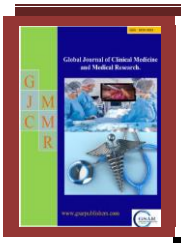

Global Journal of Clinical Medicine and Medical Research [GJCMMR]
ISSN: 2583-987X (Online)
Abbreviated key title: Glob.J.Clinic.Medici.Medica.Res.
Frequency: Monthly
Published By GSAR Publishers
Journal Homepage Link- https://gsarpublishers.com/journal-gjcmmr-home/

Evaluation of Non-Invasive Markers to Show Fibrosis in Patients with Chronic Hepatitis B

BY

Mahmut Çınar[1], Deniz İncaman[1], Mehmet Bayram[2], İskender Ekinci[3], Kader Irak[4], Ömür Tabak[4], Abdulbaki Kumbasar[5],

[1]Kastamonu Univercity, Faculty of Medicine, Internal Medicine Clinic
[2]Türkiye Hospital, Gastroenterology Clinic
[3]Bezmialem Vakif University, Internal Medicine Clinic
[4]Health Science Univercity, İstanbul Kanuni Sultan Süleyman Training and Research Hospital, Internal Medicine Clinic
[5]Health Science Univercity Bakırköy Sadi Konuk Training and Research Hospital, Internal Medicine Clinic

Article History

Received: 07/07/2024
Accepted: 12/07/2024
Published: 14/07/2024

Vol – 2 Issue – 7

PP: -07-11
DOI:
10.5281/zenodo.12739724

Abstract

Introduction: *Liver biopsy is the gold standard in the evaluation of fibrosis in the treatment plan for hepatitis B, which is an important public health problem in the world and our country. There are disadvantages such as the development of complications in the biopsy procedure and the inability to represent the liver tissue. Therefore, non-invasive, objectively evaluated, and reproducible methods that can replace biopsy are being investigated.* ***Material-Method:*** *201 patients aged 18 and over, who were diagnosed with chronic hepatitis B and applied to the internal medicine and gastroenterology outpatient clinics between January 2014 and December 2019, were included in the study.* ***Results:*** *When the diagnostic sensitivity of non-invasive markers according to the 7 tools of fibrosis was investigated in our study, it was seen that the best diagnostic test in the ≥F2 group was the King's score.* ***Conclusion:*** *Most of the non-invasive tests we evaluated can predict significant fibrosis with apparent accuracy, and with the help of non-invasive tests, the rate of unnecessary biopsies can be reduced.*

Keywords: *Hepatitis B virus, Fibrotic stage, Liver biopsy.*

INTRODUCTION

Hepatitis B virus (HBV) infection is a global public health problem due to its incidence, contagiousness, chronicity, and the development of cirrhosis and hepatocellular carcinoma. According to the 2015 data of the World Health Organization, 257 million people in the world are infected with HBV and approximately 65 million of these people are women of childbearing age (1). According to 2015 WHO data, the global prevalence of HBV was found to be 3.5% (2).

Our country is located in the middle endemic region in terms of HBV, and according to a study conducted in 2009, HBsAg positivity in the adult age group was found to be 4% in our country (3). Biochemical, serological, and molecular tests, histopathological examination, and imaging methods are used in the diagnosis, staging, treatment decision, and follow-up of chronic hepatitis B (CHB) infection (4). Although the gold standard method of CHB diagnosis is liver biopsy, there are problems such as the invasiveness of this method, the need for hospitalization, and the risk of complications after the procedure. In addition, since liver biopsy samples only 1/50 000 of the whole liver, it cannot represent the whole liver, and sampling errors can be made up to 20-30% (5).

In addition to biopsy, many non-invasive methods that show liver fibrosis are also in use. In recent years, non-invasive, easy, inexpensive, reproducible methods that can replace liver biopsy, show liver fibrosis, and perform staging have been investigated. While some of the non-invasive serum markers are frequently repeated tests in routine practice, some of them require additional study kits. 2 involves expensive, infrequently used, blood biochemical tests that cannot be studied in large patient populations (6).

Our aim in this study is to evaluate the usability of these non-invasive markers of hepatic fibrosis in chronic hepatitis b patients.

MATERIALS AND METHOD

The CHB patients older than 18 age and who were admitted to the internal medicine and gastroenterology outpatient clinics of Health Sciences University, Istanbul Kanuni Sultan

*Corresponding Author: **Mahmut Çınar**

Suleyman Training and Research Hospital between January 2014 and December 2019 were included in the study. These individuals were patients who had HBsAg positivity in serum for at least six months and had liver biopsy retrospectively.

The following laboratory data of the patients were obtained from the patient's records: blood count, ALT, AST, alkaline phosphatase (ALP), gamma-glutamyl transferase (GGT), total bilirubin, albumin, platelets, international normalized ratio (INR), prothrombin time (PTZ), hepatitis-B virus DNA (HBV-DNA), Hepatitis-B e antigen (HBeAg), Hepatitis-B surface antigen (HbsAg) Patients with decompensated liver cirrhosis, patients with a history of malignancy, hepatitis B carrier as well as other viral hepatitis diagnoses, autoimmune hepatitis, primary biliary cholangitis, patients who did not have sufficient data in the retrospectively scanned files and laboratory tests, and patients who were pregnant were excluded from the study.

Histological examination: In liver biopsy classification, histological activity is defined as "Histological activity index (HAI) - "grade" and calculated by summing the score given for each parameter. According to this system, periportal bridging necrosis, intralobular degeneration focal necrosis, portal inflammation, and fibrosis are evaluated (8). The sum of the numerical values obtained from the evaluation of the first three was determined as the histological activity index (HAI) and indicates the severity of inflammation in the liver. The maximum score is 18. Fibrosis levels are evaluated as 0,1,2,3,4,5 and 6. When fibrosis is ≥5, this indicates cirrhosis (9).

Non-invasive indexes: The non-invasive indexes used in this study are explained below and presented in detail in Table 1.

Calculated model name	Parameters that are used	Formula
APRI "AST/Platelet Ratio Index	AST, platelet count	[AST (IU/L)/ULN (IU/L)/PLT (109/L)] × 100
FIB-4 index	AST, ALT, platelet count, age	[Age (years) × AST (IU/L)] / [PLT (109/L) × ALT1/2 (IU/L)]
AST-ALT ratio (AAR)	AST, ALT	AST (IU/L)/ALT (IU/L)
King's score	Age, AST, PTZ (INR), platelet count	Age(years) × AST (IU/L) × INR/ PLT (109/L)
GUCI "Göteborg University Cirrhosis Index	AST, PT (INR), platelet count	AST (IU/L)/ULN (IU/L) × INR × 100/ PLT (109/L)
Age-platelet index (API)	Age, platelet count	Age <30=0; 30-39=1; 40-49=2; 50-59=3; 60-69 =4; ≥70=5 Platelet count (× 109 L−1): ≥225=0; 200–224=1; 175–199=2; 150–174=3; 125–149=4; API: Sum of age and platelet count score (a value between 0-10)
Abbreviations: APRI: , FIB-4: AAR:, GUCI, API: , AST:, ALT: , PTT, INR:		

Aspartate transaminase-platelet ratio index (APRI); aspartate transaminase (AST) and platelet is routinely used laboratory parameters in the follow-up of chronic hepatitis B patients. This ratio is simple, non-invasive, and predicts liver fibrosis (7).

The fibrosis index based on four factors (FIB-4) is calculated with the following formula: FIB-4 Score = (Age* x AST) / (Platelets x √(ALT)). The interpretation of the obtained score is done as below: FIB-4 score <1.45 represents approximate fibrosis stage 0-1, FIB-4 score 1.45-3.25 represents approximate fibrosis stage 2-3 and FIB-4 score >3.25 represents approximate fibrosis stage 4-6 (8).

Aspartate transaminase alanine transaminase ratio (AAR) has been widely used to distinguish between alcoholic and non-alcoholic hepatic causes of serum aminotransferase elevations (9).

King's score; is calculated using age, AST, INR, and Platelet count (9).

Göteborg University Cirrhosis Index (GUCI); It is a calculation using AST, PTZ, INR, and platelet count.

Age Score+Platelet score (API); (Age <30=0; 30–39=1; 40–49=2; 50–59=3; 60–69=4; ≥70=5) (Platelet count (×109 L−1): ≥225=0; 200–224=1; 175–199=2; 150–174=3; 125–149=4; <125=5) and a value between 0-10 emerges.

Data collection: The age and gender of the patients were recorded. The HAI rating and fibrosis stage were obtained from the liver biopsy results. The scores of the non-invasive tools including APRI, FIB-4, AAR, Kings Score, GUCI, and API were calculated.

Statistical analysis: The IBM SPSS statistic 25 program was used for statistical analysis of the data. While evaluating the

*Corresponding Author: **Mahmut Çınar**

study data, descriptive statistics (mean, standard deviation, minimum, maximum, median, first quartile, third quartile, frequency, and percentage) were presented. For continuous dependent variables, the difference was examined according to the independent variable (when there were 2 groups), while the independent group's t-test was used when the data of both groups were normally distributed, and the Mann-Whitney U test, which is one of the nonparametric statistics, was used when deviation from the normal distribution was observed. Pearson chi-square test was used to compare two categorical data. ROC (Receiver Operating Curves) analysis was performed to determine the levels and cut-off values of non-invasive indices in the prediction of moderate Fibrosis (≥2), significant Fibrosis (≥3) advanced Fibrosis (≥4), and HAI (≥6). Youden Index was calculated using the sensitivity and specificity indices obtained by ROC analysis. The highest score of the Youden index was considered a cut-off. Statistical significance was accepted as $p<0.05$.

Table 1: Non-invasive indexes of liver fibrosis

Abbreviations: APRI: AST /Platelet Ratio index, FIB-4 Fibrosis index, AAR: AST/ALT ratio, GUCI: göteborg University cirrhosis index, API: Age/platelet index, AST: aspartate aminotransferase, ALT: alanine aminotransferase, INR: İnternational normalized ratio

RESULTS

Among the patients, 85 (42.3%) were female and 116 (57.7%) were male. The ages of the cases ranged from 20 to 79, and the mean age of the patients was 42.60±12.42. There were 130 (64.7%) cases with HAI grades above 6 and 25 (12.4%) cases with F>3. The HBsAg positivity and biopsy results (HAI and Fibrosis stages) of the patients are presented in Table 2.

In the correlation analysis between non-invasive indexes and liver fibrosis stage, there was a significantly positive correlation between APRI, GUCI, King score values, and fibrosis stage. In the correlation analysis between non-invasive biomarkers and HAI scores, there was a significantly positive correlation between HAI score and APRI, GUCI, and King's score while a significantly negative correlation was found between HAI score and AAR score. The correlation analysis in detail is presented in Table 3.

The data including the HBsAg positivity, HAI score, and fibrosis stages of the patients obtained through the liver biopsy is presented in Table 2.

Table 2: HBsAg positivity, HAI rating, and Fibrosis stages of the patients

Age	Minimum-Maximum Mean±sd	20-79 42.60±12.42
Gender	Women n(%) Male n(%)	85 (%42,3) 116 (%57,7)
Hbsag	Positive n(%) 37 (18.4%) Negative n(%) 142 (70.6%) Lost data 22 (10.9)	37 (%18,4) 142 (%70,6) 22 (10.9)
HAI rating (total)	Minimum – Maximum Mean±sd	2 – 14 6.00±2.23
HAI rating	<6 n(%) >6 n(%)	71 (%35,3) 130 (%64,7)
Fibrosis stage (total)	Minimum – Maximum Mean±sd	0 – 6 1.42±1.08
Fibrosis stage	<2 n(%) >2 n(%)	118 (%58,7) 83 (41.3)
Fibrosis stage	<3 n(%) >3 n(%)	176 (%87,6) 25 (%12,4)
Fibrosis stage	<4 n(%) >4 n(%)	192 (%95,5) 9 (%4,5)
Fibrosis stage	<5 n(%) >5 n(%)	198 (%98,5) 3 (%1,5)
Fibrosis=0	n(%)	36 (%17,9)
Fibrosis=1	n(%)	82 (%40,8)
Fibrosis=2	n(%)	58 (%28,9)
Fibrosis=3	n(%)	16 (%8,0)
Fibrosis=4	n(%)	6 (%3,0)
Fibrosis=5	n(%)	2 (%1,0)
Fibrosis=6	n(%)	1 (%,5) 2

The laboratory results of the patients are presented in Table 3.

Table 3: Laboratory analysis of the patients

	N	Minimum	Maximum	Mean	Standard deviation
INR	201	,85	2.16	1.048	,152
GGT	192	,23	339.00	29.3449	35.031
ALP	187	11	248	78.73	30.316

*Corresponding Author: **Mahmut Çınar**

HBV DNA	199	0	20160315109601	101408823920.68	1429119795204.870
Platelet	201	62	398	232.32	60.805
Albumin	182	3.30	8.40	4.5482	,43789
AST	201	11	659	43.72	70.655
ALT	201	8	1149	66.12	120.148

*p <0.05

Diagnostic measures of non-invasive fibrosis markers were obtained by ROC analysis to predict fibrosis stage >2 (moderate fibrosis). A cut-off value of King's score for F2 estimation was determined as 7,503 (sensitivity=38.6% and specificity=82.2%). AAR and AP indices were found to be insufficient to predict >F2 (p>0.05).

Diagnostic measures of non-invasive fibrosis markers were obtained by ROC analysis to predict fibrosis stage > F3 (significant fibrosis).

	AUC (95% CI)	p	Cut-off index	Sensitivity (%)	Specificity (%)	Youden index
APRI	0.654 (.542-.766)	.013*	.344	76.0	58.5	0.345
FİB4	,537 (,413-660)	,554	-	-	-	-
AAR	,630 (,521-,738)	,036*	,911	67,6	56,0	0,236
AP	,544 (,414-,674)	,473	-	-	-	-
GUCİ	,666 (,558-,774)	,007*	,356	76,0	61,4	0,374
KINGSCORE	,641 (,525-,756)	,023*	5,608	64,0	65,9	0,299

*p <0.05

Diagnostic measures of non-invasive fibrosis markers were obtained by ROC analysis to predict fibrosis stage > F3 and HAI scores higher than 6 are presented in Table 5. When the diagnostic sensitivity of non-invasive markers was investigated according to the fibrosis stage, it was seen that the indices with significant predictive value for HAI >6 were APRI, AAR, GUCI, and KING SCORE. The GUCI index has the highest predictive power for the prediction of HAI >6 and the cut-off value for GUCI was 0.353 (sensitivity=56.9% and specificity=78%, 9, AUC=, 680; p=.000; CI=.608-.752). Since the AAR index is negatively associated with fibrosis, an increase in HAI was associated with a decrease in AAR. That is, patients with a value below the cut-off value (0.756) for AAR have a HAI >6 level of 90.1%. According to the Mann-Whitney U Test results, it was observed that the groups formed according to HAI<6 and HAI≥6 did not make a significant difference in terms of FIB4 and AP indexes (p>0.05). However, APRI, AAR, GUCI, and King score values differ significantly according to the HAI value (p<0.05). When the median values of these significantly different indices are examined in both groups, it can be concluded that the APRI, AAR, GUCI, and King score values of the group with HAI≥6 are significantly higher than the values of the group with HAI<6.

Table 5. ROC analysis for non-invasive parameters to predict F>3 and HAI>6

	AUC (95% CI)		P	cut-off index	Sensitivity (%)	Specificity (%)	Youden index
APRI	,668	(,595-,741)	,000*	,441	43,1	90,1	0,332
FİB4	,545	(,462-,627)	,462	-	-	-	-
AAR	,689	(,616-,763)	,000*	,756	90,1	41,5	0,317
AP	,453	(,370-,536)	,270	-	-	-	-
GUCİ	,680	(,608-,752)	,000*	,353	56,9	78,9	0,358
KINGSCORE	,665	(,592-,738)	,000*	6,00	46,2	85,9	0,321

*p <0.05

Discussion: In this study, it was found that the non-invasive markers are closely related to the liver biopsy findings, as expected. We showed that these parameters have a significant correlation with both necroinflammatory activity and fibrosis scores and that these parameters are good predictors of both fibrosis and inflammation. APRI, GUCI, and King scores were positively correlated with both F stage and HAI scores while AAR was negatively correlated with HAI score. In

*Corresponding Author: **Mahmut Çınar**

addition, APRI, AAR, GUCI, and King scores significantly predicted both the fibrosis score >3 and HAI score >6.

Significant fibrosis was detected in 9.4% of female patients and 14.7% of male patients included in our study. Age was not found to be statistically significant in the significant fibrosis group, this result was not consistent with the results reported in the literature (8). The reason for different results from previous studies can be explained by the fact that we have a small number of significant fibrosis patients. The number of cases with fibrosis grade below 3 was 176 (87.6%), and the number of cases 3 and above was 25 (12.4%). It was considered that biopsies performed in 41.3% and 87.6% of patients, respectively, in the detection of moderate fibrosis (>F2) and significant fibrosis (>F3), could predict fibrosis with the help of non-invasive tests. According to gender, the percentage of males (HAI grade >6) was statistically significantly higher than females.

When we analyzed the laboratory results according to the fibrosis stage, it was seen that the AST, AST, and GGT grades were statistically significantly higher in the group with significant hepatic fibrosis. It was considered that the reason why no difference could be observed between INR and albumin values was due to the small number of patients in our significant fibrosis group and the very small number of patients with F5 and above.

The best diagnostic test in the F4 group was GUCI (AUC=666). When the diagnostic sensitivity of noninvasive markers was investigated according to the fibrosis stage, it was seen that the indices with significant predictive value in the >F4 group were AP (AUC=0.709) and KING SCORE (AUC=0.711). APRI, FIB4, AAR, and GUCI indices were found to be insufficient to predict > F4. For HAI >6, the AUROC values of the indices in our study were as follows: GUCI index:0.680, AAR:689 APRI:668 KING SCORE:665. On the other hand, FIB4 and AP indices were insufficient to predict HAI≥6 levels.

The AUROC of the FIB-4 index, which was first developed by Sterling et al. in 2006 by researching HCV-HIV co-infected patients, and also recommended in the WHO CHB guidelines, was found to be 0.765 in estimating fibrosis stage > F4 (11). In our study, we found the AUROC value of 0.537 to indicate significant hepatic fibrosis, and AUROC:0.671 for >F4, and it was also seen as AUROC:0.545 to indicate significant hepatic necroinflammation, and we encountered a lower AUROC value than the literature.

When the diagnostic sensitivity of non-invasive markers was investigated according to the necroinflammation stage, it was seen that the index with the highest predictive power for HAI >6 was GUCI (AUC=0.680). In our study, when the diagnostic sensitivity of non-invasive markers was investigated according to the fibrosis stage, it was seen that the non-invasive markers that were significant in the diagnosis of >F2 moderate fibrosis were APRI, FIB4, GUCI, and KING SCORE, and the King's score was the best diagnostic test in the >F2 group. Significant non-invasive markers in the diagnosis of significant fibrosis were found to be APRI, AAR, GUCI, and KING SCORE. > GUCI was found to be the best diagnostic test in the F3 group. It was seen that GUCI had the highest predictive power for HAI >6 estimation. Based on these results, we think that non-invasive markers will replace invasive markers in the future. In conclusion, we think we have contributed to the Literature, but time is still needed before the biopsy takes its place.

Result: Based on the results of our study, we suggest that GUCI and King scores should be evaluated in the estimation of fibrosis. In the prediction of necroinflammation, we can recommend the GUCI scoring system. However, it should not be forgotten that; Today, an excellent biochemical serum marker that can be used instead of liver biopsy has not yet been found and standardized.

Ethics Committee Approval: On 09.12.2019, the approval of the Health Sciences University Istanbul Bakırköy Dr. Sadi Konuk Training and Research Hospital Ethics Committee (with No 2019/24/06) was obtained.
Informed Consent: Informed consent was obtained from the participants.

Author Contributions: Working Concept/Design- M.Ç, M.B, K.I; Data Collection- M.C; Data Analysis/Interpretation- İ.E., A.K. Manuscript Draft- İ.E., D.İ; Critical Review of Content- İ.E., A.K; Final Approval and Responsibility- M.Ç.; Material and Technical Support- M.Ç, D.İ.; Supervision A.K., Ö.T.

Conflict of Interest: The authors declared no conflict of interest.
Financial support: There is no financial support for this study.

*Corresponding Author: **Mahmut Çınar**