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Research Letter

A Novel Algorithm for Streamlining Diagnosis of Advanced Liver Fibrosis in CHB Patients with Concurrent Hepatic Steatosis

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Chronic hepatitis B (CHB) with concurrent hepatic steatosis (HS) is a growing challenge for public health globally. As the prevalence of HS increases in Asia where CHB is endemic,[1] the combined prevalence of HS and CHB is estimated to be 32.8%.[2] Furthermore, the co-existence of these diseases is associated with more frequent and severe adverse clinical events such as liver fibrosis than HS or CHB alone.[3] There is a need for reliable diagnostic methods to detect and manage this condition in affected individuals. It should be noted that histologic evaluation via liver biopsy remains the gold standard to diagnose advanced fibrosis in patients with chronic liver diseases. Despite its effectiveness, many patients are hesitant to undergo a liver biopsy due to its invasive nature and the potential risks involved. Some of the most well-known complications associated with liver biopsy include bleeding, infection, and damage to nearby organs. Hence, alternative non-invasive tests (NITs), including the aspartate aminotransferase to platelet ratio index (APRI) , fibrosis index based on the four factors (FIB-4), and nonalcoholic fatty liver disease fibrosis score (NFS) have been proposed.[4–6] However, the diagnostic accuracy of these tests in CHB patients with concurrent HS is not yet clear. In this retrospective study, we aimed to evaluate the performance of APRI, FIB-4, and NFS for predicting advanced fibrosis in patients with both CHB and HS. Furthermore, we introduced the combined models that call for improvements in the current diagnostic workflow.

We consecutively enrolled untreated CHB patients with concurrent HS who underwent liver biopsy from nine medical centers in China between April 2004 and September 2021 (Clinical Trials. gov identifier: NCT05766449). The liver biopsy was performed to evaluate the extent of steatosis, inflammation, and fibrosis, and to determine if patients required antiviral therapy initiation.[7,8] Patients who were hepatitis B virus surface antigen positive for more than 6 months, and had hepatic steatosis of more than 5% as confirmed by liver biopsy, were included in the study. Patients with alcoholic fatty liver or other causes of liver disease, diagnosed with hepatocellular carcinoma, or other malignancies were excluded. The study was approved by ethics committees at all participating centers. The stage of fibrosis was assessed by Scheuer's score. Based on Scheuer's classification, the stages of fibrosis were determined based on the degree of portal/periportal and lobular activity. In this system, S3 represents severe fibrosis with bridging fibrosis, and we defined ≥S3 as advanced fibrosis.[9] Clinical and laboratory data, including demographic characteristics, routine blood tests, and liver biochemistry data, were systematically extracted into standardized forms. During the data analysis process, the Kolmogorov-Smirnov test was used to determine if the data followed a normal distribution. Continuous variables were presented as the mean and standard deviation if the variables showed a normal distribution, or as the median (interquartile range). Categorical variables were expressed as numbers (percentages). Statistical analysis was performed using MedCalc software (version 19.8). The area under the receiver operating characteristic, sensitivity, specificity, positive and negative predicted values were calculated for the two estimated cut-off points for each score (the lower and higher cut-offs for ruling out and ruling in advanced fibrosis, respectively). Subsequently, we computed the proportion of patients with the correct classification (the sum of true positives and negatives) for different combinations of NITs.

We retrospectively enrolled 926 eligible patients over the study period. The patients had a median age of 38.00 years, 697 (75.27%) were male, and the median body mass index was 25.47, with 80 patients (8.64%) having diabetes. In terms of steatosis classification, 752 (81.21%) patients were categorized as grade 1, 126 (13.61%) as grade 2, and 48 (5.18%) as grade 3 (Table 1). Among them, 180 (19.44%)

Abbreviations: APRI, aspartate aminotransferase to platelet ratio index; CHB, chronic hepatitis B; FIB-4, fibrosis index based on the four factors; HS, hepatic steatosis; NFS, nonalcoholic fatty liver disease fibrosis score; NITs, non-invasive tests.

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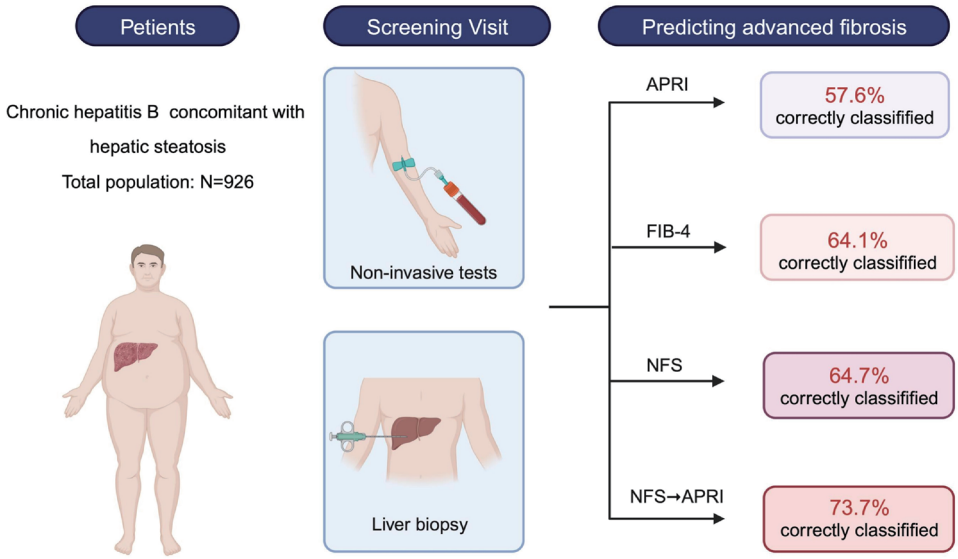
Table 1. Demographic and clinical characteristics of chronic hepatitis B patients with concurrent hepatic steatosis

Characteristics	Outcome
Male, n/total (%)	697/926 (75.27)
Age (years)	38.00 (31.00–46.00)
Body mass index (kg/m^2)	25.47 (23.38–27.74)
Diabetes mellitus, n/total (%)	80/926 (8.64)
Hypertension, n/total (%)	103/926 (11.12)
White blood cell (10^9/L)	5.57 (4.69–6.60)
Neutrophils (10^9/L)	3.05 (2.46–3.87)
Red blood cell (10^{12}/L)	4.99 (4.62–5.28)
Hemoglobin (g/L)	151.00(140.00–160.00)
Platelet (10^9/L)	195.00 (160.00–236.00)
Total bilirubin (umol/L)	14.10 (11.10–18.25)
Albumin (g/L)	44.50 (41.60–47.30)
Globulin (g/L)	27.50 (25.10–30.50)
Alanine transaminase (U/L)	44.75 (28.00–85.85)
Aspartate transaminase (U/L)	30.00 (23.00–49.25)
Alkaline phosphatase (U/L)	71.05 (58.00–90.00)
Gamma-glutamyl transferase (U/L)	34.00 (22.00–55.00)
Lactate dehydrogenase (U/L)	179.50 (160.00–204.25)
Creatinine (umol/L)	66.96±13.94
Urea (umol/L)	345.72 ± 87.22
Total Cholesterol (mmol/L)	4.58 (4.02–5.13)
Triglycerides (mmol/L)	1.23 (0.91–1.77)
Glucose (mmol/L)	5.25 (4.77–5.83)
High density lipoprotein cholesterol (mmol/L)	1.17 (0.97–1.39)
Low density lipoprotein cholesterol (mmol/L)	2.70 ± 0.75
Prothrombin time (s)	12.40 (11.10–13.20)
International normalized ratio	1.00 (0.94–1.04)
Steatosis grade, n/total (%)	
1	752/926 (81.21)
2	126/926 (13.61)
3	48/926 (5.18)
Fibrosis grade, n/total (%)	
0	29/926 (3.13)
1	464/926 (50.11)
2	253/926 (27.32)
3	129/926 (13.93)
4	51/926 (5.51)

patients had advanced fibrosis (≥S3). The scores of FIB-4, APRI, and NFS were calculated based on obtainable laboratory parameters. At the lower cut-off values (FIB-4 <1.3, APRI ≤0.5, NFS <1.455), APRI had the highest sensitivity at 65.56%, followed by 53.89% for FIB-4 and 41.11% for NFS. At the higher cut-off values (FIB-4 >2.67, APRI >1.5, NFS >0.676), NFS had the highest specificity of 97.99%, followed by FIB-4 (94.77%) and APRI (94.1%).

A total of 649 patients with CHB concurrent with HS, with a FIB-4 cutoff <1.3 were classified as non-advanced fibrosis, and 566 (61.12%) of the patients coincided with the biopsy results. Of the 695 patients with an NFS cutoff <−1.455, classified as non-advanced fibrosis, 589 (63.61%) had results consistent with the biopsy results of non-advanced fibrosis. Notably, APRI displayed the highest indeterminate rate of 30.99% compared to the other NITs. The proportions of patients who were correctly classified according to a single test were 64.7% for NFS, 64.1% for FIB-4, and 57.6% for APRI (Fig. 1).

To further improve the performance of the diagnostic



NITs	AUROC	SE	SP	PPV	NPV	TN FN	FP TP	Indeterminate
APRI								
≤0.5	0.661	65.56	66.62	32.2	88.9	497	44	287
>1.5	0.571	20	94.1	45	83	62	36	
FIB-4								
<1.3	0.649	53.89	75.87	35	87.2	566	39	210
>2.67	0.552	15.56	94.77	41.8	82.3	83	28	
NFS								
<-1.455	0.6	41.11	78.95	32	84.7	589	15	206
>0.676	0.518	5.56	97.99	40	81.1	106	10	
APRI-FIB-4								
≤0.5→<1.3	0.655	48.89	82.17	39.80	87.00	613	60	116
>1.5→>2.67	0.585	25.00	91.96	42.90	83.60	97	40	
FIB-4-APRI								
<1.3→≤0.5	0.644	46.11	82.71	39.20	86.40	617	56	116
>2.67→>1.5	0.574	22.22	92.49	41.70	83.10	97	40	
FIB-4-NFS								
<1.3→<-1.455	0.611	37.78	84.45	37	84.9	630	45	109
>2.67→>0.676	0.553	16.67	93.97	40	82.4	112	30	
NFS-FIB-4								
<-1.455→<1.3	0.608	36.11	85.52	37.60	84.70	638	37	109
>0.676→>2.67	0.550	15.00	95.04	42.20	82.30	115	27	
APRI-NFS								
≤0.5→<-1.455	0.630	40.56	85.52	40.30	85.60	638	49	93
>1.5→>0.676	0.575	21.67	93.43	44.30	83.20	107	39	
NFS-APRI								
<-1.455→≤0.5	0.602	32.22	88.20	39.70	84.40	658	29	93
>0.676→>1.5	0.547	13.33	96.11	45.30	82.10	122	24	

Fig. 1. Comparison of different non-invasive tests and combined model for predicting advanced fibrosis in chronic hepatitis B patients with concurrent hepatic steatosis. APRI, aspartate aminotransferase to platelet ratio index; AUROC, area under receiver operating characteristic; FIB-4, fibrosis index based on the four factors; FN, false negative; FP, false positive; NFS, nonalcoholic fatty liver disease fibrosis score; NITs, non-invasive tests; NPV, negative predicted values; PPV, positive predicted values; TN, true negative; TP, true positive; SE, sensitivities; SP, specificities.

model and reduce the indeterminate rate, we combined the three NITs pairwise. The algorithm, including NFS followed by APRI, allowed most patients to be correctly classified (73.7%), compared to FIB-4-NFS (71.3%), NFS-FIB-4 (71.8%), FIB-4-APRI (71.0%), APRI-FIB-4 (70.5%), APRI-NFS (71.8%), which were better than the three NITs used alone. In addition, as shown in Figure 1, the model which combined APRI and NFS achieved the lowest percentage of indeterminate cases at 10.04%.

In this study, we found that the combined models overall showed better performance than the individual NITs in CHB patients with concurrent HS. Moreover, the NFS-APRI model achieved the lowest indeterminate zone among the six combined models. Although FIB-4, APRI, and NFS have been widely utilized for detecting advanced fibrosis,[10] none of these NITs were completely suitable for CHB patients with concurrent HS. Additionally, the accuracy of these scoring systems might be affected by factors such as age, sex, ethnicity, and comorbidities. FIB-4, the most commonly used test for fibrosis stratification in HS patients had a relatively lower positive predictive value and a higher indeterminate rate.[11] Furthermore, a population-based study[12] reported that FIB-4 and NFS misclassified more than 30% of patients, leading to a non-negligible percentage of false-negatives and false-positives, especially in patients with risk factors for chronic liver disease (such as obesity and diabetes). Lin KM *et al*.[13] investigated the diagnostic accuracy of FIB-4, NFS and APRI for identifying advanced fibrosis in CHB patients with concurrent HS, and demonstrated that these NITs were better at excluding rather than including advanced fibrosis, which is consistent with our results. A previous study showed that a simple serum-based score may fall into the given test's indeterminate zone (a high uncertainty area ranging from 40% for FIB-4 to 55.8% for NFS).[14] Thus, considering the suboptimal performance of individual NITs, researchers have started using a sequential strategy to screen for fibrosis. For instance, a Danish cohort study combined FIB-4 with the Enhanced Liver Fibrosis test, to achieve a correct classification rate of 88% and reduced indeterminate zones.[15] Our study also provided evidence supporting the utilization of a sequential strategy. Given the easily-available laboratory parameters used in NITs, a sequential strategy could promote diagnostic accuracy and optimize referral pathways. Additionally, the combination of models developed during our study can be of great assistance in primary care settings. With the ability to provide targeted screening for liver fibrosis, the findings from this study will enable healthcare professionals to make informed decisions regarding treatment options and predict potential clinical outcomes. Identifying patients who may be at risk of developing liver fibrosis is a crucial step in the early detection and treatment of this condition. By implementing these models, we can ensure that patients receive the care they need and deserve. The potential benefits of utilizing these models are vast, from improving patient outcomes to reducing the burden on healthcare systems. Overall, the use of our combined models has the potential to revolutionize the way in which liver fibrosis is diagnosed and treated in primary care settings.

Our study cohort involving multiple centers provides well characterized clinical data, including liver histology information representing the current gold standard reference for comparison to the NITs. However, some limitations should be acknowledged. First, as this was a retrospective study, we did not have imaging test data for some of our subjects. Second, all the patients were recruited from tertiary centers and may not be fully representative of community-dwelling patients, which limits the generalizability of our findings to other populations.

In conclusion, the combined model of NFS-APRI developed in our study showed better performance than individual NITs. The model could reduce indeterminate zones and optimize referral pathways, highlighting it as a useful tool for targeted screening for liver fibrosis in patients afflicted with both CHB and HS. Additional research is needed to validate our study findings and extend our understanding of the clinical utility of combined NITs models, particularly in cases of CHB with concurrent HS. Further studies are needed to provide valuable insights into the efficacy of this approach, and pave the way for more widespread adoption of these models in clinical practice.

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Conflict of interest

JL has been an Editorial Board Member of *Journal of Clinical and Translational Hepatology* since 2024. The other authors have no conflict of interests related to this publication.

Author contributions

All authors contributed to this study at different levels. Study concept and design: JL and JS. Acquisition of data: XX, FR, and WN. Statistical analysis and interpretation of data: XX, FR, and WN. Drafting of the manuscript: XX and FR. Critical revision of the manuscript for important intellectual content: JL, JS and CW.

Ethical statement

This study has received approval from the Institutional Ethics Review Board of all the involved hospitals, with document numbers of 2008022 and NCT05766449. This study was carried out in accordance with the Declaration of Helsinki. As this was a retrospective study, the requirement for informed consent was waived.

Data sharing statement

We are unable to provide access to our dataset for privacy reasons. The protocol and statistical analysis methods used in the study can be requested directly from the corresponding author after approval.

References

[1] Li J, Zou B, Yeo YH, Feng Y, Xie X, Lee DH, *et al*. Prevalence, incidence, and outcome of non-alcoholic fatty liver disease in Asia, 1999-2019: a systematic review and meta-analysis. Lancet Gastroenterol Hepatol 2019;4(5):389–398. doi:10.1016/s2468-1253(19)30039-1, PMID:30902670.

[2] Zheng Q, Zou B, Wu Y, Yeo Y, Wu H, Stave CD, *et al*. Systematic review with meta-analysis: prevalence of hepatic steatosis, fibrosis and associated factors in chronic hepatitis B. Aliment Pharmacol Ther 2021;54(9):1100–1109. doi:10.1111/apt.16595, PMID:34469587.

[3] Thin KN, Tran A, Li J, Lee EY, Yang H, Rui F, *et al*. Increased Risk of Liver-Related Outcomes in Chronic Hepatitis B Patients with Metabolic Syndrome: A Systematic Review and Meta-Analysis. Dig Dis 2022;40(6):745–753. doi:10.1159/000521768, PMID:34986486.

[4] Xiao G, Yang J, Yan L. Comparison of diagnostic accuracy of aspartate aminotransferase to platelet ratio index and fibrosis-4 index for detecting liver fibrosis in adult patients with chronic hepatitis B virus infection: a systemic review and meta-analysis. Hepatology 2015;61(1):292–302. doi:10.1002/

hep.27382, PMID:25132233.
[5] Lazarus JV, Castera L, Mark HE, Allen AM, Adams LA, Anstee QM, *et al*. Real-world evidence on non-invasive tests and associated cut-offs used to assess fibrosis in routine clinical practice. JHEP Rep 2023;5(1):100596. doi:10.1016/j.jhepr.2022.100596, PMID:36644239.
[6] Xiao G, Zhu S, Xiao X, Yan L, Yang J, Wu G. Comparison of laboratory tests, ultrasound, or magnetic resonance elastography to detect fibrosis in patients with nonalcoholic fatty liver disease: A meta-analysis. Hepatology 2017;66(5):1486–1501. doi:10.1002/hep.29302, PMID:28586172.
[7] EASL Clinical Practice Guidelines: management of chronic hepatitis B. J Hepatol 2009;50(2):227–242. doi:10.1016/j.jhep.2008.10.001, PMID:19054588.
[8] Terrault NA, Bzowej NH, Chang KM, Hwang JP, Jonas MM, Murad MH. AASLD guidelines for treatment of chronic hepatitis B. Hepatology 2016;63(1):261–283. doi:10.1002/hep.28156, PMID:26566064.
[9] Scheuer PJ. Classification of chronic viral hepatitis: a need for reassessment. J Hepatol 1991;13(3):372–374. doi:10.1016/0168-8278(91)90084-o, PMID:1808228.
[10] Mózes FE, Lee JA, Selvaraj EA, Jayaswal ANA, Trauner M, Boursier J, *et al*. Diagnostic accuracy of non-invasive tests for advanced fibrosis in patients with NAFLD: an individual patient data meta-analysis. Gut 2022;71(5):1006–1019. doi:10.1136/gutjnl-2021-324243, PMID:34001645.
[11] Tincopa MA, Loomba R. Non-invasive diagnosis and monitoring of non-alcoholic fatty liver disease and non-alcoholic steatohepatitis. Lancet Gastroenterol Hepatol 2023;8(7):660–670. doi:10.1016/s2468-1253(23)00066-3, PMID:37060912.
[12] Graupera I, Thiele M, Serra-Burriel M, Caballeria L, Roulot D, Wong GL, *et al*. Low Accuracy of FIB-4 and NAFLD Fibrosis Scores for Screening for Liver Fibrosis in the Population. Clin Gastroenterol Hepatol 2022;20(11):2567–2576.e2566. doi:10.1016/j.cgh.2021.12.034, PMID:34971806.
[13] Lin KW, Kumar R, Shen F, Chan HL, Wong GL, Kumar R, *et al*. The utility of non-invasive tests to assess advanced fibrosis in Asian subjects with chronic hepatitis B and concomitant hepatic steatosis. Liver Int 2023;43(5):1008–1014. doi:10.1111/liv.15541, PMID:36855842.
[14] Pennisi G, Enea M, Falco V, Aithal GP, Palaniyappan N, Yilmaz Y, *et al*. Noninvasive assessment of liver disease severity in patients with nonalcoholic fatty liver disease (NAFLD) and type 2 diabetes. Hepatology 2023;78(1):195–211. doi:10.1097/hep.0000000000000351, PMID:36924031.
[15] Kjaergaard M, Lindvig KP, Thorhauge KH, Andersen P, Hansen JK, Kastrup N, *et al*. Using the ELF test, FIB-4 and NAFLD fibrosis score to screen the population for liver disease. J Hepatol 2023;79(2):277–286. doi:10.1016/j.jhep.2023.04.002, PMID:37088311.