p=0.000**) was a strong positive (**Error! Reference source not found.**); with AST(**r_s=0.773, p=0.000**) was a very strong positive (**Error! Reference source not found.**) and with ALP(**r_s=0.478, p=0.000**) was also a strong positive.

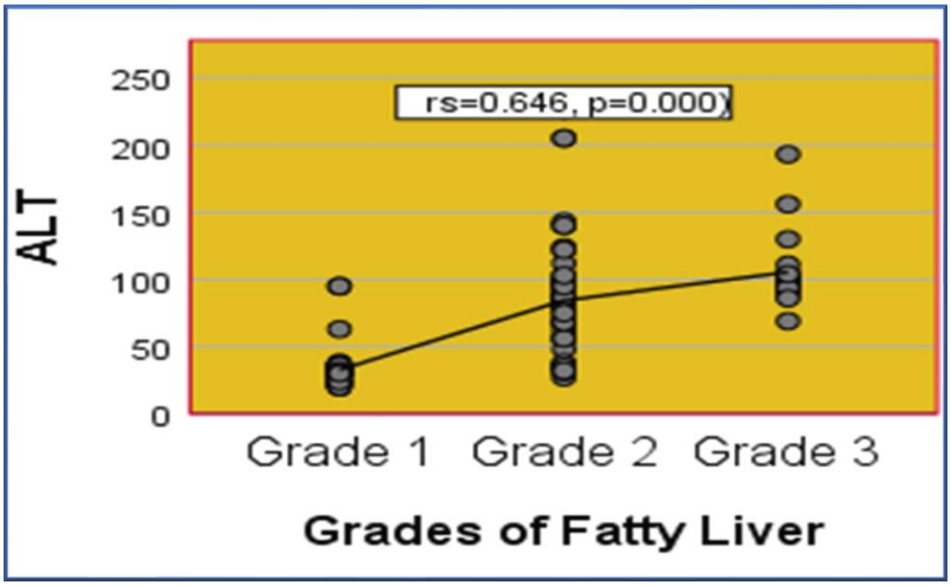

Figure 2: Scatter plot for Spearman's rank correlation between grades of fatty liver and ALT levels

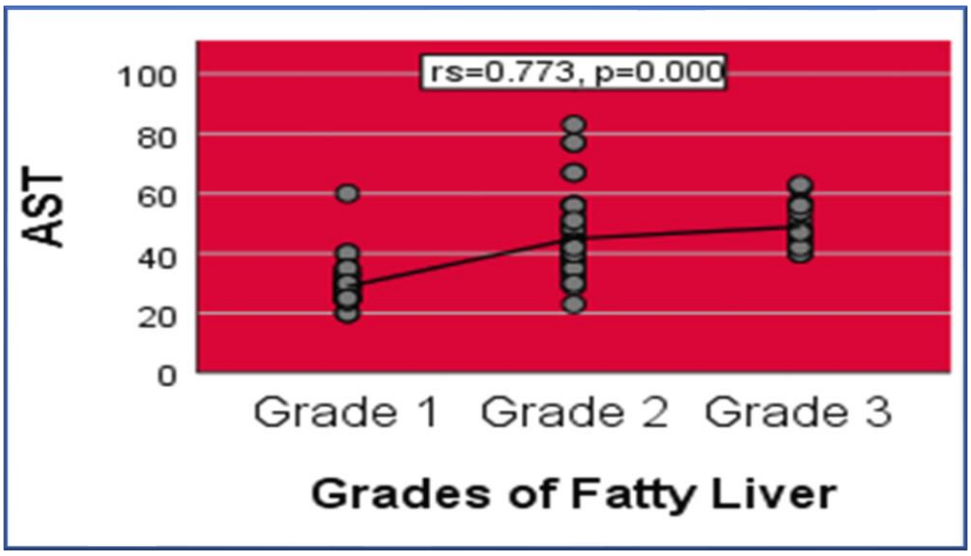

Figure 2: Scatter plot for Spearman's rank correlation between grades of fatty liver and AST levels

Sommer's delta was run to identify the association between BMI and grades of NAFLD (d=0.722, p=0.000) which was also very statistically significant. In addition, a connection between grades of NAFLD and the length of time a person has had diabetes was identified with the use of the Goodman and Kruskal gamma correlation, which was shown to be statistically significant (G=0.626, p=0.03). We used the biserial correlation co-efficient to establish the association between AST (the variable having the highest degree of association with grades of NAFLD) and dichotomous predictor variables such as gender, diabetes, and other co-morbidities, etc. because it was not possible to establish a correlation between nominal predictors and ordinal outcome variables. Thereafter, all the variables found to correlate with grades of fatty liver were used in univariate regression analysis followed by multivariate logistic regression analysis to identify multiple significant predictors of grades of NAFLD. Univariate analysis depicted that comorbidity, age, and AST levels were statistically significant (**p-value 0.01*** for all) in predicting grades of fatty liver. Furthermore, BMI and ALT levels were very highly significant (**p-value 0.000***** for both) (**Error! Reference source not found.**). However, upon adjustment, multivariate logistic regression analysis showed that only BMI, age, ALT levels, and AST levels were significant in predicting grades of fatty liver. Among these age and AST levels were moderately statistically significant with **p-values** of **0.03*** each, whereas BMI and ALT levels were very highly statistically significant with a p-value of **0.000***** as shown in **Error! Reference source not found.**.

Table-Ⅲ Univariate and multivariate logistic regression analysis of study variables

Variables	COR with p-value	AOR with p-value
BMI	7.14 (0.000***)	5.66 (0.000***)
Diabetes	9.32 (0.08)	6.15 (0.12)
Comorbidities	24.67 (0.01*)	9.32 (0.09)
Age	2.89 (0.01*)	1.98 (0.03*)
ALT	2.77 (0.000***)	1.92 (0.000***)
AST	2.45 (0.01*)	1.16 (0.03*)
Smoking	2.35 (0.09)	1.27 (0.16)

COR; crude odds ratio, AOR; adjusted odds ratio, BMI; basal metabolic rate, *ALT;* alanine aminotransferase, *AST*; aspartate aminotransferase

Discussion

The current study identified several variables that were prognostic for grades of fatty liver. We first conducted a co-relational analysis to identify variables that had a significant degree of correlation with the outcome ordinal variable i.e., Grades of NAFLD. BMI, commodities, age, ALT, and AST were crudely predictive of grades of fatty liver indicated by their COR with p-value which was 7.14 (0.000***), 24.67 (0.01*), 2.89 (0.01*), 2.77 (0.000***), 2.45 (0.01*) respectively. On adjustment BMI, age, ALT, and AST levels were incredibly significant in predicting grades of fatty liver in NAFLD patients. This was indicated by AOR with p values for BMI 5.66 (0.000***), age 1.98 (0.03*), ALT 1.92 (0.000***), and ALP 1.16 (0.03*).

Previous studies have shown that the prevalence and progression of NAFLD are associated with higher age, higher BMI, and higher levels of ALT and AST(6,7). Similar to our study, previous studies also show that there exists a significant correlation between grades of NAFLD and age(8), AST(9)(10)(11), and ALT(10–12). Our study, in contrast to previous research (13,14), did not find a significant correlation between diabetes mellitus and the grades of NAFLD. This discrepancy may be attributed to the limitations of our study, particularly the small sample size. It is essential to acknowledge the need for further investigation with larger sample sizes to obtain more reliable and representative results.

Traditionally, liver biopsy has been considered a gold standard and reference method for grading NAFLD. However, its invasive nature renders it less than ideal as an initial approach (15). In light of this, our study proposes a non-invasive predictive model as a first-line assessment method. Similar models have been developed and validated for predicting other medical conditions such as obstructive coronary disease

(16) and preeclampsia (17). These studies demonstrate the feasibility of developing practical predictive models similar to the focus of our research.

Implementing our proposed model in clinical settings can offer valuable assistance in evaluating the severity of NAFLD by utilizing factors commonly assessed in routine examinations. This approach holds particular relevance for a country like Pakistan, where determining the prevalence of NAFLD is not only highly beneficial but also cost-effective. The implementation of our model could potentially reduce the number of ultrasound examinations currently performed for disease monitoring, leading to fewer hospital admissions.

NAFLD is characterized by the increased accumulation of lipids in hepatocytes, which can occur due to either increased production or reduced clearance of fatty acids from the liver (18). This lipid accumulation leads to the development of steatosis, which is followed by inflammation, injury, and the potential progression to fibrosis and cirrhosis (19). There are two distinct causes of NAFLD: metabolic and non-metabolic. In cases of metabolic cause, the disease develops through two stages. Initially, the accumulation of fat in hepatocytes induces insulin resistance. In the second stage, there are cellular and molecular changes involving oxidative stress. Non-metabolic causes of NAFLD include conditions such as hepatitis C and HIV (19). Progressive NAFLD can ultimately progress to non-alcoholic steatohepatitis (NASH), which is a necro-inflammatory disease of the liver (18). NASH can further lead to liver failure and even hepatocellular carcinoma (19).

To accurately interpret and utilize our model, it is crucial to have a comprehensive understanding of the statistical framework that underlies it, similar to any other logistic model. This knowledge will enable healthcare professionals to effectively interpret the model's predictions and integrate them into clinical decision-making processes. While our study does provide valuable insights, it also has several limitations. First, the study lacks both internal and external validation, highlighting the need for further research to confirm the robustness of our findings. Moreover, the variables obtained from non-invasive approaches and hepatic enzymes serve as surrogate markers for grading NAFLD and cannot fully replace invasive methods. Additionally, the small sample size and cross-sectional design of our study prevent us from establishing causality, and the absence of patient follow-up limits the ability to draw definitive conclusions. It is also important to consider the potential impact of Berkson's bias, given that the data were collected from allied hospitals of Rawalpindi Medical University. Furthermore, as a retrospective multivariate analysis, our study calls for prospective multivariate analysis to validate and expand upon our findings.

In light of the above discussion, we strongly recommend further validation of our study through prospective longitudinal research involving larger sample sizes. Conducting large-scale studies will allow for a more comprehensive assessment of the efficiency and effectiveness of our proposed model. Taking into account the significant factors identified, the development of a scoring system to evaluate the grades of NAFLD can be a valuable direction for future research and clinical implementation.

Conclusion

In our study, we have successfully identified the predictors of grades of NAFLD in the Pakistani population. The variables such as BMI, age, ALT, and AST have shown significant correlations with the severity of NAFLD. Based on these findings, we have proposed a predictive model that can assist in evaluating the grades of NAFLD. However, it is important to note that our model still requires clinical validation to ensure its reliability and accuracy.

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