Application of alkaline phosphatase-to-platelet ratio as a novel noninvasive index predicts liver fibrosis in patients with chronic hepatitis B

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Abstract. Assessment of the extent of liver fibrosis is a crucial requirement for the design of antiviral treatments for patients with chronic hepatitis B (CHB). Several non-invasive predictive indices have been developed as potential alternatives to liver biopsy for fibrosis assessment. The present study aimed to establish a novel non-invasive method for predicting liver fibrosis in patients with CHB. A total of 382 patients with CHB who underwent liver biopsy and pathological examination at The Second Hospital of Anhui Medical University (Hefei, China) were enrolled into the present study. Liver fibrosis was assessed according to the meta-analysis of histological data in viral hepatitis scoring system. Logistic regression analyses were performed to explore possibly significant characteristics associated with liver fibrosis. In addition, potential correlations between the alkaline phosphatase (AKP)-to-platelet count (PLT) ratio (APPR) and the aspartate transaminase-to-platelet ratio index (APRI), fibrosis index based on four factors (FIB-4) and γ-glutamyl transpeptidase-to-platelet ratio (GPR) were assessed using Spearman's correlation analysis. Subsequently, the performance of APPR was compared with APRI, FIB-4 and GPR using receiver operating characteristic (ROC) analysis. Logistic regression analysis identified AKP and PLT to be significant independent predictors of fibrosis. Therefore, an index was then constructed for predicting the degree of fibrosis, which was expressed using the formula APPR=AKP (IU/ml)/PLT ($1x10^9$/l). APPR was found to be positively associated with the fibrotic stage of the liver in addition to being positively correlated with APRI, FIB-4 and GPR. The area under the ROC curve (AUROC) values of APPR were also significantly higher compared with those of APRI and FIB-4 in predicting significant fibrosis but were equal to those of GPR. However, for advanced fibrosis and cirrhosis, the AUROC value of APPR was shown to be higher compared with that of APRI, FIB-4 and GPR. In conclusion, these observations suggest that APPR is a viable marker that can be used to assess liver fibrosis in patients with CHB.

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Introduction

Liver fibrosis occurs during the crucial stage of disease progression from chronic hepatitis B (CHB) virus (HBV) infection to cirrhosis, which is a key determinant of the therapeutic decision taken for this condition (1,2). According to the European Association for the Study of the Liver guidelines, treatment should be initiated in patients with significant fibrosis [meta-analysis of histological data in viral hepatitis (METAVIR) scores ≥F2] (3). Therefore, accurate assessment of liver fibrosis is critical for the establishment of optimal management protocols in clinical practice.

Liver biopsy is currently the gold standard used for the assessment of liver fibrosis, although it is far from being perfect (4,5). Since it is an invasive procedure, it confers certain inevitable limitations such as pain, risk of severe complications and sampling errors that restrict its wider application to fibrosis screening in patients with CHB (6,7). Therefore, a number of non-invasive tests for fibrosis have been developed for clinical application, several of which are becoming widely applied. In particular, the aspartate transaminase (AST)-to-platelet ratio index (APRI) and the fibrosis index based on four factors (FIB-4) are non-invasive tests that have proposed widespread utility for the clinical assessment of liver fibrosis (8-10). However, various obstacles remain to be overcome, including the complexity, reproducibility, diagnostic accuracy and the cost of routine use (11). Lemoine *et al* (12) previously proposed a novel index based on the γ-glutamyl transpeptidase (GGT)-to-platelet ratio (GPR) for identifying patients with fibrosis or cirrhosis who are afflicted with CHB infections in West Africa. Subsequently, the favorable performance of GPR for predicting fibrosis and cirrhosis was verified by another previous study of HBV e antigen-positive patients with CHB in China (13). However, this novel index requires further validation in larger cohorts of patients with CHB, particularly in China.

In the present study, the potential relationship between fibrosis and a number of parameters were investigated in a large cohort of well-characterized, treatment-naïve patients with CHB. In addition, a novel non-invasive index was developed with the aim of discriminating patients with CHB with different grades of liver fibrosis.

Materials and methods

Study population. A total of 382 patients with CHB were selected from the clinic or hospitalization unit at the Department of Infectious Diseases of The Second Affiliated Hospital of Anhui Medical University (Hefei, China) between January 2014 and December 2018. None of the patients had history of treatment for HBV (interferon, nucleoside, nucleotide analogues or other drugs directed against HBV) prior to the present study. The clinical diagnosis of all selected patients complied with the standards for CHB diagnosis in the prevention and treatment guidelines recommended for liver disease by the Chinese Medical Association (14). Patients with the following conditions were excluded from the present study: i) Co-infection with another hepatitis virus or human immunodeficiency virus; ii) alcoholic liver disease; iii) nonalcoholic fatty liver disease; and/or iv) autoimmune liver disease. The present study protocol complied with the Ethical Guidelines of Anhui Medical College and The Second Hospital of Anhui Medical University. Written informed consent was obtained from all individuals included in the present study.

Table I. Clinical characteristics of the patients in the study.

Characteristics	Recorded data
Sex	
Male	268 (70.16)
Female	114 (29.84)
Age, years	38.48±19.09
Alanine transaminase, IU/l	50 (30-105)
Aspartate aminotransferase, IU/l	38 (26-67)
Total bilirubin, μmol/l	13 (10-18)
γ-glutamyl transpeptidase, IU/l	32 (19-64)
Alkaline phosphatase, IU/l	92 (74-118)
Platelet count, 10^9/l	168.33±19.79
Hepatitis B virus DNA, $\log_{10}$	5.03±2.65
Fibrosis stage	
F0	14 (3.66)
F1	188 (49.21)
F2	90 (23.56)
F3	59 (15.45)
F4	31 (8.12)

Data are presented as either n (%), mean ± SD or median (interquartile range). F0, no fibrosis; F1, portal fibrosis without septa; F2, portal fibrosis with rare septa; F3, numerous septa without cirrhosis; F4, cirrhosis.

Clinical evaluation. Clinical and laboratory parameters of all patients were extracted from the medical records, including age, sex, alanine aminotransferase (ALT), AST, alkaline phosphatase (AKP), GGT, albumin and total bilirubin levels, international normalized ratio, platelet counts (PLT) and HBV DNA levels. If a patient was admitted > once during the study period, only information obtained on the first admission was extracted.

Liver biopsy. The present protocol was approved by the Ethics Committee of the Second Hospital of Anhui Medical University, in which all patients provided informed consent prior to liver biopsy. Needle biopsy was performed using an automatic biopsy gun with a 16G disposable needle (Bard Ma x-Core Disposable Biopsy Instrument; Becton, Dickinson and Company) under sonographic guidance (thermal index for soft tissue, 0.4; mechanical index, 1.3). The liver biopsy specimens were fixed in 10% formalin at room temperature for 2 h and embedded in paraffin. The paraffin sections were stained with hematoxylin for 5 min and with eosin for 1 min, or with Masson staining for 5 min at room temperature for pathological diagnosis. Liver fibrosis was evaluated according to the METAVIR scoring system as follows: i) F0, no fibrosis; ii) F1, portal fibrosis without septa; iii) F2, portal fibrosis with rare septa; iv) F3, numerous septa without cirrhosis; and v) F4, cirrhosis. Significant fibrosis was defined as ≥F2, advanced fibrosis was defined as ≥F3 and cirrhosis was defined as F4. All biopsy samples were analyzed by two independent pathologists. These slides were also reviewed by a third hepato-pathologist in the event of any discrepancies.

Non-invasive index calculation. APRI was calculated as the APRI=[AST (IU/l)/upper limit of normal] x100/PLT (10^9/l). FIB-4 was calculated using the formula [(age, years) x (AST, U/l]/[(platelet count, 10^9/l) x√(ALT, U/l)]. GPR was calculated using the formula (GGT, IU/l)/(PLT, 10^9/l).

Statistical analysis. Quantitative data are expressed as either n (%), mean ± SD or median (interquartile range). All statistical analyses were conducted using SPSS version 16.0 (SPSS, Inc.), GraphPad Prism 5.0 (GraphPad Software, Inc.) and MedCalc statistical software 10.4 (MedCalc Software bvba).

Shapiro-Wilk's test or Kolmogorov-Smirnov test were used to test the normality of continuous variables, where those with a P>0.10 were considered to be normally distributed. Categorical variables were compared using the χ^2 test, whereas continuous variables were compared using unpaired Student's t-test, the Mann-Whitney test or the Kruskal-Wallis test (followed by Dunn's test) as appropriate. P<0.05 was considered to indicate a statistically significant difference. Correlations were evaluated using Spearman's correlation coefficient. To identify significant independent predictors of liver fibrosis (≥F2), univariate and subsequent multivariable logistic regression analyses with a backwards stepwise method were performed to identify them (15). The area under the receiver operating characteristic (AUROC) curve, sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) of the non-invasive indices were also calculated. The optimal cut-off values used for each test were determined by maximizing the Youden's index (sensibility + specificity-1).

Table II. Univariate analysis of variables associated with the presence of significant fibrosis.

Characteristics	No significant fibrosis (F0 + F1)	Significant fibrosis (F2 + F3 + F4)	P-value
Age, years	36.81±11.29	39.40±12.24	0.016
Sex, male/female	135/67	133/47	0.146
Alanine transaminase, IU/l	43 (25-86)	60 (36-134)	<0.001
Aspartate aminotransferase, IU/l	32 (23-60)	43 (31-72)	<0.001
Total bilirubin, μmol/l	12.9 (10-17.3)	13.8 (10.3-18.8)	0.234
γ-glutamyl transpeptidase, IU/l	24 (16-42)	42 (25-90)	<0.001
Platelet count, 10^9/l	180.68±51.24	162.62±62.01	<0.001
Alkaline phosphatase, IU/l	84 (68-103)	101 (82-130)	<0.001
Hepatitis B virus DNA, $\log_{10}$	5.23±1.96	5.27±1.85	0.504

Data are presented as either mean ± SD or median (interquartile range). F0, no fibrosis; F1, portal fibrosis without septa; F2, portal fibrosis with rare septa; F3, numerous septa without cirrhosis; F4, cirrhosis.

Table III. Independent predictors of significant fibrosis by multivariate logistic regression analysis.

Characteristics	Odds ratio	95% CI	P-value
Age	1.017	0.996-1.038	0.120
Sex	0.939	0.567-1.556	0.807
Alanine transaminase	1.009	1.004-1.015	0.001
Aspartate aminotransferase	0.982	0.972-0.992	<0.001
Total bilirubin	0.996	0.983-1.009	0.543
γ-glutamyl transpeptidase	1.003	0.999-1.007	0.169
Alkaline phosphatase	1.008	1.002-1.015	0.009
Platelet count	0.986	0.982-0.991	<0.001
Hepatitis B virus DNA	1.053	0.926-1.198	0.430

Results

General characteristics of the study population. In total, data from 382 patients with CHB were obtained (Table I). Among them, 14 patients (3.66%) were histologically classified as F0, 188 (49.21%) as F1, 90 (23.56%) as F2, 59 (15.45%) as F3 and 31 (8.12%) as F4.

Development of the AKP-to-PLT (APPR) index. The presence of significant liver fibrosis was found to be associated with age (P=0.016), ALT (P<0.001), AST (P<0.001), GGT (P<0.001), AKP (P<0.001) and PLT (P<0.001) according to univariate analysis (Table II). Subsequent multiple logistic regression analysis using a backwards stepwise method identified ALT (P=0.001), AST (P<0.001), AKP (P=0.009) and PLT (P<0.001) to be independent predictors of significant liver fibrosis (Table III). Based on this, a non-invasive index of APPR was developed using the formula (AKP, IU/ml)/(PLT, 10^9/l).

Association between APPR with different stages of fibrosis and other noninvasive fibrosis indices. The APPR increased with each advancing stage of fibrosis. There was no difference in the APPR of patients with F1 fibrosis compared with that in patients with F0 fibrosis. However, the APPR in patients with F2 fibrosis (P<0.0001), F3 fibrosis (P<0.0001) and F4 fibrosis (P<0.0001) was all significantly higher compared with that in patients with F1 fibrosis (Fig. 1). In addition, a significant difference in the APPR among the five stages of fibrosis was detected (P<0.0001), according to the Kruskal-Wallis test (Fig. 1). APPR was also found to be positively correlated with other established non-invasive test methods of fibrosis (Fig. 2), specifically with APRI (rho=0.504; P<0.0001), FIB-4 (rho=0.535; P<0.0001) and GPR (rho=0.591; P<0.0001).

Diagnostic performance of APPR for predicting fibrosis and cirrhosis. The performance of APPR in predicting fibrosis was subsequently evaluated using AUROC analysis with liver biopsy analysis data used as the reference standard. For predicting fibrosis, the AUROC of APPR was calculated to be significantly higher compared with that of APRI (P=0.034) and FIB-4 (P<0.001), but it was no different compared with that of GPR (Tables IV and V). The optimal cut-off value of APPR was 0.64, with 58.89% sensitivity, 79.21% specificity, 71.62% PPV and 68.37% NPV (Fig. 3).

For predicting advanced fibrosis, the AUROC of APPR was higher compared with that of APRI, FIB-4 and GPR, but there was no statistical significance (Tables IV and V). The optimal cut-off value of APPR was 0.65, with a sensitivity of 68.89%, specificity of 70.55%, PPV of 41.89% and NPV of 88.03% (Fig. 3).

Table IV. Diagnostic performance of APPR, APRI, GPR and FIB-4 for the diagnosis of fibrosis and cirrhosis.

	Significant fibrosis		Advanced fibrosis		Cirrhosis	
Characteristics	AUROC (95% CI)	Cut-off values	AUROC (95% CI)	Cut-off values	AUROC (95% CI)	Cut-off values
APPR	0.73 (0.679-0.771)	0.64	0.75 (0.704-0.793)	0.65	0.82 (0.785-0.863)	0.67
APRI	0.66 (0.617-0.713)	0.52	0.71 (0.660-0.754)	0.53	0.74 (0.693-0.783)	0.75
FIB-4	0.63 (0.581-0.681)	1.37	0.70 (0.653-0.747)	2.3	0.75 (0.704-0.793)	2.9
GPR	0.73 (0.675-0.767)	0.21	0.74 (0.701-0.790)	0.22	0.78 (0.739-0.824)	0.27

APPR, alkaline phosphatase-to-platelet ratio; APRI, aspartate transaminase-to-platelet ratio; FIB-4, fibrosis index based on four factors; GPR, γ-glutamyl transpeptidase-to-platelet ratio; AUROC, area under the receiver operating characteristic curve.

Table V. Comparative analysis of AUROCs.

	P-values		
Comparison of AUROCs	Significant fibrosis	Advanced fibrosis	Cirrhosis
APPR vs. aspartate transaminase-to-platelet ratio	0.034	0.144	0.055
APPR vs. fibrosis index based on four factors	<0.001	0.107	0.102
APPR vs. γ-glutamyl transpeptidase-to-platelet ratio	0.873	0.902	0.090

AUROCs, area under the receiver operating characteristic curve; APPR, alkaline phosphatase-to-platelet ratio.

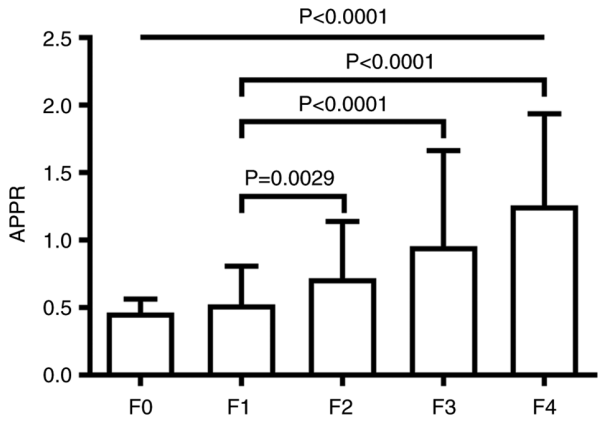

Figure 1. Association between APPR and the five stages of liver fibrosis. APPR, alkaline phosphatase-to-platelet ratio.

For predicting cirrhosis, the AUROC of APPR was also higher compared with that of APRI, FIB-4 and GPR, but there was also no statistical significance (Tables IV and V). The optimal cut-off value of APPR was 0.67, with sensitivity of 87.10%, specificity of 68.38%, PPV of 19.55% and NPV of 98.36% (Fig. 3).

Discussion

Liver fibrosis is commonly found in patients with CHB infection (16,17). Previous epidemiological studies have revealed that there are >350 million HBV carriers worldwide (18). In addition, these carriers are at increased risk of developing advanced fibrosis and cirrhosis during their lifetime. In China, there are ~20 million carriers of CHB, 10-30% of whom will progress to advanced fibrosis, cirrhosis or hepatocellular carcinoma (19). Although the pathophysiological mechanisms underlying the progression from HBV infection to liver fibrosis remain poorly understood, significant progress has been made in the diagnosis of hepatic fibrosis (20). At present, liver biopsy is the gold standard for the diagnosis of hepatic fibrosis. However, it has several disadvantages, including invasiveness, clinical complications, high cost and poor patient compliance (21). Considering these limitations, non-invasive predictors of fibrosis are now in demand. Ideally, a non-invasive method for application in clinical settings should not be too complex whilst being rapid, reproducible and cost-effective to perform (22). APRI and FIB-4 are currently widely utilized for measuring the degree of fibrosis in patients with CHB, particularly in regions with limited health care resources (23,24). In 2015, the APRI was recommended by the WHO as the preferred non-invasive tool for detecting significant fibrosis (25). However, a previous study from Singapore demonstrated the AUROC of APRI in predicting significant fibrosis to be only 0.63, suggesting APRI may not be sufficiently accurate for patients with CHB in some countries or regions (26). To the best of our knowledge, none of the non-invasive markers that have been studied have yielded satisfactory results (27).

In the present study, a potentially simpler method for predicting liver fibrosis and cirrhosis was developed using routinely available laboratory test results. ALT, AST, AKP and PLT were found to be independent predictors of significant

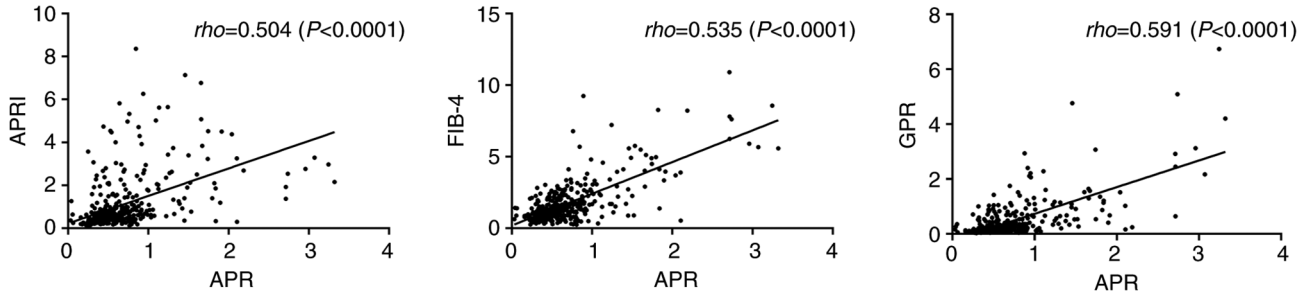

Figure 2. Correlation analysis of APPR with APRI, FIB-4 and GPR. APPR, alkaline phosphatase-to-platelet ratio; APRI, aspartate transaminase-to-platelet ratio; FIB-4, fibrosis index based on four factors; GPR, γ-glutamyl transpeptidase-to-platelet ratio.

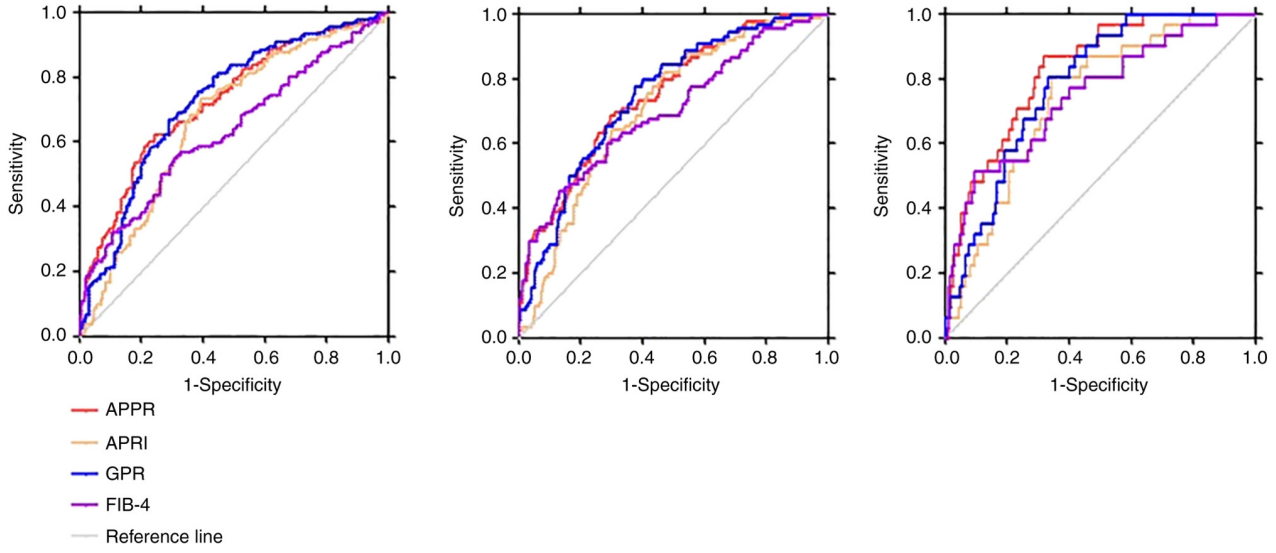

Figure 3. Receiver operating characteristics curves for the prediction of significant fibrosis, advanced fibrosis and cirrhosis by each of the four indices examined. APPR, alkaline phosphatase-to-platelet ratio; APRI, aspartate transaminase-to-platelet ratio; FIB-4, fibrosis index based on four factors; GPR, γ-glutamyl transpeptidase-to-platelet ratio.

fibrosis. All independent predictors except for AKP, form the principal components of available non-invasive indices that are currently applied, including APRI, FIB-4 and GPR. Similar to GGT, AKP is also a marker of liver fibrosis as part of the liver biochemical testing routine. As previously reported, non-invasive indices APRI, FIB-4 and GPR can be calculated using age, ALT, AST, GGT and PLT, all of which can be applied to predict fibrosis in patients with CHB (28). For this reason, it was hypothesized that the APPR may also be an useful index for distinguishing liver fibrosis from CHB. Therefore, following univariate and multivariate logistic regression analyses, an APPR two-marker index was developed. For significant fibrosis, APPR displayed superior performance compared with APRI and FIB-4. Compared with GPR, APPR exhibited similar diagnostic performance for predicting significant fibrosis. Since significant fibrosis is a determinant factor for the designation of the antiviral therapy strategy in patients with CHB, the application of APPR for significant fibrosis screening may be of clinical benefit. In addition to the superior diagnostic performance for predicting significant fibrosis, two attractive features of APPR have also been found: Ease of calculation and low cost. These advantages conferred by APPR may reduce the need for liver biopsy in patients with CHB to partially relieve the high burden of HBV infection in China. APPR can also be used to assess advanced fibrosis and cirrhosis. The AUROCs of APPR were 0.75 and 0.82 for the diagnosis of advanced fibrosis and cirrhosis in the present study, respectively. These were either somewhat superior or comparable to the AUROCs of APRI, FIB-4 and GPR. In particular, GPR predicted significant fibrosis with an AUROC of 0.73 in the present study. This was not consistent with the results of a previous study from China (AUROC, 0.66) (13). This discrepancy may have originated from the different composition of study populations. A study from Li *et al* (13) was conducted in patients positive for HBV e-antigen with high HBV DNA levels and with low levels of transaminases. This patient population is generally considered to be at low risk of developing significant fibrosis and cirrhosis. However, the present study population consisted entirely of patients with CHB. Although it remains difficult to explain why the diagnostic performance of APPR is superior to that of APRI and FIB-4, two possible explanations can be proposed. Elevated AKP levels showed the lowest coefficient of variation among all liver biochemical parameters tested, including ALT, AST and GGT (29). This finding suggests that AKP is a more stable parameter compared with other liver biochemical parameters.

In addition, the fluctuating aminotransferase levels in patients with chronic viral hepatitis can influence the predictive accuracy of non-invasive fibrosis scoring systems containing AST or ALT (30).

A number of limitations warrant consideration in the present study. This was only a single-center study, which lacked a validation cohort. Therefore, the findings in the present study require validation in multi-center trials. Furthermore, the use of liver biopsy as the 'gold standard' was a potential limitation, since this procedure may yield 'imperfect gold standard bias' (31,32). The reported data may be more convincing if APPR was compared with some other indices, such as aspartate aminotransferase-to-alanine aminotransferase ratio, fibrosis index and King scores (33). Therefore, further studies are required to clarify the relationship between the different HBV genotypes and APPR.

In conclusion, the present study found that APPR provides a potentially simple, inexpensive, rapid and non-invasive method for assessing significant fibrosis, advanced fibrosis and cirrhosis in patients with CHB, which may reduce the need for liver biopsy in China.

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Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors' contributions

KY and YP designed the study. BS collected the clinical samples and data. JC and PT analyzed and interpreted the data. KY and YP confirm the authenticity of all the raw data. All authors read and approved the final manuscript.

Ethics approval and consent to participate

The present protocol was approved by the Ethics Committee of the Second Hospital of Anhui Medical University, where all patients provided their written informed consent prior to liver biopsy. Written informed consent has been obtained from all individuals included in the present study.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Mak LY, Cloherty G, Wong DK, Gersch J, Seto WK, Fung J and Yuen MF: HBV RNA profiles in patients with chronic hepatitis B under different disease phases and antiviral therapy. Hepatology 73: 2167-2179, 2021.
2. Svicher V, Salpini R, Piermatteo L, Carioti L, Battisti A, Colagrossi L, Scutari R, Surdo M, Cacciafesta V, Nuccitelli A, *et al*: Whole exome HBV DNA integration is independent of the intrahepatic HBV reservoir in HBeAg-negative chronic hepatitis B. Gut 70: 2337-2348, 2021.
3. European Association for the Study of the Liver. EASL 2017 Clinical Practice Guidelines on the management of hepatitis B virus infection. J Hepatol 67: 370-398, 2017.
4. Kisseleva T and Brenner D: Molecular and cellular mechanisms of liver fibrosis and its regression. Nat Rev Gastroenterol Hepatol 18: 151-166, 2021.
5. Przybyłkowski A, Szeligowska J, Januszewicz M, Raszeja-Wyszomirska J, Szczepankiewicz B, Nehring P, Górnicka B, Litwin T and Członkowska A: Evaluation of liver fibrosis in patients with Wilson's disease. Eur J Gastroenterol Hepatol 33: 535-540, 2021.
6. Jing H, Yi Z, He E, Xu R, Shi X, Li L, Sun L, Liu Y, Zhang L and Qian L: Evaluation of risk factors for bleeding after ultrasound-guided liver biopsy. Int J Gen Med 14: 5563-5571, 2021.
7. Rustagi T, Newton E and Kar P: Percutaneous liver biopsy. Trop Gastroenterol 31: 199-212, 2010.
8. Lee J, Vali Y, Boursier J, Spijker R, Anstee QM, Bossuyt PM and Zafarmand MH: Prognostic accuracy of FIB-4, NAFLD fibrosis score and APRI for NAFLD-related events: A systematic review. Liver Int 41: 261-270, 2021.
9. Lai M and Afdhal NH: Liver fibrosis determination. Gastroenterol Clin North Am 48: 281-289, 2019.
10. Udompap P, Sukonrut K, Suvannarerg V, Pongpaibul A and Charatcharoenwitthaya P: Prospective comparison of transient elastography, point shear wave elastography, APRI and FIB-4 for staging liver fibrosis in chronic viral hepatitis. J Viral Hepat 27: 437-448, 2020.
11. Dong H, Xu C, Zhou W, Liao Y, Cao J, Li Z and Hu B: The combination of 5 serum markers compared to FibroScan to predict significant liver fibrosis in patients with chronic hepatitis B virus. Clin Chim Acta 483: 145-150, 2018.
12. Lemoine M, Shimakawa Y, Nayagam S, Khalil M, Suso P, Lloyd J, Goldin R, Njai HF, Ndow G, Taal M, *et al*: The gamma-glutamyl transpeptidase to platelet ratio (GPR) predicts significant liver fibrosis and cirrhosis in patients with chronic HBV infection in West Africa. Gut 65: 1369-1376, 2016.
13. Li Q, Li W, Huang Y and Chen L: The gamma-glutamyl transpeptidase-to-platelet ratio predicts liver fibrosis and cirrhosis in HBeAg-positive chronic HBV infection patients with high HBV DNA and normal or mildly elevated alanine transaminase levels in China. J Viral Hepat 23: 912-919, 2016.
14. Chinese Society of Infectious Diseases, Chinese Medical Association; Chinese Society of Hepatology, Chinese Medical Association. The guidelines of prevention and treatment for chronic hepatitis B (2019 version). Zhonghua Gan Zang Bing Za Zhi 27, 938-961, 2019. DOI: 10.3760/cma.j.issn.1007-3418.2019.12.007.
15. Steyerberg EW, Eijkemans MJ and Habbema JD: Stepwise selection in small data sets: A simulation study of bias in logistic regression analysis. J Clin Epidemiol 52: 935-942, 1999.
16. Zhan Z, Chen Y, Duan Y, Li L, Mew K, Hu P, Ren H and Peng M: Identification of key genes, pathways and potential therapeutic agents for liver fibrosis using an integrated bioinformatics analysis. PeerJ 7: e6645, 2019.
17. Ye J, Wang W, Feng S, Huang Y, Liao X, Kuang M, Xie X, Liao B and Zhong B: Precise fibrosis staging with shear wave elastography in chronic hepatitis depends on liverinflammation and steatosis. Hepatol Int 14: 190-201, 2020.
18. Winer BY, Huang T, Low BE, Avery C, Pais MA, Hrebikova G, Siu E, Chiriboga L, Wiles MV and Ploss A: Recapitulation of treatment response patterns in a novel humanized mouse model for chronic hepatitis B virus infection. Virology 502: 63-72, 2017.
19. Zheng X, Wang J and Yang D: Antiviral therapy for chronic hepatitis B in China. Med Microbiol Immunol 204: 115-120, 2015.
20. Bi J, Liu L and Qin T: Comparison of magnetic resonance elastography and transient elastography in the diagnosis of hepatic fibrosis: A systematic review and meta-analysis. Ann Palliat Med 10: 8692-8700, 2021.

21. Wang D, Wang Q, Shan F, Liu B and Lu C: Identification of the risk for liver fibrosis on CHB patients using an artificial neural network based on routine and serum markers. BMC Infect Dis 10: 251, 2010.
22. Shin YR, Kim SU, Lee S, Choi JY, Park HK, Yoo JE and Park YN: Noninvasive surrogates are poor predictors of liver fibrosis in patients with Fontan circulation. J Thorac Cardiovasc Surg 24: S0022-S5223, 2021.
23. Liu K, Qin M, Tao K, Liang Z, Cai F, Zhao L, Peng P, Liu S, Zou J and Huang J: Identification and external validation of the optimal FIB-4 and APRI thresholds for ruling in chronic hepatitis B related liver fibrosis in tertiary care settings. J Clin Lab Anal 35: e23640, 2020.
24. Huang R, Wang G, Tian C, Liu Y, Jia B, Wang J, Yang Y, Li Y, Sun Z, Yan X, *et al*: Gamma-glutamyl-transpeptidase to platelet ratio is not superior to APRI,FIB-4 and RPR for diagnosing liver fibrosis in CHB patients in China. Sci Rep 7: 8543, 2017.
25. WHO. World Health Organization. Guidelines for the Prevention, Care and Treatment of Persons with chronic Hepatitis B infection. 2015. http://whoint/hiv/pub/ hepatitis/hepatitis-b-guidelines/en/.
26. Wai CT, Cheng CL, Wee A, Dan YY, Chan E, Chua W, Mak B, Oo AM and Lim SG: Non-invasive models for predicting histology in patients with chronic hepatitis B. Liver International 26: 666-672, 2006.
27. Castera L: Invasive and non-invasive methods for the assessment of fibrosis and disease progression in chronic liver disease. Best Pract Res Clin Gastroenterol 25: 291-303, 2011.
28. Kim MN, Lee JH, Chon YE, Ha Y and Hwang SG: Fibrosis-4, aspartate transaminase-to-platelet ratio index, and gamma-glutamyl transpeptidase-to-platelet ratio for risk assessment of hepatocellular carcinoma in chronic hepatitis B patients: Comparison with liver biopsy. Eur J Gastroenterol Hepatol 32: 433-439, 2020.
29. Lazo M, Selvin E and Clark JM: Brief communication: Clinical implications of short-term variability in liver function test results. Ann Intern Med 148: 348-352, 2008.
30. Pol S, Carnot F, Nalpas B, Lagneau JL, Fontaine H, Serpaggi J, Serfaty L, Bedossa P and Bréchot C: Reversibility of hepatitis C virus-related cirrhosis. Hum Pathol 35: 107-112, 2004.
31. Zhou X, Pete C and Zhou C: Non-parametric estimation of ROC curves in the absence of a gold standard. Biometrics 61: 600-609, 2005.
32. Maharaj B, Maharaj RJ, Leary WP, CooppanRM, Naran AD, Pirie D and Pudifin DJ: Sampling variability and its influence on the diagnostic yield of percutaneous needle biopsy of the liver. Lancet 1: 523-525, 1986.
33. Deng H, Qi X, Peng Y, Li J, Li H, Zhang Y, Liu X, Sun X and Guo X: Diagnostic accuracy of APRI, AAR, FIB-4, FI, and king scores for diagnosis of esophageal varices in liver cirrhosis: A retrospective study. Med Sci Monit 21: 3961-3977, 2015.