The value of noninvasive scoring systems for the diagnosis of advanced fibrosis in Egyptian patients with nonalcoholic fatty liver disease

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Background and objectives
Nonalcoholic fatty liver disease (NAFLD) is a common chronic liver disease that includes a spectrum of liver diseases ranging from simple steatosis to steatohepatitis, fibrosis, and cirrhosis. Liver biopsy is the current gold standard for the assessment of fibrosis in patients with NAFLD. However, it is an invasive procedure and not free from complications. We aimed to analyze the diagnostic performance of simple noninvasive scoring systems for the detection of fibrosis in Egyptian patients with NAFLD.

Patients and methods
Seventy-six patients with biopsy-proven NAFLD were included in the study. Noninvasive scoring systems included AST/ALT ratio (AAR), APRI score, BARD score, FIB-4 score, and NAFLD fibrosis score (NFS). Patients were classified into two groups according to the grade of fibrosis in liver biopsy. Group 1 included 57 patients with no or mild fibrosis (stage 0–2) and group 2 included 19 patients with advanced fibrosis (stage 3–4). The sensitivity, specificity, positive predictive values, negative predictive values, and diagnostic accuracy for relevant cut-offs and area under receiver operating characteristic curves were determined.

Results
The area under receiver operating characteristic curves for advanced fibrosis were 0.936 for the FIB-4 score, 0.916 for NFS, 0.907 for the APRI score, 0.840 for AAR, and 0.556 for the BARD score. NFS and the FIB-4 score showed the best diagnostic accuracy (92.6 and 89.7%, respectively), followed by the APRI score (75%), AAR (40.8%), and the BARD score (39.5%).

Conclusion
FIB-4 and NFS can be used reliably to diagnose or exclude advanced fibrosis in NAFLD and thus reduce the burden of liver biopsies.

Keywords:
biopsy, nonalcoholic fatty liver disease, noninvasive scoring systems

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Introduction

Nonalcoholic fatty liver disease (NAFLD) is one of the most common causes of liver disease, with prevalence rates reaching up to 69.5% in some populations [1–3]. It is typically associated with type 2 diabetes mellitus and obesity and can be considered as the hepatic manifestation of the metabolic syndrome [4]. NAFLD is defined by excessive fat accumulation (steatosis) in the liver (>5% of hepatocytes) in the setting of no or little alcohol use. A subgroup of NAFLD patients has liver cell injury and inflammation in addition to excessive fat (steatohepatitis; NASH). Although simple steatosis does not correlate with increased short-term morbidity or mortality, progression of this condition to that of NASH markedly increases the risks of progressive fibrosis, cirrhosis, and hepatocellular carcinoma [5–8]. The presence and severity of fibrosis dictates both overall and liver-related mortality in patients with NAFLD [9]. The identification of the minority of patients with fibrosis among those with NAFLD is critically important for prognosis and, therefore, for the selection of patients who are candidates for existing and emerging therapeutic interventions, aiming at reversing or preventing progression of fibrosis. Furthermore, the identification of the subset of patients who have developed cirrhosis has clear importance for prophylaxis against variceal bleeding, surveillance for hepatocellular cancer, and the timing of transplantation.

Currently, liver biopsy is considered the gold standard for the assessment of fibrosis in patients with NAFLD [10,11]. However, liver biopsy is invasive and is limited by sampling error, diagnostic accuracy, and risks to the patients [12,13]. In addition, up to 90% of NAFLD patients have a benign form of the disease not requiring biopsy [14]. Moreover, given the high prevalence NAFLD in the population, the use of liver biopsy in their investigation is both practically and financially impractical [10]. Therefore, noninvasive tests that can reliably diagnose or exclude advanced fibrosis would be clinically beneficial to reduce the

need for liver biopsy. Several clinical scoring systems based on simple clinical or laboratory indices have been proposed to identify advanced fibrosis in patients with NAFLD and other liver diseases [15–20]. The aim of this study was to compare the diagnostic performance of a number of simple noninvasive scoring systems in identifying advanced fibrosis in a cohort of Egyptian patients with biopsy-proven NAFLD.

Patients and methods

Our study included 76 patients who were recruited from Kasr Al Aini hospital, Internal Medicine, and diabetes outpatient clinics. Eligible patients were older than 18 years of age and were diagnosed with liver steatosis on abdominal ultrasound (US) according to previously described criteria for the diagnosis and grading of fatty liver by ultrasound. Each patient underwent a liver US performed by the same operator using a Toshiba Aplio xv scanner equipped with a broad band 2.5–5 MHz curved-array probe to assess the presence of liver steatosis (bright liver), which was defined and graded as follows:

(1) A diffuse hyperechoic echo texture (bright liver),
(2) Increased liver echo texture compared with the kidney,
(3) Vascular blurring, and
(4) Deep attenuation [21].

Patients were subjected to a full assessment of medical history and clinical examination including measurement of blood pressure and anthropometric measurements in the form of height, weight, and waist circumference. Waist circumference (cm) was measured at the midpoint between the lower costal edge and the upper iliac crest following a normal expiration. BMI was calculated as weight (kg)/height (m^2).

Laboratory studies included complete blood count, liver function tests (aspartate aminotransferase (AST), alanine aminotransferase (ALT), γ-glutamyl transpeptidase, alkaline phosphatase, albumin, lipid profile, including total cholesterol, triglyceride, high-density lipoprotein cholesterol, and low-density lipoprotein cholesterol, fasting plasma glucose, hepatitis B surface antigen, and antihepatitis C virus antibody. Clinical and laboratory data were collected at the time of liver biopsy. Patients were excluded from the study if they had a history of or showed clinical, laboratory, or histological evidence suggesting liver diseases of other etiologies, including viral hepatitis, autoimmune hepatitis, drug-induced liver disease, primary biliary cirrhosis, biliary obstruction, hemochromatosis, Wilson's disease, or a-1-antitrypsin deficiency, or had evidence of alcohol intake of more than three drinks of any alcoholic beverage per week. The diagnoses of type 2 diabetes mellitus were recorded for any patient taking oral hypoglycemic agents or insulin or those with fasting plasma glucose of at least 126 mg/dl. Hypertension was defined as a systolic blood pressure of at least 140 mmHg or a diastolic blood pressure of at least 90 mmHg measured on two separate occasions or by the use of antihypertensive agents.

Noninvasive markers of fibrosis

The noninvasive markers used to assess fibrosis were the AST/ALT ratio (AAR), APRI score, BARD score, FIB-4 score, and NAFLD fibrosis score (NFS). These were calculated using the following equations:

(1) $\text{AAR} = \dfrac{\text{AST (IU/1)}}{\text{ALT (IU/1)}}$. [15].

(2) $\text{APRI} = \dfrac{\{\text{AST (IU/l)}/\text{upper normal value (IU/l)}\}}{\text{platelet count } (\times 10^9/\text{l})} \times 100.$ The upper normal value acccording to our lab was 37 (IU/l) [16].

(3) BARD score = sum obtained from three variables (BMI ≥ 28 = 1 point, AAR ≥ 0.8 = 2 points, diabetes = 1 point) (scale 0–4) [17].

(4) $\text{FIB-4 score} = \dfrac{\text{age} \times \text{AST (IU/l)}}{\text{platelet count } (\times 10^9/\text{l})} \times \sqrt{\text{ALT (IU/l)}}.$ [18].

(5) NFS = −1.675 + 0.037 × age (years) + 0.094 BMI (kg/m^2) + 1.13 × impaired glucose tolerance/diabetes (yes = 1, no = 0) + 0.99´AST/ALT ratio−0.013 × platelets ($\times 10^9$/l)−0.66 × albumin (g/dl) [19]

The previously published cut-off values were used for the diagnosis of advanced fibrosis: AAR ≥ 0.8, APRI ≥ 1, BARD ≥ 2, and FIB-4 have two cut-offs: lower cut-off less than 1.3 to exclude advanced fibrosis and higher cut-off of more than 2.67 to diagnose advanced fibrosis, and NFS has two cut-offs: lower cut-off less than −1.455 to exclude advanced fibrosis and higher cut-off of more than 0.676 to diagnose advanced fibrosis [15–20].

Liver biopsy

All patients enrolled in this study underwent a percutaneous liver biopsy under ultrasonic guidance according to the guidelines of liver biopsy for patients with NAFLD by the American association for study of liver diseases [22].

Biopsies were stained with hematoxylin and eosin and Massons trichrome stain, and were examined under a light microscope for histopathological diagnosis and scoring using the NASH Clinical Research Network scoring system for NAFLD, which includes both the NAFLD activity score (NAS) and the fibrosis score [23]. This scoring system addresses the full spectrum of lesions of NAFLD (steatosis, lobular inflammation, and ballooning) and allows a diagnostic categorization into NASH, borderline NASH, or no NASH. Patients with NAS equal to 5–8 are diagnosed with NASH, those with NAS equal to 3–4 are diagnosed with border line (possible) NASH, and those with NAS equal to 0–2 are diagnosed as not having NASH. The stage of fibrosis is scored separately on the basis of a five-point scale, briefly, stage 0 is equal to absence of fibrosis; stage 1 is subdivided as follows: 1a (mild, zone 3, perisinusoidal), 1b (moderate, zone 3, perisinusoidal), and 1c (portal/periportal) fibrosis; stage 2 is equal to perisinusoidal and portal/periportal fibrosis; stage 3 is equal to septal or bridging fibrosis; and stage 4 is equal to cirrhosis. Stages 3 and 4 fibrosis are classified as advanced fibrosis and stages 0, 1, and 2 as nonadvanced fibrosis. According to the grade of fibrosis on liver biopsy, patients were divided into two groups. Group 1 included 57 patients with no or mild fibrosis (G0, G1, and G2) and group 2 included 19 patients with advanced fibrosis and cirrhosis (G3 and G4).

The study protocol conformed to the ethical guidelines of the 1975 Declaration of Helsinki and was approved by Cairo University Internal Medicine Ethics Committee. Written informed consents were obtained from all participants in the study.

Statistical analysis

All statistical analyses were carried out using SPSS software version 21 (SPSS Inc., Armonk, NY, USA). Continuous normally distributed variables were represented as mean ± SD. To compare the means of normally distributed variables between groups, the Student *t*-test was performed. Qualitative variables were described as numbers and percentages. The χ^2-test or the Fisher extract test was used to determine the distribution of qualitative variables between groups. The diagnostic performance of noninvasive tests was assessed by receiver operating characteristic (ROC) curves. The area under the receiver operating characteristic (AUROC) curves was used as an index to compare the accuracy of tests, with values close to 1.0 indicating high diagnostic accuracy. The sensitivity, specificity, positive predictive values (PPV), negative predictive values (NPV), and the diagnostic accuracy for relevant cut-offs were calculated. Differences were considered significant with *P* values less than 0.05.

Results

The demographic and laboratory data of all patients are shown in Table 1.

Biopsy results showed that 49 (64.4%) patients had NASH, nine (11.8%) had simple steatosis, and 18 (23.7%) had borderline NASH. In terms of fibrosis, 37 (48.7%) patients had G0, 13 (17.2%) had G1, seven (9.2%) had G2, 17 (22.4%) had G3, and two (2.6%) had G4. In patients with NASH, 30 (61%) had no or mild fibrosis, whereas 19 (39%) had advanced fibrosis. None of the patients with simple steatosis had fibrosis and only one patient with borderline NASH had mild fibrosis (Table 2).

The clinical and laboratory features as well as noninvasive fibrosis scores and results of liver biopsy

Table 1 Demographic and laboratory characteristics of all patients

Variables	Mean ± SD
Age (years)	40.9 ± 6.43
Male/female [*n* (%)]	9 (11.8)/67 (88.2)
BMI (kg/m)	34.48 ± 2.96
Waist circumference (cm)	106.97 ± 12.20
DM [*n* (%)]	21 (27.6)
HTN [*n* (%)]	12 (15.7)
ALT (IU/l)	34.09 ± 16.64
AST (IU/l)	39.39 ± 19.63
GGT (IU/l)	44.01 ± 34.43
ALP (IU/l)	80.08 ± 39.59
ALB (g/l)	4.18 ± 0.36
PLT ($\times 10^9$/l)	242.04 ± 46.05
FBG(mg/dl)	117.14 ± 27.81
TC (mmol/l)	207.41 ± 36.58
LDL-C (mmol/l)	106.99 ± 24.10
HDL-C (mmol/l)	46.99 ± 13.27
TG (mmol/l)	164.57 ± 49.37
AAR	1.25 ± 0.59
BARD score	2.88 ± 0.99
APRI score	0.47 ± 0.30
FIB-4score	1.35 ± 0.89
NFS	−1.02 ± 1.36

ALB, albumin; ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; DM, diabetes mellitus; FPG, fasting plasma glucose; GGT, γ-glutamyl transpeptidase; HDL-C, high-density lipoprotein cholesterol; HTN, hypertension; LDL-C, low-density lipoprotein cholesterol; NFS, NAFLD fibrosis score; PLT, platelets; TC, total cholesterol; TG, triglycerides.

Table 2 Biopsy results in nonalcoholic fatty liver disease

Fibrosis score (%)	NASH (*n* = 49) (64.4%)	Borderline NASH (*n* = 18) (23.7%)	Simple steatosis (*n* = 9) (11.8%)
G0 37 (48.7)	11	17	9
G1 13 (17.2)	12	1	0
G2 7 (9.2)	7	0	0
G3 17 (22.4)	17	0	0
G4 2 (2.6%)	2	0	0

of 57 patients with no/mild fibrosis (stage 0–2) were compared with those of 19 patients with advanced fibrosis (stage 3–4) (Table 3). The mean age, BMI, and waist circumference were higher in patients with advanced fibrosis ($P < 0.001$, $P = 0.002$, and 0.004, respectively). Also, patients with advanced fibrosis had a significantly higher level of AST and significantly lower levels of platelets and serum albumin compared with patients with no or mild fibrosis ($P < 0.001$ in all).

There were significantly higher mean values of AAR, FIB-4 score, APRI score, and NFS in patients with advanced fibrosis compared with those with no or mild fibrosis ($P < 0.001$ in all). There was no significant difference between the two groups in the BARD score.

Table 3 Comparison between patients with fibrosis stages 0–2 and 3–4

Variables	No–mild fibrosis (G0–G2) (*n* = 57)	Advanced fibrosis (G3–G4) (*n* = 19)	*P*-value
Male/female [*n* (%)]	8/49 (14/86)	1/18 (5.3/94.7)	0.4
Age (years)	38.84 ± 5.91	45.42 ± 5.42	<0.001
BMI (kg/m)	33.89 ± 2.68	36.24 ± 3.14	0.002
Waist circumference (cm)	104.68 ± 11.34	113.84 ± 12.39	0.004
DM [*n* (%)]	15 (26.3)	6 (31.6)	0.8
HTN [*n* (%)]	9 (15.8)	3 (15.8)	1
ALT (IU/l)	33.35 ± 13.23	36.32 ± 24.55	0.6
AST (IU/l)	33.47 ± 15.83	57.16 ± 19.55	<0.001
GGT (IU/l)	40.39 ± 25.42	54.89 ± 52.55	0.3
ALP (IU/l)	81.9 ± 42.94	74.53 ± 27.45	0.5
ALB (g/l)	4.31 ± 0.25	3.77 ± 0.33	<0.001
Platelets (×10 /l)	257.58 ± 41.06	195.42 ± 23.09	<0.001
FBG (mg/dl)	116.74 ± 27.62	118.37 ± 29.09	0.8
TC (mmol/l)	209.93 ± 33.32	199.84 ± 45.16	0.4
HDL-C (mmol/l)	48.05 ± 13.89	43.79 ± 10.95	0.3
LDL-C (mmol/l)	108.40 ± 23.73	102.74 ± 2 5.35	0.2
TG (mmol/l)	164 ± 47.77	166 ± 55.24	0.9
AAR	1.04 ± 0.38	1.88 ± 0.66	<0.001
BARD score	2.81 ± 1.06	3.11 ± 0.74	0.2
APRI score	0.97 ± 0.58	2.47 ± 0.67	<0.001
FIB-4 score	0.37 ± 0.23	0.79 ± 0.27	<0.001
NFS	−1.62 ± 0.80	0.80 ± 1.02	<0.001

ALB, albumin; ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; DM, diabetes mellitus; FPG, fasting plasma glucose; GGT, γ-glutamyl transpeptidase; HDL-C, high-density lipoprotein cholesterol; HTN, hypertension; LDL-C, low-density lipoprotein cholesterol; NFS, NAFLD fibrosis score; PLT, platelets; TC, total cholesterol; TG, triglycerides.

The FIB-4 score showed the best AUROC curve (0.936), followed by NFS (0.916), APRI score (0.907), AAR (0.840), and BARD score (0.556) (Fig. 1). The sensitivity, specificity, PPVs, NPVs, and diagnostic accuracy of all noninvasive scores using previously published cut-offs [15–20] are shown in Table 4. Figure 2 shows the fibrosis stage according to the noninvasive scores compared with liver biopsy.

The AUROC for FIB-4 was 0.936 [95% confidence interval (CI), 0.884–0.988]. Using the high cut-off point (>2.67), the sensitivity was 63.2% and the specificity was 93%. Sixteen patients had a score of more than 2.67, 12 patients (with stage 3 or 4 fibrosis) were correctly staged (true positive), whereas four (25%) were overstaged (false positive). The PPV of this cut-off for stage 3 or 4 fibrosis was 75%. Using the low cut-off point (<1.30), for exclusion of advanced fibrosis, 52 patients had a score of less than 1.3, 49 patients (without stage 3 or 4 fibrosis) were correctly staged (true negatives), whereas only three were understaged (false negative). The NPV of this cut-off for stage 3 or 4 fibrosis was 94.2%. A total of 68 patients (89.4% of the cohort) had a FIB-4 score of less than 1.30 or more than 2.67; FIB-4 identified the absence or presence of advanced fibrosis with 89.7% accuracy in these 68 patients. Eight patients (10.6%) had FIB-4 values in the indeterminate range (1.30–2.67). For the biopsies of specimens that showed indeterminate results in the FIB-4 score, 37.5% of the samples corresponded to G1, 12.5% to G2, and 50% to G3. If liver biopsies were only performed in patients with a FIB-4 score above the low cut-off point (≥1.3), 68% of biopsies could be avoided, with 94% staged correctly (Table 5)

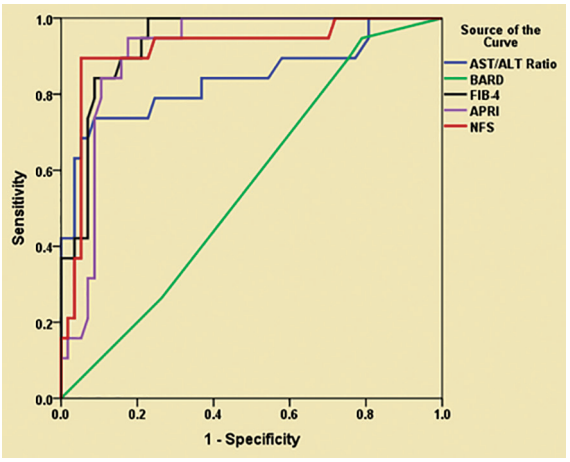
The AUROC for NFS was 0.916 (95% CI, 0.834–0.998). Using the high cut-off (>0.676), the sensitivity was 78.9% and the specificity was 94.7%. Eighteen patients had a score of more than 0.676, 15 patients (with stage 3 or 4 fibrosis) were correctly staged (true positive), whereas three were overstaged (false positive). The PPV of this cut-off for stage 3 or 4 fibrosis was 83.3%. Using the low cut-off (<−1.455), the sensitivity was 89.4% and the specificity was 84.2%. Fifty patients had a score less than −1.455, 48 (patients without stage 3 or 4 fibrosis) were correctly staged (true negative),

Table 4 Comparison of the performance of each test for the diagnosis of advanced fibrosis in patients with nonalcoholic fatty liver disease

Score	Cut-off	AUC (95% CI)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy
AAR	0.8	0.840 (0.717–0.963)	89.5	24.6	28.3	87.5	40.8
BARD score	2	0.556 (0.417–0.696)	94.7	21.1	28.6	92.3	39.5
APRI score	1	0.907 (0.839–0.974)	21.1	93.0	50.0	77.9	75.0
FIB-4 score	2.67 (1.3)	0.936 (0.884–0.988)	63.2 (84.2)	93.0 (86.9)	75.0 (66.6)	88.3 (94.2)	89.7
NFS	0.67 (−1.45)	0.916 (0.834–0.998)	78.9 (89.5)	94.7 (84.7)	83.3 (65.3)	93.1 (96.0)	92.6

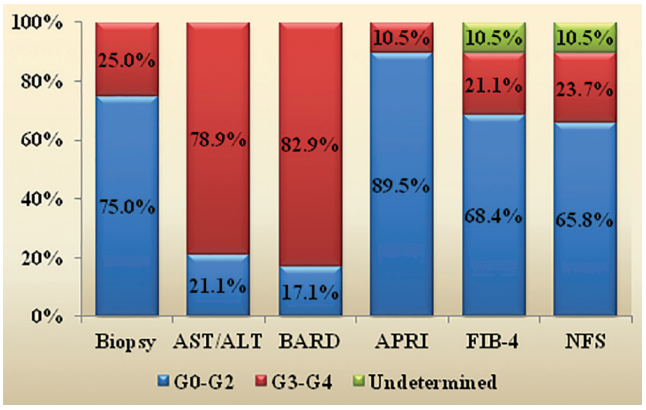
NFS, NAFLD fibrosis score; NPV, negative predictive value; PPV, positive predictive value.

Figure 1



Receiver operating characteristic curves for the noninvasive scores for the diagnosis of advanced fibrosis (stages 3–4). NFS, NAFLD fibrosis score.

Figure 2



Fibrosis stage according to the noninvasive scores compared with liver biopsy. ALT, alanine aminotransferase; AST, aspartate aminotransferase; NFS, NAFLD fibrosis score.

Table 5 Percentage of patients who can avoid liver biopsy using non invasive tests

Score	Cut-off	Patients avoiding liver biopsy (%)	False negative results (%)	Correctly staged (%)
FIB-4 score	<1.3	52/76 (68)	3/52 (5.8)	94.2
NFS	<−1.455	50/76 (66)	2/50 (4)	96
APRI score	<1	68/76 (89)	15/68 (22)	77.9
AAR	<0.8	16/76 (21)	2/16 (12.5)	87.5
BARD score	<2	13/76 (17)	1/13 (8)	92.3

NFS, NAFLD fibrosis score.

whereas only two were understaged (false negative). The NPV of this cut-off for stage 3 or 4 fibrosis was 96%. A total of 68 patients (89.4% of the cohort) had an NFS score less than −1.455 or more than 0.676; NFS identified the absence or presence of advanced fibrosis with 92.6% accuracy in these 68 patients. Eight patients (10.6%) had NFS values in the indeterminate range (−1.455 to 0.676). For the biopsies of specimens that showed indeterminate results in the NFS, 12.5% corresponded to G0, 12.5% to G1, 50% to G2, and 25% to G3. If liver biopsies were only performed in patients with an NFS score above the low cut-off point (≥−1.45), 66% of biopsies could be avoided, with 96% staged correctly (Table 5).

The AUROC for the APRI score was 0.907(95% CI, 0.839–0.974). At a cut-off of at least 1, the sensitivity was 21.1% and the specificity was 93%. Sixty eight of the 76 patients had an APRI score of less than 1. Of these, 53 patients, without advanced fibrosis, were staged correctly, whereas 15 patients were understaged. The NPV was 77.9%. Eight patients had APRI score of at least 1. Of these, four were staged correctly and four were overstaged; the PPV was 50%. The diagnostic accuracy was 75%. If liver biopsy was performed only for patients with APRI score of at least 1, 89.4% of biopsies could be avoided, with 78% staged correctly (Table 5).

The AAR had an AUROC of 0.840 (95% CI, 0. 0.717–0.963). At a cut-off of at least 0.8, the sensitivity was 89.5% and the specificity was 24.6%. Sixteen patients had a score of less than 0.8; of these, 14 patients, without advanced fibrosis, were staged correctly, whereas two patients were understaged. The NPV was 87.5%. Sixty patients had a score of at least 0.8; of these, 17 were staged correctly, whereas 43 were understaged, and the PPV was 28.3%. The diagnostic accuracy was 40.8%. If liver biopsies were only performed in patients with AAR of at least 0.8, 21% of biopsies could be avoided, with 87.5% staged correctly (Table 5).

The BARD score had an AUROC of 0.556 (95% CI, 0.417–0.696). At a cut-off of at least 2, the sensitivity was 94.7% and the specificity was 21.1%. Sixty-three patients had a score of at least 2, 18 patients with advanced fibrosis were staged correctly, and 45 were overstaged; the PPV was 28.6%. Thirteen patients had a score of less than 2, 12 patients without advanced fibrosis were staged correctly, and one was understaged. The NPV was 92.3% and the diagnostic accuracy was 39.5%. If liver biopsy was only performed in patients with BARD score of at least 2, only 17% of biopsies could be avoided, with 92% staged correctly (Table 5).

Discussion

The presence and severity of fibrosis may be the most important factors in the prognosis of NAFLD and in the prediction of the risk of progression to cirrhosis and

its complications [9]. Many noninvasive strategies have been developed to predict the stage of liver fibrosis in NAFLD. The factors found to predict the development of progressive fibrosis and cirrhosis more consistently include obesity, type 2 diabetes, age older than 45 years, an elevated AST/ALT ratio, hypertension, hyperlipidemia, and the metabolic syndrome [24,25].

In contrast with our results, Pérez-Gutiérrez *et al.* [26], found a significantly higher prevalence of diabetes mellitus in NAFLD patients with advanced fibrosis compared with patients with mild fibrosis. They also found that high BMI was not related to the degree of fibrosis, which is already known to be a risk factor for the development of fibrosis [27]. In the present study, the BMI was significantly higher in patients with advanced fibrosis (P = 0.002). In the study by Pérez-Gutiérrez *et al*. [26], only 23% of the patients had a BMI of at least 30 kg/m^2 and 1.7% had a BMI of at least 40 kg/m^2, whereas in our study, all patients had a BMI of at least 30 kg/m^2 and 6.6% had a BMI of at least 40 kg/m^2. Increased waist circumference was associated with fibrosis in a large series from the NASH Clinical Research Network [28]. In the present study, the waist circumferences was significantly higher in patients with advanced fibrosis (P = 0.004).

Also, patients with advanced fibrosis were significantly older than patients with mild fibrosis (P < 0.001), which was consistent with previous reports [26,29].

In the study by Pérez-Gutiérrez *et al.* [26], the only laboratory parameter that differed between the two groups was the platelet count. Similarly, in our study, patients with advanced fibrosis had significantly lower platelet counts (P < 0.001). This was also observed by McPherson *et al.* [29]. Also, in our study, patients with advanced fibrosis had significantly lower serum albumin (P < 0.001), which was consistent with a previous report [30].

The AST level was significantly higher in patients with advanced fibrosis in our study (P < 0.001). A previous multicenter study in children showed that serum AST level was associated with the severity of hepatic fibrosis [31]. However, other studies found no difference in the AST levels between patients with or without advanced fibrosis [26,29].

As the diagnosis of fibrosis is a major predictor of disease progression in patients with NAFLD, clinicians cannot rely solely on a single marker to identify fibrosis in these patients, particularly in view of the seriousness of this condition. Several clinical scoring systems based on simple clinical or laboratory indices have been proposed to identify advanced fibrosis in patients with NAFLD and other liver diseases. The AST/ALT ratio and the BARD score are the simplest and can be calculated easily when a patient is reviewed in the clinic. The FIB-4 and NAFLD fibrosis scores require more complex calculation, but the relevant details can easily be entered onto a predesigned Excel spreadsheet that can produce an instant result in the presence of the patient. Therefore, introduction of the use of these tests into daily practice should be relatively simple and will not result in extra costs [26]. The usefulness of the fibrosis scoring systems has been evaluated in different NAFLD populations worldwide [26,29,30,32].

In the present study, we compared the diagnostic performance of these simple noninvasive tests in identifying advanced fibrosis in a cohort of Egyptian patients with biopsy-proven NAFLD.

Elevated serum ALT and AST levels are the primary abnormality observed in patients with NAFLD and tend to be higher in patients with NASH compared with simple steatosis. However, with increasing liver fibrosis, the ALT typically decreases and the AST remains stable or increases, and as a result, the AST/ALT ratio increases [24]. Despite its simplicity, this ratio has a good NPV (93%) and can be used to rule out the presence of advanced fibrosis, as reported in previous studies [26,29]. In our study, the AST/ALT ratio showed an AUROC of 0.840 and at a cut-off 0.8, the NPV was 87.5%, whereas the PPV was 28.3% and the diagnostic accuracy was 40.8%. This low diagnostic accuracy raises a question about its reliability in the diagnoses of advanced fibrosis in our population. At a cut-off of less than 0.8, only 21% of patients could have avoided the liver biopsy, with 87.5% of these classified correctly.

The BARD score was developed by Harrison *et al.* [17], to identify patients with advanced fibrosis. A BARD score of at least 2 was associated with an odds ratio of 17 (95% CI, 9.2–31.9) and an AUROC of 0.81 for detecting advanced fibrosis [17]. However, in recent studies, the use of the BARD score has been associated with lower AUROCs, ranging from 0.65 to 0.7 [20,26,29]. Also, Sumida *et al*. [30] and Fujii *et al.* [33] reported significantly poorer applicability of the BARD score in Japanese patients with NAFLD compared with White patients. In our study, the BARD score showed the lowest AUROC (0.556). At a cut-off 2, the NPV was 92.3%, the PPV was 28.6%, and the diagnostic accuracy was 39.5%. At a cut-off less than 2, only 17% of patients could avoid the liver biopsy, with 92% of these staged correctly. These data show that the BARD score has a poor diagnostic value for advanced fibrosis in our population.

The APRI score was originally developed for assessing fibrosis in patients with liver cirrhosis [16], but it has been validated recently for NAFLD [34]. McPearson *et al.* [29] assessed the usefulness of this system and found a sensitivity of 27%, a specificity of 89%, a PPV of 37%, and an NPV of 95%. Another assessment of NAFLD, in a French cohort, showed a sensitivity of 66%, a specificity of 90%, a PPV of 72%, and an NPV of 87%; these were the highest sensitivity and PPV reported [32]. Pérez-Gutiérrez *et al.* [26] showed results similar to the values reported by McPherson *et al.* [29]. Our results showed a pattern similar to the results of McPherson *et al.* [29], but with lower sensitivity (21.1%) and NPV (77.9%) and higher specificity (93%) and PPV (50%) and a diagnostic accuracy of 75%. Using the APRI score at a cut-off of less than 1, 89.4% of patients could avoid liver biopsy, with 78% staged correctly, which means that APRI can be used to exclude advanced fibrosis in patients with NAFLD in our population.

The NFS was created by Angulo *et al.* [19] to evaluate fibrosis in fatty liver. The NFS score has two cut-offs: a score of more than 0.67 predicts the presence of advanced fibrosis, whereas a score less than −1.455 predicts the absence of advanced fibrosis. In the study by Angulo *et al.* [19], this system showed sensitivity, specificity, NPP, and PPV of 22, 100, 93, and 90%, respectively. Later studies have reported sensitivity, specificity, NPP, and PPV in the ranges of 22–78, 58–100, 92–100, and 26–81% [26,29,35]. In the present study, the NFS showed an AUROC of 0.916 and a diagnostic accuracy of 92.6%. At a cut-off less than −1.455, the sensitivity was 89.4%, the specificity was 84.2%, and the NPV was 96% and at a cut-off more than 0.67, the sensitivity was 78.9%, the specificity was 94.7%, and the PPV was 83.3%. However, a major drawback of this score is that a large percentage of patients fall in the indeterminate category and cannot be classified as having a high or a low probability of advanced fibrosis. In the present study, a total of nine patients (11.8%) had NFS values in the indeterminate range (−1.455 to 0.676). If liver biopsies were only performed in patients with NFS score of at least −1.45, 65.7% of biopsies could be avoided, with 96% staged correctly. These data provide support for the use of this marker for the detection or exclusion of advanced fibrosis in our population.

The FIB-4 index was originally developed to stage liver fibrosis in patients with hepatitis C virus infection and it has been used in NAFLD patients with promising results. The FIB-4 has two cut-offs to discriminate between the presence (2.67) and the absence (1.3) of fibrosis. Using a cut-off value of less than 1.3, the FIB-4 index has a NPV of 90–95% to rule out advanced fibrosis [20,26,29]. Using the high cut-off (>2.67), Shah *et al.* [20] found a PPV of 80%. Interestingly, when the FIB-4 index was compared with other noninvasive markers of fibrosis, including the AST/ALT ratio, BARD score, and the NFS, it had the highest AUROC for predicting advanced fibrosis (0.80–0.86) [20,29]. In the present study, the FIB-4 score showed the greatest AUROC curve (0.936). Using the high cut-off (>2.67), the FIB-4 index had a sensitivity of 63.2%, a specificity of 93%, and a PPV of 75%. Using the low cut-off (<1.3), the FIB-4 index had a sensitivity of 89%, a specificity of 86.9%, and an NPV of 94.2%. A total of 68 patients (89.4%) had a FIB-4 score of less than 1.30 or more than 2.67; FIB-4 identified the absence or presence of advanced fibrosis with 89.7% accuracy in these 68 patients. If liver biopsies were only performed in patients with FIB-4 of at least 1.3, 68.4% of biopsies could be avoided, with 94% staged correctly. Our results suggest that the FIB-4 score can be used reliably to exclude or diagnose advanced fibrosis in our population.

In the present study, in samples that produced indeterminate results in the NFS, 50% were graded G3 and in the FIB-4 score, 25% of the indeterminate group were graded G3. There is no consensus on the need for liver biopsy in this patient group; however, this group would be the most likely candidate for biopsy for adequate staging of fibrosis as part of appropriate surveillance and monitoring.

Conclusion

In the present study, the FIB-4 score and NFS showed the best AUROC curve and diagnostic accuracy and can be used reliably to diagnose or exclude advanced fibrosis. Given the large numbers of patients with NAFLD who are currently being referred to liver clinics for evaluation, the use of these noninvasive tests could markedly reduce the number of liver biopsies being performed. Liver biopsy could be considered only in patients who have a value above the lower cut-off for the chosen noninvasive score to confirm the presence of advanced fibrosis and to determine the need for long-term monitoring for cirrhosis and its complications. This would result in significant benefit to patients by directing liver biopsy to those more likely to have advanced liver disease as well as lead to cost savings. These tools are very useful in countries such as Egypt, with a high prevalence of overweight, obesity, and diabetes mellitus and a higher risk of developing fatty liver and fibrosis. This study may serve as a pilot study and studies on larger population samples are needed to validate the use of the noninvasive markers of fibrosis in our population.

Acknowledgements

Conflicts of interest

There are no conflicts ofinterest.

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