Assessment of Nonalcoholic Fatty Liver Disease in Diabetic and Prediabetic Patients Using Noninvasive Methods

Lobna Farag Eltoony, Hussein Ahmed El Amin, Lobna Abdel Wahid Ahmed, Mishael Melad Fekry*, Mohamed Abdelhakim Mahdy.
Internal Medicine Department, Faculty of Medicine, Assiut University, Assiut, Egypt.
***Corresponding Author:** Mishael Melad Fekry.
E-mail: dr_misheal@aun.edu.eg

Abstract

Background: This study intended to evaluate the validity of hepatic steatosis and fibrosis scores to predict the existence and risk stratification of liver steatosis and fibrosis amongst diabetic, prediabetic, and non-diabetic groups. These scores include fibrosis-4 index (FIB4), nonalcoholic fatty liver disease (NAFLD) fibrosis score (NFS) for prediction of fibrosis, and hepatic steatosis index (HIS) and fatty liver index (FLI) for steatosis. Transient elastography with a controlled attenuation parameter (CAP) is a widely accepted benchmark in our research. It will also assess NAFLD occurrence in prediabetic cases.

Patients and Method: This cross-sectional research was done on 150 adults who were admitted or attended the outpatient clinics of the Internal Medicine Department at Assiut University Hospital between the 1st of January 2021 and December 2021. The investigated markers' diagnostic effectiveness was evaluated using AUROC (area under the receiver operating characteristic curve).

Results: Using FIB-4 at a cutoff value of 1.45 and NFS at a cutoff value of 0.67 were good predictors for detecting liver fibrosis. However, FIB-4 was more sensitive for detecting liver fibrosis, even in prediabetic individuals. Using HSI at a cutoff value of 36 and FLI at a cutoff value of 30 were good predictors for detecting liver steatosis.

Conclusion: The diagnostic accuracy of the proposed scores had a dependable and acceptable value in recognizing individuals who might have severe fibrosis and/or steatosis using readily accessible clinical and laboratory data.

Keywords: NAFLD, Diabetic, Prediabetic, transient elastography.

Introduction

NAFLD is a significant issue in public health, with a high incidence of between 10% and 30% worldwide. It is also the third most common reason for liver transplantation and a frequent cause of chronic liver diseases **[1, 2]**. NAFLD manifestations are wide, in the absence of inflammation; it can be presented with simple steatosis, while more severely, it can be presented with liver cirrhosis, hepatocellular carcinoma (HCC), and even nonalcoholic steato-hepatitis **[3]**.

Obesity, hyperlipidemia, hypertriglyceridemia, type 2 diabetes mellitus (T2DM), and hypertension are regarded as hazard factors for NAFLD **[4].**

The gold standard method in NAFLD diagnosis is liver biopsy, although it can occasionally result in consequences such as hemorrhage, bile leak, infection, and

other potentially fatal problems **[5]**. Computed tomography, MRI, and ultrasound imaging had poor diagnostic accuracy for NAFLD. Thus, fibroscan was applied **[6]**. This imaging technique is noninvasive, easy to use, and highly accurate for measuring hepatic fat deposition and liver stiffness. The basis of noninvasive diagnostics involves scoring models, algorithms, and biochemical and clinical markers with appropriate sensitivity, specificity, and reliability **[7, 8]**.

The current study's primary objective is to assess NAFLD amongst diabetic, prediabetic, and non-diabetic individuals utilizing noninvasive techniques such as Fibroscan and routine biochemical scores.

1. Patients and Methods:

1.1 Studied Participants

The participants of this study were 150 adult Egyptians who were admitted or attended the outpatient clinic of the Internal Medicine Department, Assiut University Hospital, Egypt, from the 1st of January 2021 to December 2021. This study adhered to the guidelines of Assiut University's Ethical Committee (IRB No: 17101297). The clinical protocol has been recorded on ClinicalTrials.gov, and the registration number is NCT04553796. Every participant in this study provided written informed consent.

1.2 Eligible Criteria

Adult patients of both sexes attending inpatient and outpatient general clinics of Assiut University Hospital during the study period and who were accepted to participate in this research were enlisted in the present investigation. The exclusion criteria were cases aged less than 18 years, those with viral hepatitis infection (HCV or HBV), those at risk of developing secondary hepatic steatosis (due to excessive alcohol or medication use), those with a history of any liver disease (involving autoimmune hepatitis, primary biliary cirrhosis, drug-induced liver injury, primary sclerosing cholangitis, and -1 antitrypsin deficiency), and patients with body mass index>35 (to minimize the risk of fibroscan failure), pregnant women, patient with end-organ disease, those who refused to take part in this research were also excluded.

1.3 Sample Size Estimation:

Using the statistical software EPI info 2000, the sample size was estimated. The total sample size required was 150 subjects. The sample size was divided into three groups (diabetic, prediabetic, and non-diabetic, with 50 patients for each group).

1.4 Methodology:

The eligible individuals underwent a comprehensive process of obtaining their complete medical and personal histories, anthropometric assessment, and detailed systematic examination. Furthermore, CBC, fasting plasma glucose, glycated haemoglobin (HbA1C), liver and kidney function tests, hepatitis marker (HBs Ag, Anti-HCV Ab), and lipid profile were also done.

- **Calculation of fibrosis scores**
  - **NFS [9]:** NFS <-1.5 for low, NFS ≥-1.5 to 0.67 for intermediate, and NFS ≥0.67 for great likelihood of fibrosis.
  - **FIB-4 Score [10]**: The expected fibrosis stage is 2-3 for a score result of 1.45-3.25 and 4-6 for a score >3.25
- **Calculation** of steatosis scores:
  - **FLI [10]**: FLI <30 for low, FLI 30 to <60 for Indeterminate, and FLI ≥ 60 for high-risk steatosis.
  - **HIS [10]**: NAFLD can be ruled out if the HSI value is less than

30. HSI readings of 36 and higher suggest that a positive diagnosis of NAFLD is very likely.

- **Imaging Assessment:** The used device is fibroscan echosens at Assiut Liver Center.

1.5 Statistical Analysis:

SPSS (Statistical Package for the Social Science, version 22) was utilized throughout the data collection and analysis. The quantitative data were analyzed using the One Way ANOVA test with post-hoc analysis or the Kruskal Wallis test, and the findings were shown as the mean accompanied by the standard deviation (SD) or the median followed by the range. While the nominal data were shown as a number (n) and a percentage (%), the Chi2-test and the Fisher Exact test were utilized to contrast the data. The Pearson correlation test was carried out to analyze the degree of connection among several distinct variables. The optimal cutoff values were calculated using AUROC to verify the prediction of hepatic fibrosis or steatosis in NAFLD cases. The significance level for the P-value was set at 0.05, and the level was considered significant.

2. Results

Demographic Characteristics:

The mean age of the studied diabetic cases was more than the other two groups (53.20 ± 6.03 vs. 45.06 ± 4.56 vs. 46.52 ± 10.62, P < 0.001) in the three studied respectively, the BMI was also significantly greater among diabetic cases contrasted to the other two groups (29.32 ± 2.96 vs. 26.41 ± 3.04 vs. 27.34 ± 3.43, P < 0.001), the same was found concerning to the waist circumference (91.32 ± 9.05 vs. 84.52 ± 8.73 vs. 78.36 ± 8.89, P < 0.001) in the three studied respectively. The sex distribution and the presence of hypertension were comparable between the three studied groups with no significant variance (P=0.571 and 0.314), respectively.

Metformin was the commonest anti-diabetic medication received by the studied diabetic patient in 62.0%, 14 cases (28.0%) received Sulphonylureas, and five cases (10.0%) received DPP4 inhibitors. The median disease duration among diabetic patients was five years (ranged 1-15 years) **(Table 1)**.

Laboratory Data:

(Table 2) showed that the prothrombin time and INR were significantly shorter, and the prothrombin concentration was significantly lesser among the diabetic group than the other two groups (P < 0.001, for all). Regarding liver function, the diabetic group has significantly lower total protein and albumin levels compared to the prediabetic and control groups (P < 0.001, for both), while the diabetic group has significantly greater GGt contrasted with controls (P < 0.001). Regarding kidney function, the serum urea level was significantly greater in diabetic and prediabetic groups contrasted with the control group (P < 0.001), while the diabetic group had significantly greater serum creatinine levels in comparison to the other two groups (P=0.003). The diabetic group also had significantly higher serum cholesterol, triglyceride, and LDL levels contrasted with the other two groups (P < 0.05) and significantly lower HDL levels contrasted with the other two groups (P < 0.001). Meanwhile, CBC parameters were comparable between the three studied groups.

Fibrosis and steatosis assessment of all enrolled groups:

Risk stratification was as follows: Steatosis grade [S0 (normal), S1 (>11%), S2 (>34%), and S3 (>67%)], and Fibrosis grade [F0, 1 (normal to low-grade insignificant), F2 Low-grade fibrosis), F3 (Advanced fibrosis]. The steatosis and fibrosis grade distribution between the three studied groups is presented in

Figures 1, 2.

We contrasted the noninvasive fibrosis markers in all individuals (FIB-4 and NFS). FIB-4 was significantly greater in cases with diabetes contrasted with the other two groups (1.19 ± 0.49 vs. 1.13 ± 0.42 vs. 0.87 ± 0.4, P=0.001) in the three studied groups, respectively. Cases with T2DM also had a significantly greater mean NFS score (-1.01 ± 1.16 vs. -1.30 ± 0.95 vs. -1.54 ± 1.25, P=0.020) than that of prediabetic and control participants. Also, the steatosis assessment was analyzed by contrasting the noninvasive steatosis markers (HIS and FLI). The mean HSI was significantly greater in cases with diabetes contrasted with the other two groups (38.41 ± 5.49 vs. 35.18 ± 4.73 vs. 37.13 ± 9.57, P=0.007) in the three studied groups, respectively. However, no significant variances were noticed in the mean score of FLI among the three studied groups (P=0.151), as revealed in (**Table 3)**.

Prediction of liver fibrosis:

(Table 4) and (Figure 3) showed the predictive ability of FIB-4 and NFS to detect liver fibrosis among the studied participants using the ROC curve analysis. For FIB-4, at a cutoff value of 1.45, the areas under the ROC curves were 85.4% (95%CI: 0.762 – 0.945, P < 0.001) with a sensitivity of 83.3%, specificity of 81.9%, and accuracy of 84.7%. For NFS, at a cutoff value of –1.5, the areas under the ROC curves were 92.2% (95%CI: 0.842 – 1.0, P < 0.001) with a sensitivity of 95.8%, specificity of 59.5% and accuracy of 65.3%, while at a cutoff value of 0.67; the areas under the ROC curves was 92.2% (95%CI: 0.842 – 1.0, P < 0.001) with a sensitivity of 41.7%, specificity of 100.0% and accuracy of 90.7%.

Prediction of liver steatosis:

(Table 5) and (Figure 4) showed the predictive ability of HIS and FLI to detect liver steatosis among the studied participants using the ROC curve analysis. For HIS, at a cutoff value of 30, AUROC was 88.7% (95%CI: 0.835 – 0.939, P < 0.001) with a sensitivity of 90.9%, specificity of 43.1%, and accuracy of 74.7%, while at a cutoff value of 36 the AUROC was 88.7% (95%CI: 0.835 – 0.939, P < 0.001) with a sensitivity of 83.8%, specificity of 78.4%, and accuracy of 82.0%. For FLI, at a cutoff value of 30 the AUROC was 86.5% (95%CI: 0.800 – 0.930, P < 0.001) with a sensitivity of 80.8%, specificity of 74.5%, and accuracy of 78.7%, while at a cutoff value of 60 the AUROC was 86.5% (95%CI: 0.800 – 0.930, P < 0.001) with a sensitivity of 68.7%, specificity of 88.2%, and accuracy of 75.3%.

Tables and Figures

Table 1: Demographic characteristics of all enrolled groups

Variable name	Diabetic group (n=50)	Prediabetic group (n=50)	Control group(n=50)	P-value
Age (years)				
• Mean ± SD	53.20±6.03	45.06±4.56	46.52±10.62	
• Range	40–63	38-57	22-64	
	P^1<0.001	**P^2<0.001**	**P^3<0.001**	P^4=0.597
Sex				0.571
• Male	26 (52.0)	21 (42.0)	25 (50.0)	
• Female	24(48.0)	29 (58.0)	25 (50.0)	
BMI (kg/m^2)				
• Mean ± SD	29.32±2.96	26.41±3.04	27.34±3.43	
• Range	23.5–38.3	22.5–33.7	22.6–38.3	
	P^1<0.001	**P^2<0.001**	**P^3=0.016**	P^4=0.300
Waist circumference (cm)				
• Mean ± SD	91.32±9.05	84.52±8.73	78.36±8.89	
• Range	72–111	63-110	65-97	
	P^1<0.001	**P^2=0.001**	**P^3<0.001**	**P^4=0.002**
Hypertension	20 (40.0)	18 (36.0)	13 (26.0)	0.314
Drug used				
• Metformin	31(62.0)			
• Sulphonylureas	14 (28.0)			
• DPP4 inhibitors	5 (10.0)			
Disease duration (years)	5 (1–15)			

Data are presented as mean ± SD and range or number (percentage). Significance defined by $p < 0.05$.

P-value[1]: Comparison among all groups

P-value[2]: Comparison between Diabetic and Prediabetic group

P-value[3]: Comparison between Diabetic and control group

P-value[4]: Comparison between Prediabetic and control group

Table 2: Laboratory data among the three studied groups

Laboratory data	Diabetic group (n=50)	Prediabetic group (n=50)	Control group (n=50)	P-value
CBC parameters				
• Hemoglobin (g/dl)	12.43±1.68	12.11±1.04	12.73±1.79	0.136
• Platelets (103/ul)	277.96±68.73	274.94±50.03	291.22±71.72	0.405
• WBCs (103/ul)	7.23±1.97	6.38±1.89	6.66±1.86	0.082

Laboratory data	Diabetic group (n=50)	Prediabetic group (n=50)	Control group (n=50)	P-value
Coagulation profile				
• PT (sec)	11.23±1.16 **P1<0.001**	12.08±1.01 **P2<0.001**	11.53±1.05 **P3=0.354**	P^4=0.028
• PC (%)	99.89±14.01 **P1<0.001**	106.26±11.04 **P2=0.035**	114.55±12.79 **P3<0.001**	**P4=0.004**
• INR	0.91±0.09 **P1<0.001**	1.06±0.14 **P2<0.001**	1.02±0.17 **P3<0.001**	P^4=0.293
Liver function				
• Total bilirubin (umol/l)	6.0 (5.0 - 8.0)	7.5 (5.5 - 10.0)	9.5 (4.4 – 10.8)	0.152
• Direct bilirubin (umol/l)	1.6 (1.0-2.0)	1.6 (1.2-2.5)	1.2 (0.3-2.3)	0.134
• Total protein (ng/l)	65.94±5.54 **P1<0.001**	69.74±4.23 **P2=0.002**	75.07±6.32 **P3<0.001**	**P4<0.001**
• Albumin (g/l)	37.10 ± 3.56 **P1<0.001**	40.84±4.00 **P2<0.001**	41.54±4.14 **P3<0.001**	**P4=0.644**
• AST (U/L)	26 (21–31)	24 (19–30)	23 (19–30)	0.439
• ALT (U/L)	28 (22–35)	25 (17–39)	24 (16–30)	0.059
• AST/ALT ratio	0.86 (0.73-1.33)	0.86 (0.76-1.09)	0.87 (0.76-1.65)	0.453
• ALP (U/L)	79 (62–90)	70 (64–85)	80 (68–96)	0.197
• GGt (U/L)	35.34±11.28 **P^1<0.001**	33.72±11.41 P^2=0.694	26.16±6.17 **P^3<0.001**	**P^4=0.001**
Kidney function				
• Urea (mmol/L)	6.86±7.42 **P^1<0.001**	8.36±3.05 P^2=0.251	4.06±1.31 **P^3=0.009**	**P^4<0.001**
• Creatinine (mmol/L)	91.18±43.37 **P^1=0.003**	72.08±17.69 **P^2=0.003**	76.76±17.02 **P^3=0.035**	P^4=0.696
Lipid profile				
• Cholesterol (mg/dl)	192.22±46.81 **P^1=0.008**	170.36±40.45 **P^2=0.028**	167.94±38.75 **P^3=0.013**	P^4=0.956
• Triglyceride (mg/dl)	240.08±87.56 **P^1<0.001**	197.02±60.81 **P^2=0.005**	132.30±50.89 **P^3<0.001**	**P^4<0.001**
• HDL (mg/dl)	37.22±4.64 **P^1<0.001**	45.20±8.49 **P^2<0.001**	46.53±13.99 **P^3<0.001**	P^4=0.778
• LDL (mg/dl)	141.03±33.97 **P^1<0.001**	80.15±25.78 **P^2<0.001**	69.63±22.67 **P^3<0.001**	P^4=0.146
• HBA1c (%)	8.18±1.39 **P^1<0.001**	5.93±0.17 **P^2<0.001**	4.61±0.39 **P^3<0.001**	**P^4<0.001**

PT: Prothrombin time; PC: Prothrombin concentration; INR: international normalized ratio; ALT: Alanine transaminase; AST: aspartate aminotransferase; ALP: alkaline phosphatase; GGt: Gamma-GlutamylTransferase; HDL: high-density lipoprotein; LDL: low-density lipoprotein; HBA1c: hemoglobinA1c.

Table 3: Non-invasive fibrosis and steatosis assessment of all enrolled groups

Fibrosis assessment	Diabetic group (n=50)	Prediabetic group (n=50)	Control group(n=50)	P-value
FIB-4	1.19±0.49	1.13±0.42	0.87±0.46	
• P-value	**P^1=0.001**	P^2=0.787	**P^3=0.002**	**P^4=0.015**
NFS	-1.01±1.16	-1.30±0.95	-1.54±1.25	
• P-value	**P^1=0.020**	P^2=0.070	**P^3=0.014**	P^4=0.102
HSI	38.41±5.49	35.18±4.73	37.13±9.57	
• P-value	**P^1=0.007**	**P^2=0.001**	P^3=0.051	P^4=0.338
FLI	52.72±23.91	43.22±25.64	46.12±22.99	0.151

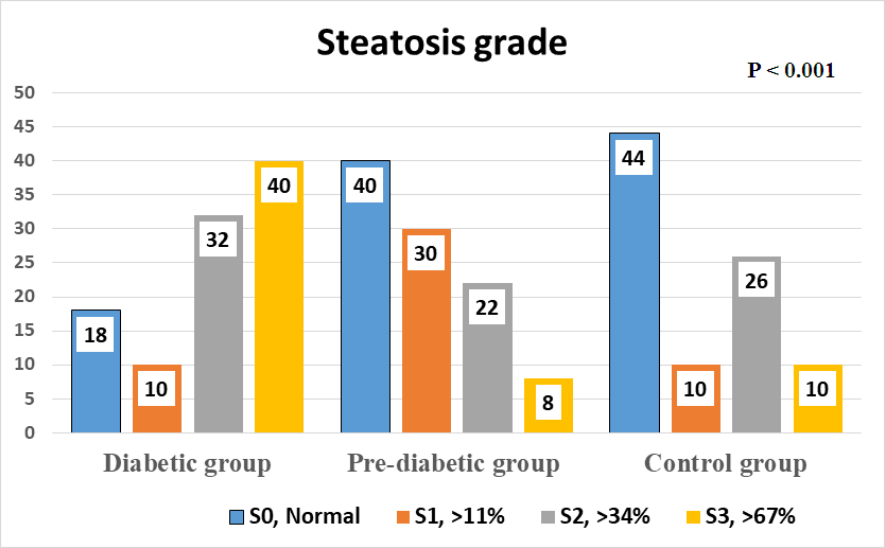

Figure 1 The distribution of steatosis grade between the three studied groups.

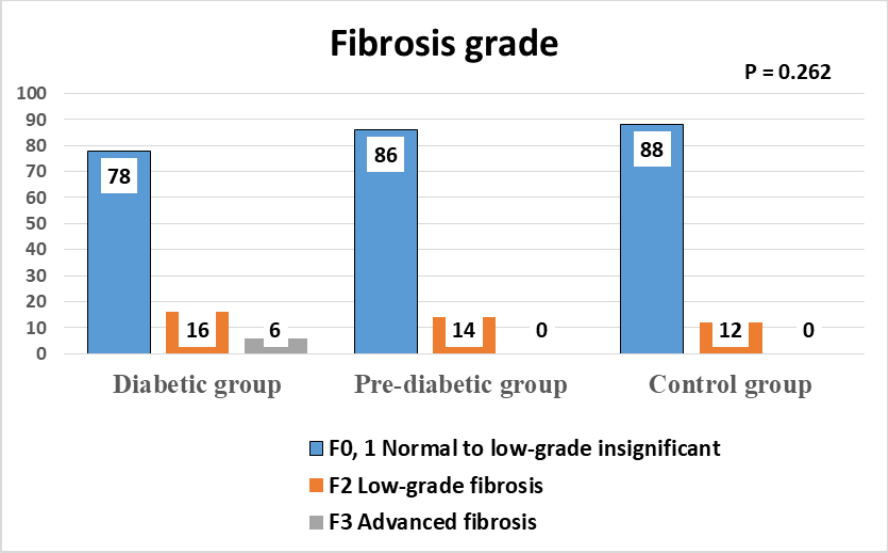

Figure 2: The distribution of fibrosis grade between the three studied groups.

Table 4: The best sensitivity and specificity for liver fibrosis detection.

	Cut off	95%CI	Sensitivity	Specificity	PPV	NPV	Accuracy	AUC	P-value
FIB-4	1.45	0.762-0.945	83.3%	81.9%	51.3%	96.4%	84.7%	0.854	**<0.001**
NFS	- 1.5	0.842–1.0	95.8%	59.5%	31.1%	98.7%	65.3%	0.922	**<0.001**
NFS	0.67	0.842–1.0	41.7%	100.0%	100.0%	90.0%	90.7%	0.922	**<0.001**

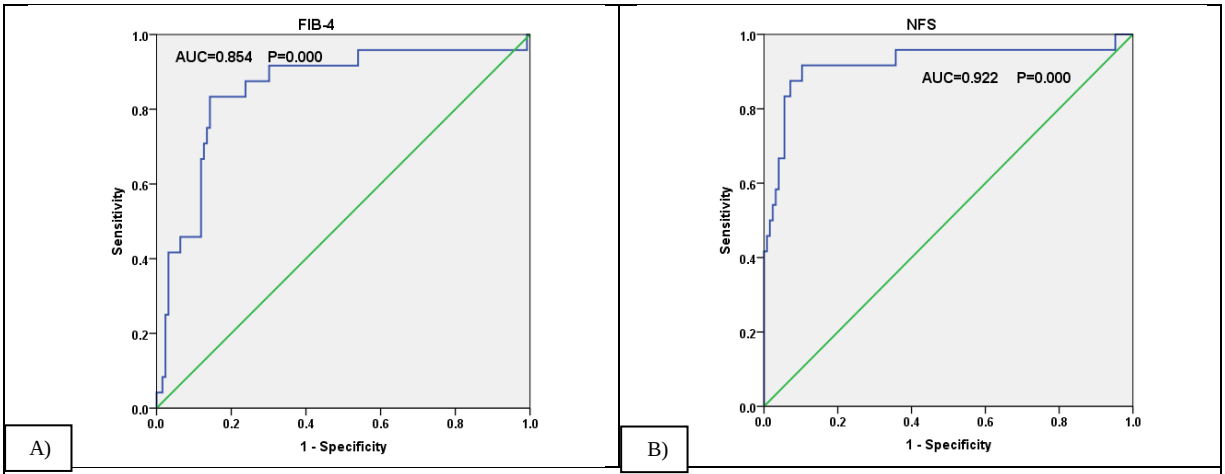

Figure 3 ROC curves for liver fibrosis detection in cases with NAFLD. A) FIB-4, B) NFS

Table 5 The best sensitivity and specificity for liver steatosis detection.

	Cut off	95%CI	Sensitivity	Specificity	PPV	NPV	Accuracy	AUC	P-value
HIS	30	0.835–0.939	90.9%	43.1%	75.6%	70.9%	74.7%	0.887	**<0.001**
HIS	36	0.835–0.939	83.8%	78.4%	88.3%	71.4%	82.0%	0.887	**<0.001**
FLI	30	0.800–0.930	80.8%	74.5%	86.0%	66.7%	78.7%	0.865	**<0.001**
FLI	60	0.800–0.930	68.7%	88.2%	91.9%	59.2%	75.3%	0.865	**<0.001**

FLI: Fatty liver index

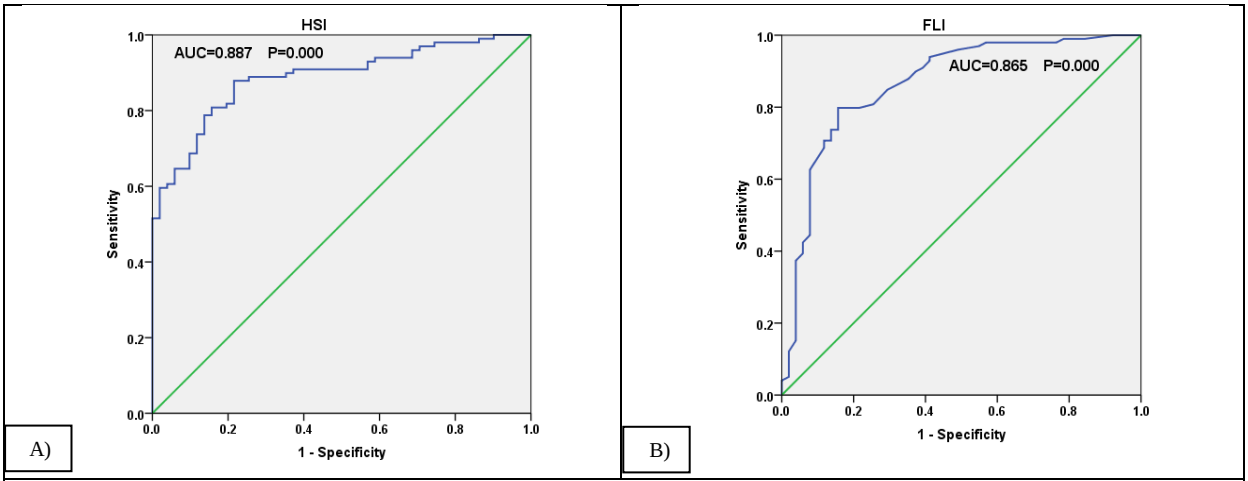

Figure 4ROC curves for liver steatosis detection in cases with NAFLD. A) HIS, B) FLI

4. Discussion

In the present research, we sought to assess the use of noninvasive scores in identifying and risk-stratifying liver fibrosis and steatosis.

The assessment of fibrosis and steatosis among enrolled participants using fibroscan showed that diabetic cases had significantly greater mean fibrosis readings than that of the prediabetic and control groups. Furthermore, the prediabetic group had substantially greater fibrosis reading than the control group. Regarding steatosis, diabetic cases had significantly greater mean steatosis results than the other two groups, while no significant variance was observed amongst the prediabetic and control groups.

We observed that FIB-4 was significantly greater among diabetic and prediabetic patients than controls. At the same time, NFS was markedly greater in diabetic cases than controls, with no significant variance among diabetic and prediabetic cases. This finding indicates that FIB-4 was more sensitive for monitoring liver fibrosis, even in prediabetic individuals.

Also, we observed that HSI was significantly greater in diabetic cases contrasted with prediabetic patients (P=0.001) and contrasted with the control group with borderline significance (P=0.051). Meanwhile, no significant variances were noticed in the mean score of FLI among the three studied groups; however, its mean was higher among diabetic patients.

At the published cutoffs FIB-4 **[10]** and NFS **[11],** we evaluate their predictive ability to detect liver fibrosis using the ROC curve analysis. We observed that for FIB-4 at a cutoff value of 1.45, the AUROC was 85.4% (95%CI: 0.762 – 0.945, P < 0.001) with a sensitivity of 83.3%, specificity of 81.9%, for NFS at a cutoff value of –1.5; the AUROC was 92.2% (95%CI: 0.842 – 1.0, P < 0.001) with a sensitivity of 95.8%, specificity of 59.5% and accuracy of 65.3%, while at a cutoff value of 0.67; the areas under the ROC curves was 92.2% (95%CI: 0.842 – 1.0, P < 0.001) with a sensitivity of 41.7%, specificity of 100.0% and accuracy of 90.7%. Based on this finding, we could be contributing that using FIB-4 at a cutoff value of 1.45 and NFS at a cutoff value of 0.67 were good predictors for detecting liver fibrosis.

According to several comparative investigations, the FIB-4 diagnostic panel holds the highest promise for differentiating steatosis from NASH. In 576 Japanese patients with biopsy-proven NAFLD, the AUROC for the FIB-4 index was 0.87, which is superior to other scoring systems (APRI, NFS, AP index, AAR, BARD score, 0.86, 0.82, 0.81, and 0.76, respectively) for discriminating between mild and advanced and fibrosis **[12]**. A research of 165 Caucasian NAFLD patients, compared to the other panels, produced the highest AUROC score for FIB 4 (0.96) **[13]**. Despite having a lower AUROC value of 0.80, different research also found that the most accurate predictor method for advanced fibrosis is the FIB-4 test **[14]**.In a cohort of 228 Latino patients, the inferior diagnostic value FIB-4 among noninvasive evaluation techniques was recorded, with an AUROC score of 0.74 **[15]**.

Most research has proven the NFS to be accurate. Twelve papers were included in a current meta-analysis; the determination of advanced fibrosis had a

summary AUC of 0.85 **[16]**. 79% of cases did not get a liver biopsy in a test of reliability for a Chinese population, and the NPV was 91%. In a sample of 267 patients, Demir et al. achieved the greatest AUROC (0.96) for NFS **[17].**

We evaluate the predictive ability of HSI [18] and FLI [19] to detect liver steatosis using the ROC curve analysis. We observed that for HSI at a cutoff value of 30, the areas under the ROC curves were 88.7% (95%CI: 0.835 – 0.939, P < 0.001) with a sensitivity of 90.9%, specificity of 43.1%, and accuracy of 74.7%, while at a cutoff value of 36, the areas under the ROC curves was 88.7% (95%CI: 0.835 – 0.939, P < 0.001) with a sensitivity of 83.8%, specificity of 78.4%, and accuracy of 82.0%, for FLI at a cutoff value of 30; the AUROC was 86.5% (95%CI: 0.800–0.930, P 0.001) with a sensitivity of 80.8%, specificity of 74.5%, and accuracy of 78.7%, while at a cutoff value of 60; the AUROC was 86.5% (95%CI: 0.800–0.930, P < 0.001) with a sensitivity of 68.7%, specificity of 88.2%, and accuracy of 75.3%. Based on this finding, we could be contributing that using HSI at a cutoff value of 36 and FLI at a cutoff value of 30 were good predictors for detecting liver steatosis.

The identification of simple steatosis is frequently made using a FLI algorithm. It has an AUROC=0.834 for NAFLD **[20,21]**. Borman et al. suggested limited usage of FLI in obese cases with small AUROC (0.67) **[22].** Lee et al. reported that HIS has an AUROC of 0.81 for diagnosing liver steatosis **[18]**.

Conclusion

According to our results, using FIB-4 at a cutoff value of 1.45 is recommended as a good predictor for early detection of liver fibrosis, and using HSI at a cutoff value of 36 for steatosis. Using these easily applied scores in routine clinical practice could help in early detection and better management of NAFLD.

References

1. European Association for the Study of the Liver (EASL), European Association for the Study of Diabetes (EASD), European Association for the Study of Obesity (EASO). EASL-EASD-EASO Clinical Practice Guidelines for the management of non-alcoholic fatty liver disease. Diabetologia. 2016;59:1121-40.
2. Lv S, Jiang S, Liu S, Dong Q, Xin Y, Xuan S. Noninvasive quantitative detection methods of liver fat content in nonalcoholic fatty liver disease. J Clin Transl Hepatol. 2018 Jun 6;6(2):217.
3. Bazick J, Donithan M, Neuschwander-Tetri BA, Kleiner D, Brunt EM, Wilson L, et al. Clinical model for NASH and advanced ***fibrosis*** in adult patients with diabetes and NAFLD: guidelines for referral in NAFLD. Diabetes Care. 2015 Jul 1;38(7):1347-55.
4. Dharmalingam M, Yamasandhi PG. Nonalcoholic fatty liver disease and type 2 diabetes mellitus. Indian J ***Endocrinol*** Metab. 2018 May;22(3):421.
5. Esterson YB, Grimaldi GM. Radiologic imaging in nonalcoholic fatty liver disease and nonalcoholic ***steatohepatitis***. Clin Liver Dis. 2018 Feb 1;22(1):93-108.
6. De Lédinghen V, Vergniol J, Foucher J, Merrouche W, Le Bail B. Noninvasive diagnosis of liver steatosis using controlled attenuation

parameter (CAP) and transient elastography. Liver Int. 2012 Jul;32(6):911-8.

7. Castera L, Friedrich-Rust M, Loomba R. Noninvasive assessment of liver disease in patients with nonalcoholic fatty liver disease. Gastroenterology. 2019 Apr 1;156(5):1264-81.
8. Wang CC, Jhu JJ. On the Application of Clustering and Classification ***Techniques*** to Analyze Metabolic Syndrome Severity Distribution Area and Critical Factors. Int J Environ Res Public Health. 2019;16(9):1575.
9. Treeprasertsuk S, Björnsson E, Enders F, Suwanwalaikorn S, Lindor KD. ***NAFLD*** fibrosis score: a prognostic predictor for mortality and liver complications among NAFLD patients. World J Gastroenterol. 2013 Feb 2;19(8):1219.
10. Sterling RK, Lissen E, Clumeck N, Sola R, Correa MC, Montaner J, et al. Development of a simple, noninvasive index to ***predict*** significant fibrosis in patients with HIV/HCV coinfection. Hepatology. 2006 Jun 1;43(6):1317-25.
11. Angulo P, Hui JM, Marchesini G, Bugianesi E, George J, Farrell GC, et al. The NAFLD fibrosis score: a ***noninvasive*** system that identifies liver fibrosis in patients with NAFLD. Hepatology. 2007 Apr;45(4):846-54.
12. Sumida Y, Yoneda M, Hyogo H, Itoh Y, Ono M, Fujii H, et al. Validation of the FIB4 index in a Japanese nonalcoholic fatty liver disease population. BMC Gastroenterol. 2012 Dec;12:1-9.
13. Younossi ZM, Jarrar M, Nugent C, Randhawa M, Afendy M, Stepanova M, et al. A novel diagnostic biomarker panel for obesity-related ***nonalcoholic*** steatohepatitis (NASH). Obes Surg. 2008 Nov;18:1430-7.
14. Shah AG, Lydecker A, Murray K, Tetri BN, Contos MJ, Sanyal AJ, Nash Clinical Research Network. ***Comparison*** of noninvasive markers of fibrosis in patients with nonalcoholic fatty liver disease. Clin Gastroenterol Hepatol. 2009 Oct 1;7(10):1104-12.
15. Pérez-Gutiérrez OZ, Hernández-Rocha C, Candia-Balboa RA, Arrese MA, Benítez C, Brizuela-Alcántara DC, et al. Validation study of systems for noninvasive diagnosis of fibrosis in nonalcoholic fatty liver disease in Latin population. Ann Hepatol. 2013 May 15;12(3):416-24.
16. Musso G, Gambino R, Cassader M, Pagano G. Meta-analysis: natural history of nonalcoholic fatty liver disease (NAFLD) and diagnostic accuracy of noninvasive tests for liver disease severity. Ann Med. 2011 Dec 1;43(8):617-49.
17. Demir M, Lang S, Schlattjan M, Drebber U, Wedemeyer I, Nierhoff D, et al. NIKEI: a new inexpensive and noninvasive scoring system to exclude advanced fibrosis in patients with NAFLD. PLoS One. 2013 Mar 26;8(3)
18. Lee JH, Kim D, Kim HJ, Lee CH, Yang JI, Kim W, et al. Hepatic steatosis ***index***: a simple screening tool reflecting nonalcoholic fatty liver disease. Dig Liver Dis. 2010 Jul 1;42(7):503-8.
19. Bedogni G, Bellentani S, Miglioli L, Masutti F, Passalacqua M, Castiglione A, et al. The Fatty Liver Index: a ***simple*** and accurate predictor of hepatic steatosis in the general

population. BMC Gastroenterol. 2006 Dec;6(1):1-7.

20. Huang X, Xu M, Chen Y, Peng K, Huang Y, Wang P, et al. Validation of the fatty liver index for nonalcoholic fatty liver disease in middle-aged and elderly Chinese. Medicine (Baltimore). 2015 Oct;94(40).
21. Khang AR, Lee HW, Yi D, Kang YH, Son SM. The fatty liver index, a simple and useful predictor of metabolic syndrome: analysis of the Korea National Health and Nutrition Examination Survey 2010–2011. Diabetes Metab Syndr Obes. 2019 Jan 24:181-90.
22. Borman MA, Ladak F, Crotty P, Pollett A, Kirsch R, Pomier-Layrargues G, et al. The Fatty Liver Index has limited utility for the detection and quantification of hepatic steatosis in obese patients. Hepatol Int. 2013 Jun.,7:592-9.