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Serum markers for predicting advanced fibrosis in patients with chronic hepatitis B and nonalcoholic fatty liver disease

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Abstract

To compare the diagnostic utility of serum markers in nonalcoholic fatty liver disease (NAFLD) patients with chronic hepatitis B (CHB).

This study enrolled 118 consecutive biopsy-proven NAFLD patients with or without CHB. Fibrosis scores of each marker were compared against histological fibrosis staging. Receiver operating characteristic curve (ROC) analysis helped assess the accuracy of each marker.

In patients with both diseases, 12.96% (7/54) had advanced fibrosis on biopsy and aspartate aminotransferase (AST) to platelet ratio index was the best performing marker for predicting advanced fibrosis. The sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and area under the ROC (95% confidence interval) for AST to platelet ratio index (APRI) were 0%, 93.62%, 0%, 86.27%, and 0.676 (0.524–0.828), respectively. The markers ranked as follows from highest to lowest with respect to their accuracy: APRI; BARD; fibrosis-4; and AST to ALT ratio. In patients without CHB, fibrosis-4 was the best performing marker for predicting advanced fibrosis. The sensitivity, specificity, PPV, NPV, and area under the ROC (95% confidence interval) for fibrosis-4 were 77.78%, 85.45%, 46.67%, 95.92%, and 0.862 (0.745–0.978), respectively.

Serum markers are less reliable in predicting advanced fibrosis in NAFLD patients with CHB; APRI is the most accurate predictor of the absence of advanced fibrosis.

Abbreviations: AAR = AST/ALT ratio, ALP = alkaline phosphatase, ALT = alanine aminotransferase, APRI = AST to platelet ratio index, AST = aspartate aminotransferase, AUROC = area under the ROC curve, BMI = body mass index, BARD = body mass index (BMI), AST to alanine aminotransferase (ALT) ratio, and diabetes Score, CHB = chronic hepatitis B, CIs = confidence intervals, CPRs = clinical prediction rules, FIB-4 = fibrosis-4, GGT = gamma-glutamyl transpeptidase, HDL–cholesterol = high-density lipoprotein-cholesterol, INR = international normalized ratio, mean ± SD = mean ± standard, NAFLD = nonalcoholic fatty liver disease, NASH = nonalcoholic steatohepatitis, NPV = negative predictive value, PPV = positive predictive value, ROC = receiver operating characteristic curve.

Keywords: nonalcoholic steatohepatitis, aspartate aminotransferase to platelet ratio index, chronic hepatitis B, fibrosis stage, non-invasive markers

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The study was performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments and obtained approval from an institutional review board (approval number: 2017-016-01).

The institutional review board approved the study with consent exemption.

The authors have no conflicts of interests to disclose.

The data that support the findings of this study are available from a third party, but restrictions apply to the availability of these data, which were used under license for the current study, and so are not publicly available. Data are available from the authors upon reasonable request and with permission of the third party.

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1. Introduction

Nonalcoholic fatty liver disease (NAFLD) is a major global cause of liver disease with the increasing obesity epidemic. NAFLD is the most common liver disease in the United States, with a prevalence rate of 27.37% in Asia.[1] Estes et al projected a modest growth in total NAFLD cases (0%–30%) between 2016 and 2030, with the highest growth in China as a result of urbanization and the lowest growth in Japan owing to shrinking population.[2] NAFLD is a clinical syndrome characterized by predominant macrovesicular steatosis of the liver in patients who have little or no alcohol consumption. Nonalcoholic steatohepatitis (NASH) is a sub-phenotype associated with disease progression resulting in fibrosis, cirrhosis, and liver cancer.[3] With emerging therapeutic interventions for NASH, early detection and non-invasive monitoring of fibrosis in patients at highest risk of disease progression are a critical step to reduce complications. Ekstedt et al observed that fibrosis stage predicts both overall and liver-specific mortality.[4] However, another study found that only significant fibrosis (grade >2) is an independent predictor of long-term mortality.[5]

The current gold standard for fibrosis staging in NAFLD patients is liver biopsy.[6] The limitations of biopsy include sampling error, intra- and inter-observer variability in interpretation of histology, high cost, and more importantly, invasive procedure associated with adverse effects.[7] Therefore, non-invasive markers are needed for the diagnosis of advanced fibrosis in NAFLD patients. Markers commonly used in clinical setting are the NAFLD fibrosis score; body mass index (BMI), aspartate aminotransferase (AST) to alanine aminotransferase (ALT) ratio (AAR), and diabetes (BARD) score; AST to alanine aminotransferase (ALT) ratio (AAR); NASH Clinical Research Network (CRN) model; fibrosis-4 (FIB-4), which combines biochemical values (platelets, ALT, and AST) and age; and AST to platelet ratio index (APRI). They have been widely validated for predicting advanced fibrosis in NAFLD.[6] However, the accuracy for predicting fibrosis with the aforementioned non-invasive markers has not been fully explored in NAFLD patients with chronic hepatitis B (CHB). Thus, we designed a cross-sectional study to enroll consecutive CHB patients who had liver biopsy-proven NAFLD. We aimed to validate the accuracy of non-invasive index scores for predicting advanced fibrosis in these patients by comparing them with paired liver biopsy findings.

2. Methods

2.1. Study design and patient selection

The present study was a cross-sectional analysis of a retrospective patient cohort with CHB and biopsy-proven NAFLD. Adult (age ≥18 years) HBV mono-infected and treatment-naïve patients who attended the liver clinic or inpatient services in Beijing Ditan Hospital from June 2015 to August 2016, with liver biopsy-proven NAFLD were eligible for the study. In our study, CHB is defined by positive HBsAg for more than 6 months. The study was performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments, and this study was approved by the Institutional Review Board of Beijing Ditan Hospital Capital medical University (number: 2017-016-01). The major exclusion criteria were as follows: presence of other hepatic diseases and secondary causes of hepatic steatosis, such as hemochromatosis, Gilbert Syndrome, Wilson disease, glycogen storage disease, cholestatic liver disease, alpha-1 antitrypsin deficiency, autoimmune hepatitis, primary biliary cholangitis, sclerosing cholangitis, liver cancer, and vascular liver disease; men and women who consumed >40 g and >20 g of alcohol per day, respectively, or other substance abuse; patients on hemolysis or immunosuppressive therapy; evidence of co-infection with hepatitis C or human immunodeficiency virus; other clinical or laboratory evidence of secondary NAFLD due to major nutritional and iatrogenic gastrointestinal disorders; clinical or laboratory evidence of decompensated liver disease (Child-Pugh score >7 points); connective tissue diseases or other significant systemic illnesses; pregnant patients; or patients without blood test results for the analysis of non-invasive markers.

2.2. Data collection, clinical assessment and methods

Using an electronic medical record system and paper charts, the following data from the clinic and inpatient services in our center were collected for analysis: demography information before liver biopsy; history of alcohol consumption, liver disease, or hepatocellular carcinoma; hepatitis B disease course, treatment history, and complications; list of medications; pertinent physical findings and BMI; and laboratory information including HBV virological markers, chemistry panel results, and imaging results. Data regarding medications and liver disease complications were also collected. Data were assessed within 60 days of liver biopsy. Assessment of laboratory values included ALT, AST, alkaline phosphatase (ALP), gamma-glutamyl transpeptidase (GGT), total bilirubin, direct bilirubin, albumin, hemoglobin A1c, fasting glucose, prothrombin time, international normalized ratio (INR), fasting lipid panel, free fatty acids, C-reactive protein, and platelet count.

Blood tests of the patients were performed in the hospital's central laboratory. Comprehensive chemistry including ALT level was tested using a Hitachi 7600 fully automatic biochemical analyzer (Wako Pure Chemical Industries, Ltd., Osaka, Japan), with the upper limit of normal of ALT set at 40 U/L. HBV serological markers were measured using chemiluminescent microparticle immunoassay (Architect i2000 analyzer; Abbott Diagnostics, Abbott Park, IL). Serum HBV DNA was tested by real-time quantitative PCR (Shanghai Kehua Bio-engineering Co., Ltd., China), with a lower limit of quantitation of 500 copies/ml. The diagnosis of metabolic syndrome was calculated using the International Diabetes Federation definition, which included central obesity (BMI >30 kg/m^2) and at least 2 of the following: triglycerides ≥1.69 mmol/L (150 mg/dl) or specific treatment for elevated triglycerides, reduced high-density lipoprotein-cholesterol (HDL) <1.04 mmol/L (40 mg/dl) in males or <1.29 mmol/L (50 mg/dl) in females or specific treatment for low HDL, elevated systolic blood pressure ≥130 mmHg or diastolic ≥85 mmHg or treatment of previously diagnosed hypertension, and elevated fasting plasma glucose ≥5.55 mmol/L (100 mg/dl) or previously diagnosed type 2 diabetes.[8]

2.3. Histological assessment

All liver biopsy samples of the patients were re-evaluated and scored by an experienced liver pathologist who was blinded to the patients' clinical, laboratory, and image data. The histological scoring system of the NASH CRN was used for our study.[9] Our pathologist scored hepatic ballooning with 3-point scales (0, 1,

2); hepatic steatosis with 4-point scales (0, 1, 2, 3); and fibrosis with 5-point scales (0, 1, 2, 3, and 4). The NAFLD activity score, ranged from 0 – 8 was the sum scores of hepatic steatosis, hepatic ballooning, and lobular inflammation scores. In this study, advanced fibrosis was defined as F3 or F4. The potential markers of fibrosis were then correlated with liver biopsy findings of necro-inflammation and fibrosis. The primary assessment of noninvasive markers in predicting advance fibrosis was defined as the accuracy for predicting stage III to IV fibrosis when compared with the gold standard of liver biopsy.

2.4. Assessments of serum markers and statistical methods

We calculated the values of several commonly used serum markers for fibrosis including FIB-4, AAR, APRI, and BARD. The cut-off values recommended by the American Association for the Study of Liver Diseases guidelines in 2018 were adopted in our study for diagnosis of fibrosis stage III to IV, as follows: AAR >1, APRI >1.5, FIB-4 >3.25, and BARD >2.[10] As a primary assessment, receiver operating characteristic (ROC) curve analysis was performed with each noninvasive marker for the accuracy of predicting advanced fibrosis based on the liver biopsy report in NAFLD patients with CHB. The area under the ROC curve (AUROC) was used to measure the overall performance of the calculated ROC curves. Ninety-five percent confidence intervals (CIs) were calculated for each AUROC using its standard error. The AUROCs from different markers were ranked from highest to lowest. The sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were also calculated for each ROC curve for predicting stage III to IV fibrosis.[5] The secondary assessments were as follows:

1. performed the aforementioned prespecific measurements of each marker for the accuracy of predicting advanced fibrosis in NAFLD patients without CHB;
2. ranked these markers with respect to their performance (i.e., accuracy) from highest to lowest based on their AUROCs, if available in the 2 groups of NAFLD patients with and without CHB; and
3. compared the markers with the best performance in predicting significant fibrosis between the 2 groups.

The aforementioned data analyses were performed using the Statistical Package for Social Science for Windows, Version 13.0 (SPSS Inc., Chicago, IL). Demographic data, clinical values, laboratory data, and histological scores with staging were calculated and summarized. Descriptive values were expressed as medians and interquartile ranges (median, Q1, Q3) or means ±standard (mean±SD) deviations, depending on the underlying distribution of the data. All tests were two-tailed with 95% CI. Statistical significance was set at a P value <.05.

3. Results

Among the 152 eligible patients who received care from June 2015 to August 2016 in our institution and were screened, 34 were excluded due to the following reasons: excessive alcohol intake (n=19), drug-induced liver injury (n=8), and other exclusion criteria (n=7). As a result, a total of 118 patients were analyzed (Figure 1). Among enrolled patients, 63.5% (75/118) were male with mean ± SD age of 40.17 (±12.22) years and mean ± SD BMI of 27.12±3.34 kg/m^2. Of the 118 NAFLD patients

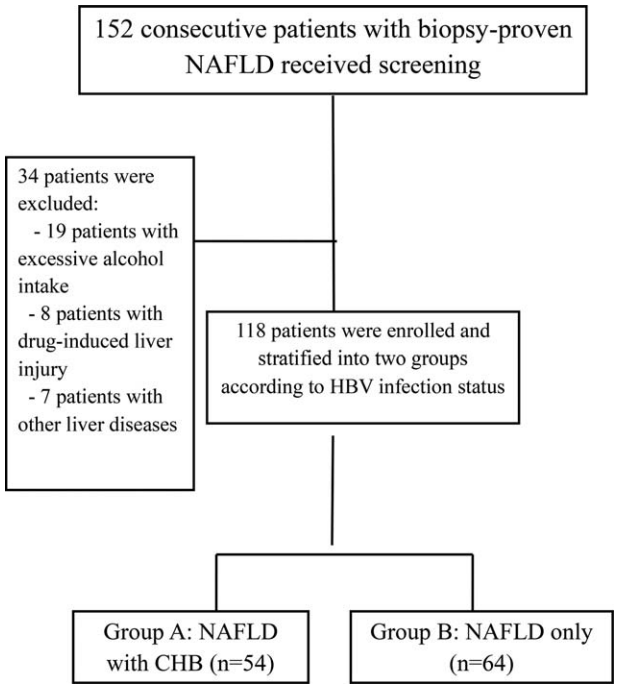

Figure 1. Patient enrollment and depositions. A total of 34 patients were excluded from the study owing to excessive alcohol intake. Among 54 NAFLD patients with CHB, 36.17% were HBeAg positive and 89.13% had HBV viremia.

with or without CHB, 34 (28.81%) patients had no NASH, 53 (44.92%) had borderline NASH, and 31 (26.27%) patients had definite NASH. The percentage of patients in the aforementioned 3 categories among patients without CHB was 31.25% (20/64), 42.19% (27/64), and 26.56% (17/64), respectively. In the NAFLD with CHB group, the percentage of patients in these categories was 25.93% (14/54), 48.15% (26/54), and 25.93% (14/54), respectively. The disease phases of most CHB patients were in either the immune tolerance or inactive phase defined by investigators. They were all treatment naïve. In our cohort, HBeAg was present in 36.17% of patients.

The clinical and biochemical data of 118 patients with NAFLD were divided into 2 groups including NAFLD with and without CHB for comparison (Table 1). Only few variables, including older age and higher levels of gamma-glutamyl transferase in the NAFLD without the CHB group when compared with those in the NAFLD with the CHB group, significantly differed between the 2 groups. The histological findings of the subjects are shown in Table 2. Based on the biopsy reports, 13.56% (16/118) of the patients met the definition of advanced fibrosis as they had stage III or IV fibrosis. However, only 1 patient had stage IV fibrosis (cirrhosis), but 15 of 118 (12.71%) had stage III fibrosis. A similar distribution of stage III fibrosis was observed in the NAFLD groups with CHB (n=7/54, 12.96%) and without CHB (n=8/64, 12.50%). We correlated stage III and IV fibrosis in liver biopsy with staging by serum fibrosis markers. These markers included FIB-4, AAR, APRI, and BARD. There were 9.32% of patients with fibrosis stage 0 on biopsy, and the percentage of patients with stage 0 did not significantly differ between the groups.

The primary assessments of the aforementioned serum markers including their sensitivity, specificity, PPV, NPV, and AUROC in NAFLD patients with CHB are shown in Table 3. In addition, the

Table 1

Clinical characteristic of NAFLD patients with or without CHB.

Variables, median (Q1, Q3) or specified	All study patients (n = 118)	NAFLD with CHB† (n = 54)	NAFLD only‡ (n = 64)	*P* value
Male patients (%)	74 (62.71%)	41 (75.93%)	33 (51.56%)	
Age, mean±SD	40.17 ± 12.22	36.89 ± 9.65	42.94 ± 13.48	*P* = .005
Height (cm), mean±SD	168.03 ± 8.99	171.19 ± 7.53 (18 lost)	165.89 ± 9.34 (11 lost)	*P* = .011
Weight (kg), mean±SD	76.56 ± 12.80	80.29 ± 11.87 (1 lost)	73.48 ± 12.80	*P* = .065
BMI (kg/m^2), mean±SD	27.12 ± 3.34	27.89 ± 3.07 (18 lost)	26.62 ± 3.47 (11 lost)	*P* = .827
HBeAg positivity (%)		17 (36.17%) (7 lost)		
HBV detectable (%)		41 (89.13%) (8 lost)		
ALT (U/L), median (Q1, Q3)	67.15 (37.78, 98.65)	57.85 (37.78, 90.15)	72.65 (34.98, 120.43)	.367
AST (U/L), median (Q1, Q3)	40.0 (29.8, 68.1)	35.85 (29.73, 56.98)	41.7 (29.8, 79.03)	.147
ALP (U/L), median (Q1, Q3)	70.95 (60.4, 84.98)	70.45 (58.85, 78.33) (8 lost)	71.15 (62.4, 96.2) (8 lost)	.202
GGT (U/L), median (Q1, Q3)	42.85 (26.75, 72.43)	34.6 (23.23, 55.8) (8 lost)	47.15 (30.85, 91.88) (8 lost)	.019
Total bilirubin (mmol/L), median (Q1, Q3)	11.55 (9.1, 14.75)	11.5 (9.08, 14.95)	11.6 (9.5, 14.65)	.783
Albumin (g/L), median (Q1, Q3)	46.55 (42.9, 49.12)	47.15 (43.9, 48.83)	46.0 (41.53, 49.20)	.234
Glucose (mmol/L), median (Q1, Q3)	6.04 (5.48, 6.9)	6.04 (5.54, 6.8) (11 lost)	6.0 (5.39, 7.15) (2 lost)	.982
Triglycerides (mmol/L), median (Q1, Q3)	1.44 (1.0, 1.88)	1.42 (1.08, 1.78) (21 lost)	1.45 (0.99, 2.01) (17 lost)	.718
Total cholesterol (mmol/L), median (Q1, Q3)	4.70 (3.83, 5.50)	4.62 (3.87, 5.50) (21 lost)	4.75 (3.83, 5.5) (17 lost)	.845
LDL-cholesterol (mmol/L), median (Q1,Q3)	2.9 (2.09, 3.43)	2.74 (2.09, 3.36) (21 lost)	2.94 (2.09, 3.47) (17 lost)	.689
HDL-cholesterol (mmol/L), median (Q1,Q3)	1.07 (0.90, 1.26)	1.03 (0.88, 1.21) (21 lost)	1.08 (0.95, 1.27) (17 lost)	.307
Platelet count (10^9/L), median (Q1,Q3)	202.15 (154.00, 240.75)	201.15 (169.75, 245.25)	206 (143.25, 237.25)	.282
Prothrombin time, median (Q1,Q3)	11.1 (10.6, 11.6)	11.2 (10.7, 11.6) (1 lost)	11.0 (10.5, 11.5) (2 lost)	.127
INR, median (Q1,Q3)	0.99 (2.0, 3.0)	0.98 (0.96, 1.03) (1 lost)	0.995 (0.95, 1.03) (2 lost)	.848

† For NAFLD patients with CHB, the missing data included the following: BMI (n = 18), HBeAg status (n = 7), HBV DNA levels (n = 8), and INR (n = 1).

‡ Among NAFLD patients without CHB, 11 and 2 patients had missing data on BMI and INR at the visit of liver biopsy.

The other missing data were not the variables involved in the calculation of serum markers for fibrosis. Thus, the missing data had no impact on the assessment of fibrosis scales.

BMI = body mass index, AST = aspartate aminotransferase, ALT = alanine aminotransferase, ALP = alkaline phosphatase, GGT = gamma-glutamyl transferase, HDL–cholesterol = high-density lipoprotein-cholesterol, LDL-cholesterol = low-density lipoprotein-cholesterol, INR = international normalized ratio.

ROC curves of the aforementioned 4 clinical prediction rules are presented in Figure 2A. In this group, analyses of FIB-4 with the cut off value (>3.25) revealed that 11 out of 54 (20.37%) patients had F3-F4 when compared with 7 out of 54 (12.96%) patients proven by biopsy. Thus, the sensitivity and specificity of FIB-4 for predicting advanced fibrosis were 0% and 76.60%, respectively. The NPV was 83.72%. The AUROC was 0.590 (0.432–0.747) 6goi0p+

. Analyses of APRI values with the cutoff score (>1.5) showed that 3 out of 54 (5.56%) patients had F3-F4. The sensitivity and specificity of APRI for predicting advanced fibrosis were 0 and 93.62%, respectively. The NPV was 86.72%. The AUROC was 0.676 (0.524–0.828). Using the cutoff value (>1.0) for AAR, 12 out of 54 patients (22.22%) had advanced fibrosis. When compared with the aforementioned biopsy results on fibrosis staging, the sensitivity and specificity of AAR for predicting advanced fibrosis were 14.29% and 76.60%, respectively. The NPV was 85.71%. The AUROC was 0.500 (0.298–0.702). Assessment of advanced fibrosis with BARD score in this group showed that 7 out of 54 (12.96%) patients had F3-F4 (values >2.0). Compared with staging by biopsy, the sensitivity and specificity of BARD for predicting advanced fibrosis were 14.29% and 87.23%, respectively. The NPV was 87.23%. The AUROC was 0.611 (0.391–0.831).

In assessments of serum markers in NAFLD patients without CHB, we based the aforementioned cut-off values in each marker and classified that 18/64 (28.12%), 9/64 (14.06%), 15/64 (23.44%), and 12/64 (18.75%) of patients had advanced fibrosis when they were assessed with AAR, FIB-4, APRI, and BARD, respectively. We calculated the sensitivity, specificity, PPV, NPV, and AUROC of these markers from the staging data obtained by biopsy. The results are shown in Table 4, and the receiver operating ROC curves of the aforementioned clinical prediction rules are presented in Figure 2B.

Based on the AUROCs, the serum markers ranked as follows from highest to lowest with respect to their accuracy in predicting

Table 2

Histological profile of NAFLD patients with or without hepatitis B.

Variables (mean ± SD) or specified	All study patients n=118	NAFLD with CHB (n=54)	NAFLD only (n=64)	*P* value
Steatosis				
0	0 (0)	0 (0)	0 (0)	
1	52 (44.07%)	22 (40.74%)	30 (46.87%)	.99
2	52 (44.07%)	26 (48.15%)	26 (40.63%)	.412
3	14 (11.86%)	6 (11.11%)	8 (12.50%)	.816
Lobular inflammation				
0	1 (0.85%)	0 (0)	1 (1.56%)	1.00
1	73 (61.86)	35 (64.81%)	38 (59.38%)	.544
2	41 (34.75%)	18 (33.33%)	23 (35.93%)	.767
3	3 (2.54%)	1 (1.86%)	2 (3.13%)	1.00
Ballooning				
0	62 (52.54%)	29 (53.70%)	33 (51.56%)	.816
1	40 (33.90%)	18 (33.33%)	22 (34.38%)	.905
2	16 (13.56)	7 (12.97%)	9 (14.06%)	.862
Fibrosis				
0	11 (9.32%)	7 (12.96%)	4 (6.25%)	.211
1	73 (61.86%)	35 (64.81%)	38 (59.38%)	.554
2	18 (15.25%)	5 (9.26%)	13 (20.31%)	.096
3	15 (12.71%)	7 (12.96%)	8 (12.50%)	.94
4	1 (0.85%)	0 (0)	1 (1.56%)	1.00

advanced fibrosis in NAFLD patients with CHB: APRI, BARD, FIB-4, and AAR. However, the ranking of performance in NAFLD patients without CHB was as follows: FIB-4, BARD, APRI, and AAR. Therefore, the serum markers with the best performance for predicting advanced fibrosis were APRI and FIB-4 in NAFLD patients with and without CHB, respectively. In our NAFLD patients without CHB, FIB-4 provided significantly better accuracy for predicting advance fibrosis than APRI ($P=.015$).

4. Discussion

In the present study, 4 scoring systems, namely, APRI, BARD, FIB-4, and AAR, were assessed for their accuracy in predicting advanced fibrosis. To our knowledge, this study is by far the largest one conducted in Asian NAFLD patients with CHB. We observed that the AUROC (95% CI) of the markers ranged from 0.500 (0.298–0.702) to 0.676 (0.524–0.828). APRI was the best performing marker. By contrast, the AUROC (95% CI) for the study markers ranged from 0.656 (0.427–0.884) to 0.862 (0.745–0.978) in NAFLD patients without CHB. The best performing marker was FIB-4.

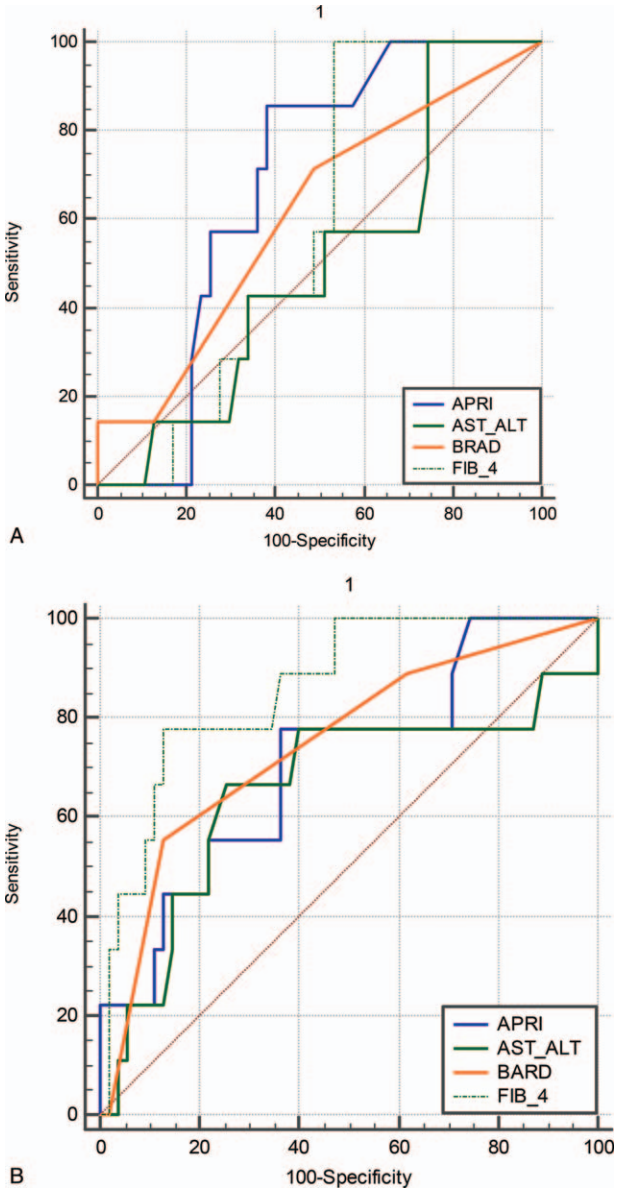

Figure 2. Receiver operating characteristic (ROC) curves for each serum marker and their correlations with biopsy staging. A: NAFLD patients with CHB; B: NAFLD patients without CHB. ROC curves for the 4 noninvasive serum markers for predicting biopsy-proven advanced fibrosis in patients with chronic hepatitis B: aspartate aminotransferase (AST) to alanine aminotransferase (ALT) ratio (AAR); AST to platelet ratio index (APRI); body mass index (BMI), AST to alanine aminotransferase (ALT) ratio, and diabetes (BARD) Score; d fibrosis-4 (FIB-4).

Table 3

Comparison of noninvasive scores with histological fibrosis in NAFLD patients with CHB.

Serum markers	Sensitivity	Specificity	PPV	NPV	AUROC (95% CI)	*P* values (compared to APRI)
APRI	0.00%	93.62%	0.00%	86.27%	0.676 (0.524–0.828)	N/A
BARD	14.29%	87.23%	14.29%	87.23%	0.611 (0.391–0.831)	.581
FIB-4	0.00%	76.60%	0.00%	83.72%	0.590 (0.432–0.747)	.265
AAR	14.29%	76.60%	8.33%	85.71%	0.500 (0.298–0.702)	.113

ALT = alanine aminotransferase, APRI = AST to platelet ratio index, AST = aspartate aminotransferase, AUROC = area under the ROC curve, FIB-4 = fibrosis-4, ALT, and AST and age, NAFLD = nonalcoholic fatty liver disease, NPV = negative predictive value, PPV = positive predictive value, ROC = receiver operating characteristic curve.

Table 4

Comparison of non-invasive scores with histological fibrosis in NAFLD patients without CHB.

Serum markers	Sensitivity	Specificity	PPV	NPV	AUROC (95% CI)	*P* values (compared to FIB-4)
FIB-4	77.78%	85.45%	46.67%	95.92%	0.862 (0.745–0.978)	N/A
BARD	55.56%	87.27%	41.67%	92.31%	0.745 (0.565–0.926)	.262
APRI	40.00%	90.74%	44.44%	89.09%	0.709 (0.525–0.893)	.015
AAR	55.56%	76.36%	27.78%	91.30%	0.656 (0.427–0.884)	.117

ALT = alanine aminotransferase, APRI = AST to platelet ratio index, AST = aspartate aminotransferase, AUROC = area under the ROC curve, FIB-4 = fibrosis-4, ALT, and AST and age, NAFLD = nonalcoholic fatty liver disease, NPV = negative predictive value, PPV = positive predictive value, ROC = receiver operating characteristic curve.

A recent study by De Silva et al compared 90 South Asian NAFLD patients with 85 white NAFLD patients.[11] They observed significant differences in the performance of noninvasive serum markers with respect to accuracy between the groups. The authors suggested that ethnicity should be considered when assessing NAFLD patients for fibrosis. Although both hepatitis B and NAFLD have high prevalence in Asia,[1,2,12] data on the accuracy of serum markers for predicting advanced fibrosis in Asian NAFLD patients with CHB are limited. Wadhva et al investigated NAFLD patients with hepatitis and end-stage renal diseases in Pakistan and found that APRI is more accurate in predicting advanced fibrosis. However, only 3 out of 109 patients analyzed in the study had hepatitis B.[13]

Among CHB patients without NAFLD, several studies have identified FIB-4 to predict CHB-related fibrosis with moderate sensitivity and accuracy.[14–18] However, the role of APRI for predicting advanced fibrosis in CHB patients without NAFLD remains controversial as 2 meta-analyses showed conflicting results.[14,17] Although our study patients shared some clinical features of those with CHB, the NAFLD status may have effects on hepatitis B progression and serum markers related to hepatic necroinflammatory activities.[19,20] Therefore, our findings on the best performing marker differed from those in studies that enrolled CHB patients without NAFLD or NAFLD patients without CHB. In the present study, the best performing marker in NAFLD patients without CHB was FIB-4, which has been well documented as the best choice in many studies and also recommended by recent published association guidelines for NAFLD patients.[6,21–26]

This study has 2 major limitations. First, the best performing marker had poor sensitivity. This could be attributed to the small number of patients with stage III or IV fibrosis. Further studies on advanced fibrosis with a larger sample size may provide additional information on the sensitivity of APRI in NAFLD patients with CHB. Second, the CHB patients in our study were mainly at the phase of immune tolerance or inactive phase of hepatitis B. Our findings could not be generalized to all CHB patients with NAFLD.

In conclusion, our study showed that APRI was the best performing marker for predicting advanced fibrosis in Asian patients with CHB and NAFLD. The sensitivity, specificity, PPV, NPV, and AUROC (95% CI) for APRI in this patient group were 0%, 93.62%, 0%, 86.27%, and 0.676 (0.524–0.828), respectively. The markers ranked as follows from highest to lowest with respect to their accuracy in predicting advanced fibrosis: APRI, BARD, FIB-4, and AAR. In contrast, FIB-4 was the best performing marker for predicting advanced fibrosis in NAFLD patients without CHB. The sensitivity, specificity, PPV, NPV, and AUROC (95% CI) for FIB-4 in this group were 77.78%, 85.45%, 46.67%, 95.92%, and 0.862 (0.745–0.978), respectively. The markers ranked as follows from highest to lowest with respect to their accuracy in predicting advanced fibrosis: FIB-4, BARD, APRI, and AAR. When the 2 groups of NAFLD patients with and without CHB were compared, serum markers were less reliable in predicting advanced fibrosis; however, APRI showed the best accuracy for predicting the absence of advanced fibrosis in NAFLD patients with CHB. Therefore, histopathological evaluation with liver biopsy still plays an important role in the diagnosis of advanced fibrosis in these NAFLD patients with CHB until new reliable mathematical models are formulated.

Author contributions

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