Clinical usefulness of Mac-2 binding protein glycosylation isomer for diagnosing liver cirrhosis and significant fibrosis in patients with chronic liver disease
A retrospective single-center study

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Abstract

Accurate diagnosis of liver cirrhosis (LC) and significant fibrosis in patients with chronic liver disease (CLD) is important. The Mac-2 binding protein glycosylation isomer (M2BPGi) has emerged as a novel serum biomarker for liver fibrosis; however, insufficient clinical data of M2BPGi are available in patients with CLD. Therefore, we performed a retrospective cohort study to investigate the clinical usefulness of serum M2BPGi for assessing LC and significant fibrosis in CLD patients. We retrospectively reviewed the CLD patients with measured serum M2BPGi at Kosin University Gospel Hospital between January 2016 and December 2019. Multivariate logistic regression analyses were conducted to identify the independent factors associated with LC. The diagnostic power of serum M2BPGi for LC and significant fibrosis (≥F2) was evaluated and compared to that of other serum biomarkers using receiver operating characteristic curve and area under the curve (AUC). A total of 454 patients enrolled in this study. M2BPGi (adjusted odds ratio [aOR], 1.77; 95% confidence interval [CI], 1.52–2.07) and fibrosis index based on four factors (aOR, 1.23; 95% CI, 1.11–1.37) were identified as significant independent factors for LC. The AUC of M2BPGi for LC (0.866) and significant fibrosis (0.816) were comparable to those of fibrosis index based on four factors (0.860, 0.773), aspartate aminotransferase-to-platelet ratio index (0.806, 0.752), and gamma-glutamyl transpeptidase-to-platelet ratio (0.759, 0.710). The optimal cut-off values for M2BPGi for LC and significant fibrosis were 1.37 and 0.89, respectively. Serum M2BPGi levels were significantly correlated with liver stiffness measurements ($\rho = 0.778$). Serum M2BPGi is a reliable noninvasive method for the assessment of LC and significant fibrosis in patients with CLD.

Abbreviations: ALD = alcoholic liver disease, ALT = alanine aminotransferase, aOR = adjusted odds ratio, APRI = aspartate aminotransferase-to-platelet ratio index, AST = aspartate aminotransferase, AUC = area under the curve, CLD = chronic liver disease, COI = cut-off index, FIB-4 = fibrosis index based on four factors, GPR = gamma-glutamyl transpeptidase-to-platelet ratio, HBV = hepatitis B virus, HCV = hepatitis C virus, HSCs = hepatic stellate cells, INR = international normalized ratio, LC = liver cirrhosis, LSM = liver stiffness measurement, M2BPGi = Mac-2 binding protein glycosylation isomer, NAFLD = nonalcoholic fatty liver disease.

Keywords: biomarkers, chronic hepatitis, fibrosis, liver cirrhosis, Mac-2 binding protein glycosylation isomer

1. Introduction

Chronic liver disease (CLD) is a serious health issue worldwide because it can lead to liver fibrosis and cirrhosis.[1,2] Causes of CLD include hepatitis B virus (HBV) and hepatitis C virus (HCV) infections, alcoholic liver disease (ALD), nonalcoholic fatty liver disease (NAFLD), and autoimmune hepatitis (AIH). Liver cirrhosis (LC), the end stage of fibrosis, leads to increased

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morbidity and mortality due to various complications such as gastrointestinal bleeding, encephalopathy, ascites, hepatorenal syndrome, or development of hepatocellular carcinoma.[3,4] Accurate diagnosis of LC and liver fibrosis in patients with CLD is clinically important, since management and prognosis are mainly dependent on it.[5–7] However, diagnosing LC and liver fibrosis is still challenging.

Liver biopsy is considered as the gold-standard for diagnosing LC and liver fibrosis[8]; however, it is invasive and painful, with potential complications, thereby making it difficult to repeat the procedure multiple times in clinical practice.[8,9] Moreover, liver biopsy has several limitations, such as interobserver variability and sampling errors.[10,11] Therefore, many noninvasive methods have been developed to assess LC and liver fibrosis.[12–14] Conventional serum markers using routine laboratory parameters include the fibrosis index based on four factors (FIB-4), aspartate aminotransferase-to-platelet ratio index (APRI), and gamma-glutamyl transpeptidase-to-platelet ratio (GPR). Furthermore, these serum markers have been extensively reported to accurately predict LC; however, it is inconvenient to calculate their values using formulas.[12–16] Transient elastography (FibroScan®; Echosens, Paris, France) is a validated noninvasive ultrasonography-based method and is the most widely used for diagnosing LC and fibrosis in patients with various CLD etiologies.[17–21] Nevertheless, Fibroscan® (Echosens) cannot be performed on certain patients with morbid obesity or ascites, and the results may not be reliable in cases of acute hepatitis,[22] extrahepatic cholestasis,[23] or congestive heart failure.[24]

Recently, the Mac-2 binding protein glycosylation isomer (M2BPGi, *Wisteria floribunda* agglutinin-positive Mac-2 binding protein, WFA+ -M2BP) has emerged as a novel serum biomarker for the assessment of LC and liver fibrosis.[25,26] Several recent studies have reported that serum M2BPGi is a predictive biomarker of LC and liver fibrosis in patients with HCV,[27,28] HBV,[29,30] ALD,[31] NAFLD,[32] AIH,[33] and primary biliary cirrhosis[34]; however, limited real-life clinical data on M2BPGi are available for assessing LC and liver fibrosis in patients with CLD.

In the present study, we aimed to evaluate the diagnostic power of M2BPGi and to identify the cut-off values of M2BPGi for LC and significant fibrosis in patients with CLD presenting various etiologies.

2. Materials and methods

2.1. Study design and setting

This study is a retrospective cohort study. We retrospectively reviewed the patients diagnosed with CLD whose serum M2BPGi was measured at Kosin University Gospel Hospital between January 2016 and December 2019.

The inclusion criteria:

(1) The diagnosis of CLD defined as decreased hepatic function which lasts over a period of 6 months, including chronic hepatitis to LC based on clinical features, histologic, laboratory, and radiological findings.
(2) The causes of CLD consisted of HBV, HCV, ALD, NAFLD, and AIH.

The exclusion criteria:

(1) Patients with conditions that may elevate serum M2BPGi (i.e., metastatic liver cancer, pancreatic cancer, chronic pancreatitis, idiopathic pulmonary fibrosis).[35]
(2) Insufficient clinical data for statistical analysis.

2.2. Patients

A total of 512 patients were initially reviewed, 58 patients were excluded. Among the 58 patients who were excluded, there were
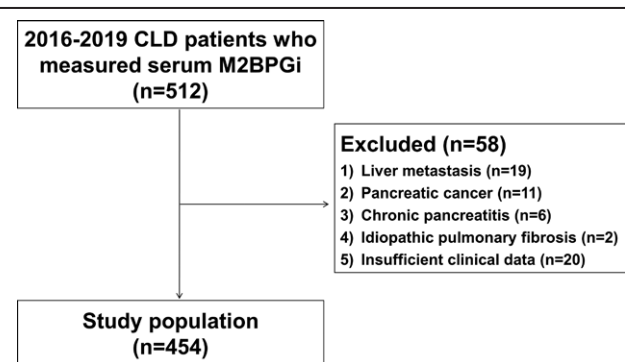

Figure 1. Flowchart of study population. CLD = chronic liver disease, M2BPGi = Mac-2 binding protein glycosylation isomer.

19 patients with metastatic liver cancer, 11 patients with pancreatic cancer, 9 patients with chronic pancreatitis, 2 patients with idiopathic pulmonary fibrosis, and 20 patients with insufficient clinical data. Finally, 454 patients enrolled in this study. Figure 1 illustrates the flow chart of study population.

Clinicopathological data including age, sex, height/weight, HBV and HCV infection status, cirrhosis status, and alcohol consumption were collected from patients' medical records. Laboratory data included platelet count, international normalized ratio (INR), bilirubin (total, direct), sodium, albumin, ammonia, gamma-glutamyltransferase, aspartate aminotransferase (AST), alanine aminotransferase (ALT), and M2BPGi. Imaging data including ultrasonography, CT, MRI, and FibroScan® (Echosens) were obtained.

2.3. Main outcomes

Main outcomes of this study were LC and significant fibrosis. LC was diagnosed by histology and/or radiological findings (ultrasonography, CT, MRI) with clinical features such as gastroesophageal varices and ascites.[12] Significant fibrosis (≥F2) was defined by histology and/or FibroScan® (Echosens) with clinical features.

2.4. Variables

Patients with HBV were defined as those positive for HBsAg, and patients with HCV were defined as positive for both anti-HCV Ab and HCV RNA. ALD referred to consumption of at least 40 g of alcohol per day for men and 20 g alcohol per day for women without other causes of CLD.[36] AIH was diagnosed by the revised international AIH group modified scoring system.

The values of conventional serum biomarkers were evaluated using the following equations: FIB-4 = [age (years) × AST (U/L)]/ [platelet count (10^9/L) × ALT $(U/L)^{0.5}$]; APRI = [AST (U/L)/upper limit normal (ULN)]/platelet count (10^9/L) × 100; GPR = [GGT (U/L)/ULN]/platelet count (10^9/L) × 100.[13,14]

Serum M2BPGi levels were measured using an automated HISCL-5000 immunoanalyzer (Sysmex Co., Kobe, Japan) via 2-step sandwich chemiluminescent enzyme immunoassay.[26,37] The values of M2BPGi were expressed as the cut-off index (COI) and were calculated using the following equation: COI = ([M2BPGi]$_{sample}$–[M2BPGi]$_{NC}$)/([M2BPGi]$_{PC}$– [M2BPGi]$_{NC}$), where [M2BPGi]$_{sample}$ is the M2BPGi value of the serum sample, [M2BPGi]$_{NC}$ is the value of the negative control, and [M2BPGi]$_{PC}$ is the positive control value. The positive control was provided as a calibration solution preliminarily standardized to yield a COI of 1.0. M2BPGi values >1.0 were considered positive (1+, $1.0 \leq COI < 3.0$; 2+, $3.0 \leq COI$).

Liver stiffness measurement (LSM) was performed using a FibroScan® (Echosens) by certified operators. At least 10 valid

measurements were obtained for each patient. The median value of valid measurements was achieved as a representative of liver stiffness. Results were considered reliable when the interquartile range/median for LSM was ≤ 30%.[38] The results were expressed in kPa. Liver stiffness cut-off values for stages F2 and F4 were set by FibroScan® (Echosens) as 7.2 and 14.6 kPa, respectively.[17,18]

2.5. Ethical statement

This study was approved by the Kosin University Gospel Hospital Institutional Review Board prior to the study (date: 24/09/2020, No. 2020-09-005). The requirement for written informed consent was waived for this retrospective study.

2.6. Statistical analysis

Continuous variables are presented as mean ± standard deviation or medians with ranges. Student t test was used to compare mean values between the LC and non-LC groups. The Mann–Whitney U test and Kruskal–Wallis test were used to compare median values between the 2 groups and multiple groups, respectively. Categorical variables were expressed as frequencies and compared using the chi-square test or Fisher exact test, as appropriate. Univariate and multivariate logistic regression analyses were conducted to identify the independent factors associated with LC. The degree of collinearity among the independent variables was identified using variance inflation factors. The receiver operating characteristic curve and area under the curve (AUC) were used to evaluate the diagnostic power of M2BPGi, FIB-4, APRI, and GPR for LC. The optimal cut-off values for assessing LC were determined using receiver operating characteristic curve analysis. Spearman rank correlation analysis was performed to evaluate the serum M2BPGi levels

with LSM (expressed in ρ). Statistical analysis was conducted using SPSS (version 23.0, SPSS Inc., Chicago, IL). Statistical significance was set at $P < .05$.

3. Results

3.1. Baseline characteristics

Baseline characteristics of the 454 patients are summarized in Table 1. The mean (±SD) age was 57.6 (±13.4) years, and the majority of the patients were male (69.8%). Among the study population, 144 (31.7%) patients were positive for HBV and 64 (14.1%) were positive for HCV. Eight (1.8%) patients were positive for both HBV and HCV. Moreover, 140 (30.8%) patients presented ALD. The mean (±SD) values of M2BPGi, FIB-4, APRI, and GPR were 3.77 (±4.12), 6.65 (±7.96), 1.85 (±2.78), and 1.51 (±3.11), respectively. Of the 454 patients, 285 (62.8%) were diagnosed with LC. The mean value of M2BPGi in the LC group was 5.31 (±4.29), which was significantly higher than that of M2BPGi in the non-LC group ($P < .001$) (Table 1). The FIB-4, APRI, and GPR values were also significantly higher in the LC group than those in the non-LC group (all $P < .001$).

3.2. Univariate and multivariate analyses for cirrhosis

Univariate analyses revealed that M2BPGi, FIB-4, APRI, and GPR were significantly associated with LC (Table 2). Multivariate logistic regression analysis was performed for age, sex, HBV, HCV, ALD, platelet count, albumin, INR, total bilirubin, ALT, sodium, M2BPGi, FIB-4, APRI, and GPR to identify the independent factors for LC. Four different multivariate analysis models for M2BPGi, FIB-4, APRI, and GPR were used to avoid multicollinearity. In the M2BPGi model, M2BPGi was significantly associated with LC (adjusted odds ratio [aOR], 1.77;

Table 1				
Baseline characteristics of study population.				
	Total (n = 454)	**Cirrhosis (n = 285)**	**Noncirrhosis (n = 169)**	**_P_ value**
Age, yr*	57.6 (±13.4)	59.8 (±10.4)	53.8 (±16.6)	<.001
Male sex†	317 (69.8)	220 (77.2)	97 (57.4)	<.001
Positive HBsAg†	144 (31.7)	111 (38.9)	33 (19.5)	<.001
Positive anti-HCV†	64 (14.1)	51 (17.9)	13 (7.7)	.003
ALD†	140 (30.8)	94 (33.0)	46 (27.2)	<.001
BMI*	25.7 (±6.3)	23.9 (±3.8)	28.8 (±8.2)	<.001
Platelet count, ×10⁹/L*	160.8 (±98.6)	123.2 (±81.7)	224.1 (±92.2)	<.001
Albumin, g/L*	3.7 (±0.7)	3.4 (±0.7)	4.1 (±0.6)	<.001
INR*	1.2 (±0.3)	1.3 (±0.4)	1.0 (±0.2)	<.001
Total bilirubin, μmol/L*	2.1 (±3.4)	2.7 (±4.1)	1.0 (±1.5)	<.001
Direct bilirubin, μmol/L*	1.2 (±2.3)	1.6 (±2.8)	0.5 (±0.9)	<.001
r-GTP, U/L*	164.9 (±262.0)	184.0 (±277.2)	132.7 (±231.2)	.044
AST, U/L*	77.2 (±99.8)	84.0 (±90.0)	65.6 (±113.9)	.057
ALT, U/L*	54.2 (±100.0)	46.3 (±84.2)	67.6 (±121.1)	.028
Sodium, mmol/L*	138.1 (±4.0)	136.7 (±4.1)	138.1 (±4.0)	<.001
Ammonia, μmol/L*	54.0 (±22.0)	56.1 (±22.8)	48.2 (±18.7)	.002
M2BPGi*	3.77 (±4.12)	5.31 (±4.29)	1.17 (±1.93)	<.001
FIB-4*	6.65 (±7.96)	9.04 (±8.93)	2.64 (±3.18)	<.001
APRI*	1.85 (±2.78)	2.33 (±2.82)	1.05 (±2.52)	<.001
GPR*	1.51 (±3.11)	1.95 (±3.71)	0.76 (±1.39)	<.001

ALD = alcoholic liver disease, ALT = alanine aminotransferase, APRI = aspartate aminotransferase-to-platelet ratio index; AST = aspartate aminotransferase, BMI = body mass index, FIB-4 = fibrosis index based on four factors, GPR = gamma-glutamyl transpeptidase-to-platelet ratio, HBsAg = hepatitis B surface antigen, HCV = hepatitis C virus, INR = international normalized ratio, M2BPGi = Mac-2 binding protein glycosylation isomer, r-GTP = r-glutamyltransferase.
*Values are presented as median (range).
†Values are presented as number (%).

Variables	Univariate analysis		Multivariate analysis			
			M2BPGi model		FIB-4 model	
	OR (95% CI)	*P* value	OR (95% CI)	*P* value	OR (95% CI)	*P* value
Age, yr	1.04 (1.02–1.05)	<.001	1.03 (1.01–1.05)	.015	1.01 (0.98–1.03)	.726
Male sex	0.40 (0.26–0.61)	<.001	0.48 (0.25–0.89)	.021	0.52 (0.27–0.97)	.04
Positive HBsAg	2.62 (1.68–4.12)	<.001	3.73 (2.05–6.84)	<.001	4.59 (2.45–8.58)	<.001
Positive anti-HCV	2.62 (1.38–4.97)	.003	1.99 (0.79–5.02)	.144	3.75 (1.58–8.93)	.003
ALD	2.16 (1.46–3.20)	<.001	2.83 (1.51–5.32)	.001	2.72 (1.48–5.01)	.001
Platelet count, ×10^9/L	0.99 (0.98–0.99)	<.001	0.99 (0.99–1.00)	<.001	1.00 (0.99–1.00)	.021
Albumin, g/L	0.17 (0.11–0.25)	<.001	0.62 (0.33–1.15)	.129	0.43 (0.25–0.75)	.003
INR	1599.37 (285.51–8959.55)	<.001	2.73 (0.32–23.36)	.358	9.69 (1.30–72.12)	.027
Total bilirubin, µmol/L	1.70 (1.40–2.06)	<.001	0.93 (0.80–1.08)	.343	0.94 (0.79–1.11)	.435
r-GTP, U/L	1.00 (1.00–1.00)	.05	-	-	-	-
AST, U/L	1.00 (1.00–1.00)	.065	-	-	-	-
ALT, U/L	1.00 (1.00–1.00)	.038	1.00 (0.99–1.00)	.004	1.00 (0.99–1.00)	.003
Sodium, mmol/L	0.91 (0.86–0.96)	<.001	1.03 (0.95–1.12)	.452	1.03 (0.95–1.11)	.516
Ammonia, µmol/L	1.02 (1.01–1.03)	.002	-	-	-	-
M2BPGi	1.87 (1.63–2.15)	<.001	1.77 (1.52–2.07)	<.001	-	-
FIB-4	1.47 (1.34–1.60)	<.001	-	-	1.23 (1.11–1.37)	<.001
APRI	1.44 (1.23–1.68)	<.001	-	-	-	-
GPR	1.44 (1.21–1.71)	<.001	-	-	-	-

ALD = alcoholic liver disease, ALT = alanine aminotransferase, APRI = aspartate aminotransferase-to-platelet ratio index, AST = aspartate aminotransferase, FIB-4 = fibrosis index based on four factors, GPR = gamma-glutamyl transpeptidase-to-platelet ratio, HBsAg = hepatitis B surface antigen, HCV = hepatitis C virus, INR = international normalized ratio, M2BPGi = Mac-2 binding protein glycosylation isomer, r-GTP = r-glutamyltransferase.

95% CI, 1.52–2.07). Furthermore, other independent variables, such as age, sex, HBV, ALD, and platelet count, were significantly associated with LC. FIB-4 was significantly associated with LC in the FIB-4 model (aOR, 1.23; 95% CI, 1.11–1.37). In addition, other independent variables, such as sex, HBV, HCV, ALD, albumin, and INR, were significantly associated with LC (Table 2). In contrast, APRI (aOR, 1.18; 95% CI, 0.95–1.46) and GPR (aOR, 1.15; 95% CI, 0.99–1.34) were not significantly associated with LC.

3.3. Diagnostic power of M2BPGi for cirrhosis and significant fibrosis compared to other fibrosis markers

The AUC of M2BPGi (AUC, 0.866; 95% CI, 0.830–0.902) for diagnosing LC (F4) was comparable to that of FIB-4 (AUC, 0.860; 95% CI, 0.823–0.896), APRI (AUC, 0.806; 95% CI, 0.762–0.850), and GPR (AUC, 0.759; 95% CI, 0.711–0.808) (Fig. 2). The optimal cut-off value of M2BPGi for LC was 1.37, with a sensitivity of 86.7% and specificity of 78.7%. Moreover, the AUC of M2BPGi (AUC, 0.816; 95% CI, 0.773–0.858) for diagnosing significant fibrosis (≥F2) was comparable to that of FIB-4 (AUC, 0.773; 95% CI, 0.726–0.819), APRI (AUC, 0.752; 95% CI, 0.700–0.804), and GPR (AUC, 0.710; 95% CI, 0.654–0.766) (Fig. 3). Furthermore, the optimal cut-off value for M2BPGi for significant fibrosis was 0.89, with a sensitivity and specificity of 84.1% and 62.4%, respectively.

3.4. Correlation between M2BPGi and LSM

LSM was performed in 151 of the 454 patients. The fibrosis stage was categorized as F0/F1, F2/F3, and F4. Forty-five (29.8%) patients were F0/F1, 43 (28.5%) patients were F2/F3, and 63 (41.7%) patients were F4. The median levels of serum M2BPGi were 0.44 (F0/F1), 0.66 (F2/F3), and 4.03 (F4), respectively (Fig. 4). The Kruskal–Wallis test revealed a significant difference

between the fibrosis stages (*P* < .001). Moreover, significant differences were observed between the F0/F1 and F2/F3 stages (*P* = .038), between the F2/F3 and F4 stages (*P* < .001), and between the F0/F1 and F4 stages (*P* < .001). Furthermore, LSM was significantly correlated with M2BPGi (ρ = 0.778, *P* < .001), FIB-4 (ρ = 0.704, *P* < .001), APRI (ρ = 0.702, *P* < .001), and GPR (ρ = 0.685, *P* < .001), respectively.

4. Discussion

Noninvasive methods to assess LC and liver fibrosis need to be accurate, convenient, safe, and repeatable; however, conventional serum markers and transient elastography do not fully satisfy these requirements. This study confirmed that serum M2BPGi is a reliable method for assessing LC and significant fibrosis in patients with CLD.

In this study, serum M2BPGi levels were found to be independently associated with LC. This finding was in accordance with that of previous clinical studies in patients with CLD.[28,29] And we found that the median levels of serum M2BPGi increased as fibrosis progressed. This is supported by the fact that M2BPGi is crucial in the process of liver fibrosis. In an in vitro study, Bekki et al[39] showed that M2BPGi, which is secreted by hepatic stellate cells (HSCs), promotes Kupffer cells to express Mac-2 and induces the activation of HSCs. Activated HSCs are known to secrete inflammatory cytokines and produce substantial amounts of extracellular matrix, thereby resulting in liver fibrosis.[40] Therefore, serum M2BPGi would be useful for assessing significant fibrosis in clinical setting.

In the present study, we identified the strong diagnostic power of serum M2BPGi for LC (AUC: 0.866) and significant fibrosis (AUC: 0.816), which was comparable to that of other serum markers (FIB-4, APRI, and GPR). The cut-off value of M2BPGi for LC and significant fibrosis was 1.37 and 0.89, respectively. These results were consistent with those of previous studies.
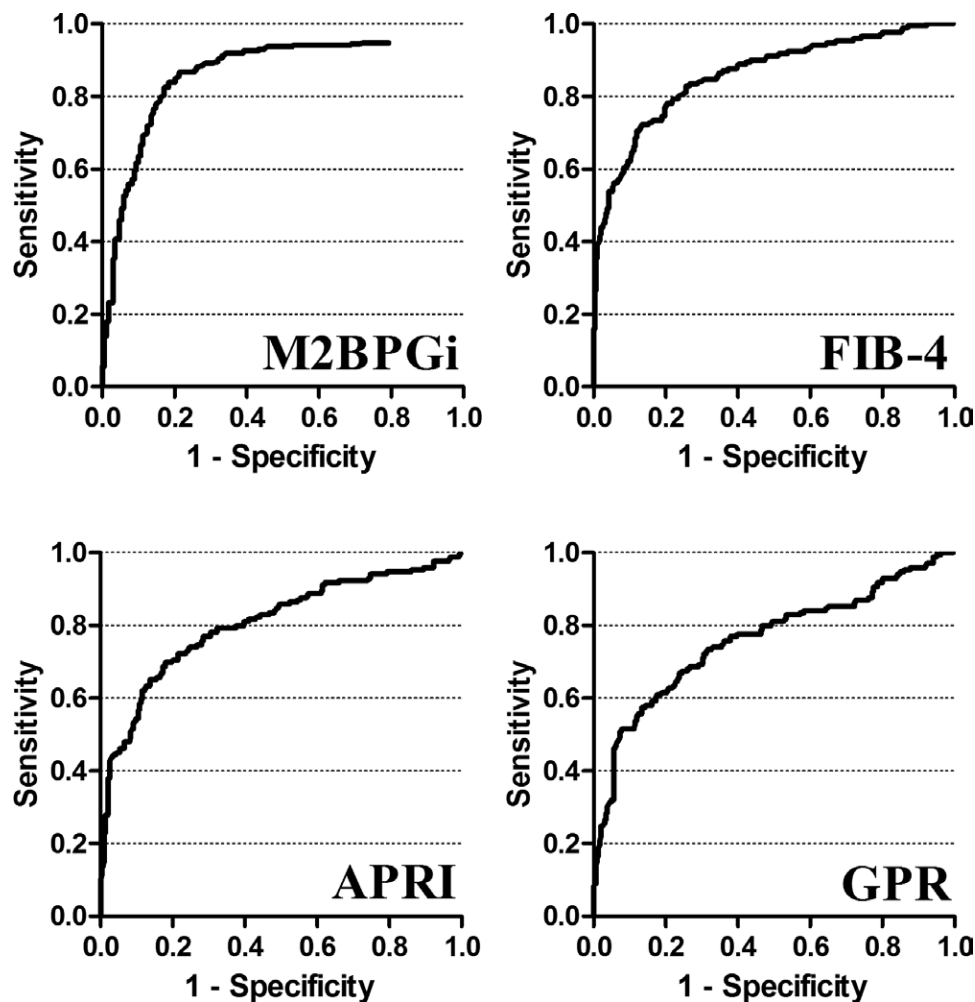
Figure 2. Receiver operating characteristic curves of serum M2BPGi, FIB-4, APRI, and GPR for diagnosing cirrhosis (F4). APRI = aspartate aminotransferase-to-platelet ratio index, AUC = area under the curve, FIB-4 = fibrosis index based on four factors, GPR = gamma-glutamyl transpeptidase-to-platelet ratio, M2BPGi = Mac-2 binding protein glycosylation isomer.

Based on a study conducted against 680 HCV patients and 164 healthy controls in China, the AUCs of serum M2BPGi for LC and significant fibrosis were 0.892 and 0.774, respectively.[27] And the cut-off values of M2BPGi for LC and significant fibrosis were 1.36 (sensitivity 86.8%, specificity 78.6%) and 0.95 (sensitivity 74.5%, specificity 69.4%), respectively. In a study comprising 327 HBV patients, the AUC of serum M2BPGi for F4 was 0.914 and the cut-off value of M2BPGi for F4 was 0.96, with a sensitivity of 83.3% and a specificity of 92.7%.[29] A study of 220 patients with NAFLD showed that the AUC and cut-off value of serum M2BPGi for F4 were 0.840 and 0.70, respectively, with a sensitivity of 85.7% and a specificity of 72.8%.[41] Although this study could not demonstrate that the diagnostic power of serum M2BPGi for LC and significant fibrosis was superior to that of other serum biomarkers, it was found that the diagnostic power of serum M2BPGi was comparable to that of other serum markers. Therefore, diagnostic performance for LC and significant fibrosis may be improved by using serum M2BPGi with conventional serum biomarkers in clinical practice.

Serum M2BPGi levels were found to be significantly correlated with LSM in this study. These findings were consistent with those of previous studies.[27–30,42] Fibroscan® (Echosens) is an already validated noninvasive method for diagnosing LC and fibrosis in patients with various CLD etiologies. Therefore, combination of serum M2BPGi and Fibroscan® (Echosens) would improve the diagnostic performance for LC and liver fibrosis.

4.1. Limitations

This study also has several limitations. First, because this study was a single-center retrospective study, careful interpretation is required due to selection and information bias. Second, we were unable to identify a correlation between M2BPGi and the results of liver biopsy, because of lack of patients who underwent liver biopsy. Third, Fibroscan® (Echosens) was conducted on only 151 patients out of total 454 patients. Thus, we could not separate the F0/F1 and F2/F3 stages due to the relatively small sample size of the F1 and F3 stages. Therefore, further large-scale prospective studies are required.

4.2. Conclusions

In this study, we confirmed that serum M2BPGi is a reliable noninvasive method for the assessment of LC and significant fibrosis in patients with CLD. Serum M2BPGi would help the physicians in clinical diagnosis of LC and significant fibrosis in patients with CLD.

Author contributions

Conceptualization: Hyunyong Hwang, Kwang Il Seo, Myunghee Yoon.
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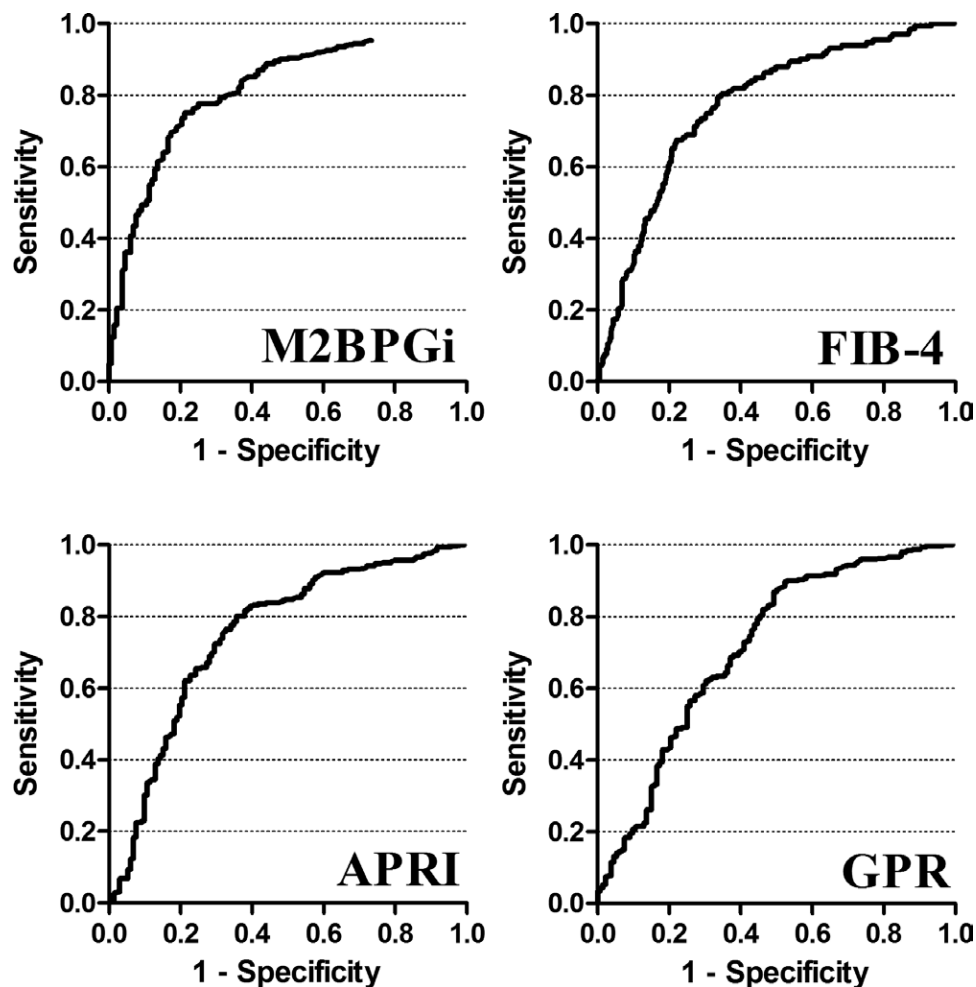**Interpretation of data:** Byung Cheol Yun, Kwang Il Seo.

Figure 3. Receiver operating characteristic curves of serum M2BPGi, FIB-4, APRI, and GPR for assessing significant fibrosis (≥F2). APRI = aspartate amino-transferase-to-platelet ratio index, AUC = area under the curve, FIB-4 = fibrosis index based on four factors, GPR = gamma-glutamyl transpeptidase-to-platelet ratio, M2BPGi = Mac-2 binding protein glycosylation isomer.

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References

[1] Friedman SL. Seminars in medicine of the Beth Israel Hospital, Boston. The cellular basis of hepatic fibrosis. Mechanisms and treatment strategies. N Engl J Med. 1993;328:1828–35.

[2] Poynard T, Mathurin P, Lai CL, et al. A comparison of fibrosis progression in chronic liver diseases. J Hepatol. 2003;38:257–65.

[3] D'Amico G, Morabito A, D'Amico M, et al. Clinical states of cirrhosis and competing risks. J Hepatol. 2018;68:563–76.

[4] D'Amico G, Garcia-Tsao G, Pagliaro L. Natural history and prognostic indicators of survival in cirrhosis: a systematic review of 118 studies. J Hepatol. 2006;44:217–31.

[5] Jang JW. Current status of liver diseases in Korea: liver cirrhosis. Korean J Hepatol. 2009;15(Suppl 6):S40–9.

[6] Majumdar A, Kitson MT, Roberts SK. Treatment of hepatitis C in patients with cirrhosis: remaining challenges for direct-acting antiviral therapy. Drugs. 2015;75:823–34.

[7] Wright TL. Introduction to chronic hepatitis B infection. Am J Gastroenterol. 2006;101(Suppl 1):S1–6.

[8] Bravo AA, Sheth SG, Chopra S. Liver biopsy. N Engl J Med. 2001;344:495–500.

[9] Cadranel JF, Rufat P, Degos F. Practices of liver biopsy in France: results of a prospective nationwide survey. For the group of epidemiology of the French Association for the Study of the Liver (AFEF). Hepatology. 2000;32:477–81.

[10] Colloredo G, Guido M, Sonzogni A, et al. Impact of liver biopsy size on histological evaluation of chronic viral hepatitis: the smaller the sample, the milder the disease. J Hepatol. 2003;39:239–44.

[11] Rousselet MC, Michalak S, Dupré F, et al. Sources of variability in histological scoring of chronic viral hepatitis. Hepatology. 2005;41:257–64.

[12] Lurie Y, Webb M, Cytter-Kuint R, et al. Non-invasive diagnosis of liver fibrosis and cirrhosis. World J Gastroenterol. 2015;21:11567–83.

[13] Wai CT, Greenson JK, Fontana RJ, et al. A simple noninvasive index can predict both significant fibrosis and cirrhosis in patients with chronic hepatitis C. Hepatology. 2003;38:518–26.

[14] Sterling RK, Lissen E, Clumeck N, et al. Development of a simple non-invasive index to predict significant fibrosis in patients with HIV/HCV coinfection. Hepatology. 2006;43:1317–25.

[15] Vallet-Pichard A, Mallet V, Nalpas B, et al. FIB-4: an inexpensive and accurate marker of fibrosis in HCV infection. Comparison with liver biopsy and fibrotest. Hepatology. 2007;46:32–6.

[16] Liu DP, Lu W, Zhang ZQ, et al. Comparative evaluation of GPR versus APRI and FIB-4 in predicting different levels of liver fibrosis of chronic hepatitis B. J Viral Hepat. 2018;25:581–9.

[17] Foucher J, Chanteloup E, Vergniol J, et al. Diagnosis of cirrhosis by transient elastography (FibroScan): a prospective study. Gut. 2006;55:403–8.

[18] Ganne-Carrié N, Ziol M, de Ledinghen V, et al. Accuracy of liver stiffness measurement for the diagnosis of cirrhosis in patients with chronic liver diseases. Hepatology. 2006;44:1511–7.

[19] Ziol M, Handra-Luca A, Kettaneh A, et al. Noninvasive assessment of liver fibrosis by measurement of stiffness in patients with chronic hepatitis C. Hepatology. 2005;41:48–54.

[20] O EA, Büchter M, H AB, et al. Performance and utility of transient elastography and non-invasive markers of liver fibrosis in patients
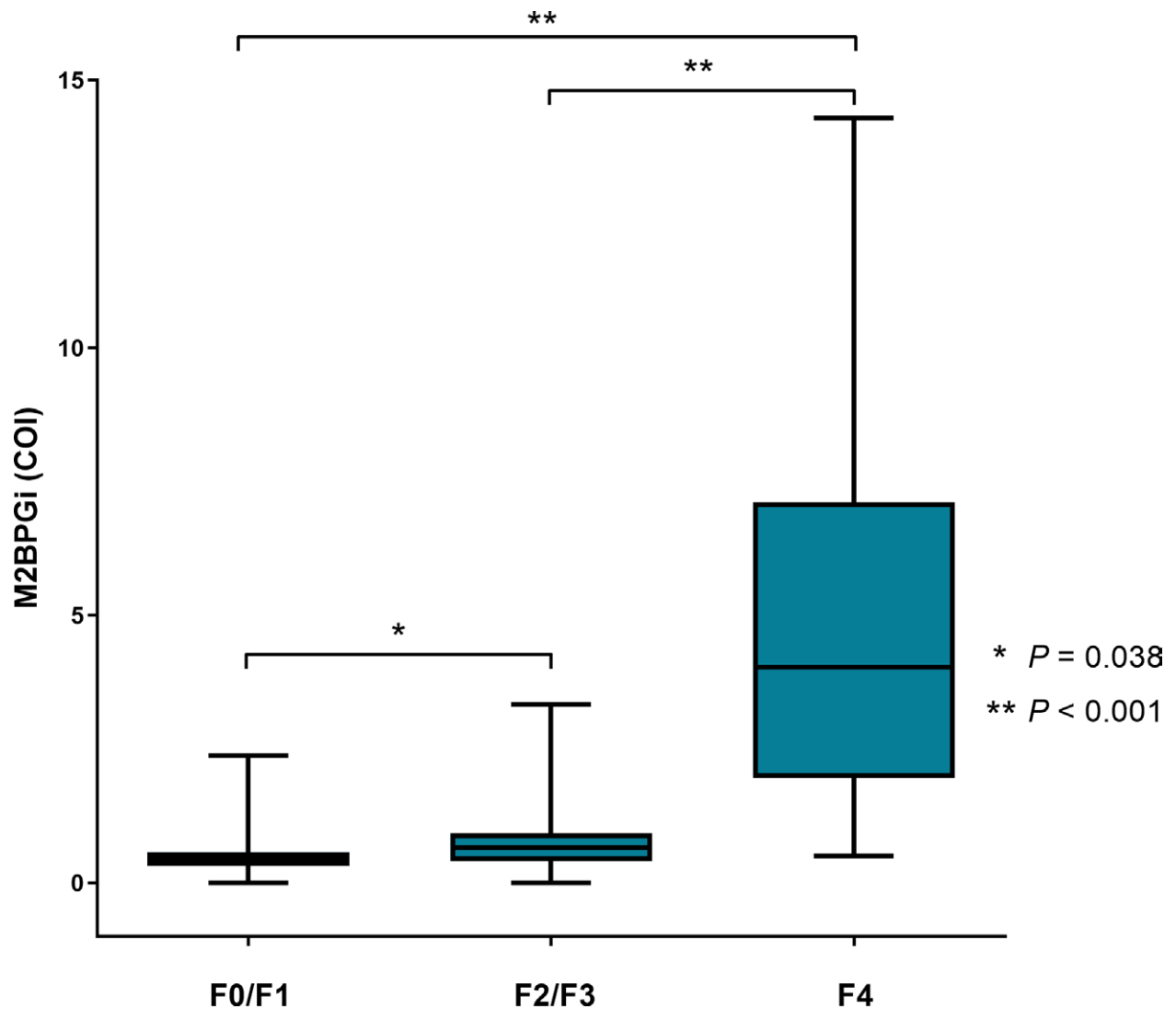
Figure 4. Box plots of serum M2BPGi levels based on the fibrosis stage. Horizontal lines within boxes indicate medians; the upper and lower borders of boxes indicate the 75th and 25th percentiles, respectively. The vertical whiskers indicate the minimum and maximum levels. COI = cut-off index, M2BPGi = Mac-2 binding protein glycosylation isomer.

with autoimmune hepatitis: a single centre experience. Hepat Mon. 2016;16:e40737.

[21] Wong VW, Vergniol J, Wong GL, et al. Diagnosis of fibrosis and cirrhosis using liver stiffness measurement in nonalcoholic fatty liver disease. Hepatology. 2010;51:454–62.

[22] Coco B, Oliveri F, Maina AM, et al. Transient elastography: a new surrogate marker of liver fibrosis influenced by major changes of transaminases. J Viral Hepat. 2007;14:360–9.

[23] Millonig G, Reimann FM, Friedrich S, et al. Extrahepatic cholestasis increases liver stiffness (FibroScan) irrespective of fibrosis. Hepatology. 2008;48:1718–23.

[24] Millonig G, Friedrich S, Adolf S, et al. Liver stiffness is directly influenced by central venous pressure. J Hepatol. 2010;52:206–10.

[25] Kuno A, Ikehara Y, Tanaka Y, et al. A serum "sweet-doughnut" protein facilitates fibrosis evaluation and therapy assessment in patients with viral hepatitis. Sci Rep. 2013;3:1065.

[26] Kuno A, Sato T, Shimazaki H, et al. Reconstruction of a robust glycodiagnostic agent supported by multiple lectin-assisted glycan profiling. Proteomics Clin Appl. 2013;7:642–7.

[27] Xu H, Kong W, Liu L, et al. Accuracy of M2BPGi, compared with Fibro Scan®, in analysis of liver fibrosis in patients with hepatitis C. BMC Gastroenterol. 2017;17:62.

[28] Nishikawa H, Takata R, Enomoto H, et al. Proposal of a predictive model for advanced fibrosis containing Wisteria floribunda agglutinin-positive Mac-2-binding protein in chronic hepatitis C. Hepatol Res. 2017;47:E74–84.

[29] Mak LY, Wong DK, Cheung KS, Seto WK, Lai CL, Yuen MF. Role of serum M2BPGi levels on diagnosing significant liver fibrosis and cirrhosis in treated patients with chronic hepatitis B virus infection. Clin Transl Gastroenterol. 2018;9:163.

[30] Zou X, Zhu MY, Yu DM, et al. Serum WFA(+) -M2BP levels for evaluation of early stages of liver fibrosis in patients with chronic hepatitis B virus infection. Liver Int. 2017;37:35–44.

[31] Nguyen-Khac E, Chatelain D, Tramier B, et al. Assessment of asymptomatic liver fibrosis in alcoholic patients using fibroscan: prospective comparison with seven non-invasive laboratory tests. Aliment Pharmacol Ther. 2008;28:1188–98.

[32] Abe M, Miyake T, Kuno A, et al. Association between Wisteria floribunda agglutinin-positive Mac-2 binding protein and the fibrosis stage of non-alcoholic fatty liver disease. J Gastroenterol. 2015;50:776–84.

[33] Nishikawa H, Enomoto H, Iwata Y, et al. Clinical significance of serum Wisteria floribunda agglutinin positive Mac-2-binding protein level and high-sensitivity C-reactive protein concentration in autoimmune hepatitis. Hepatol Res. 2016;46:613–21.

[34] Umemura T, Joshita S, Sekiguchi T, et al. Serum Wisteria floribunda agglutinin-positive Mac-2-binding protein level predicts liver fibrosis and prognosis in primary biliary cirrhosis. Am J Gastroenterol. 2015;110:857–64.

[35] Shirabe K, Bekki Y, Gantumur D, et al. Mac-2 binding protein glycan isomer (M2BPGi) is a new serum biomarker for assessing liver fibrosis: more than a biomarker of liver fibrosis. J Gastroenterol. 2018;53:819–26.

[36] Becker U, Deis A, Sørensen TI, et al. Prediction of risk of liver disease by alcohol intake, sex, and age: a prospective population study. Hepatology. 1996;23:1025–9.

[37] Narimatsu H. Development of M2BPGi: a novel fibrosis serum glyco-biomarker for chronic hepatitis/cirrhosis diagnostics. Expert Rev Proteomics. 2015;12:683–93.

[38] Boursier J, Zarski JP, de Ledinghen V, et al. Determination of reliability criteria for liver stiffness evaluation by transient elastography. Hepatology. 2013;57:1182–91.

[39] Bekki Y, Yoshizumi T, Shimoda S, et al. Hepatic stellate cells secreting WFA(+) -M2BP: Its role in biological interactions with Kupffer cells. J Gastroenterol Hepatol. 2017;32:1387–93.

[40] Yi HS, Jeong WI. Interaction of hepatic stellate cells with diverse types of immune cells: foe or friend? J Gastroenterol Hepatol. 2013;28(Suppl 1):99–104.

[41] Lai LL, Chan WK, Sthaneshwar P, Nik Mustapha NR, Goh KL, Mahadeva S. Serum Wisteria floribunda agglutinin-positive Mac-2 binding protein in non-alcoholic fatty liver disease. PLoS One. 2017;12:e0174982.

[42] Moon HW, Park M, Hur M, Kim H, Choe WH, Yun YM. Usefulness of enhanced liver fibrosis, glycosylation isomer of Mac-2 binding protein, galectin-3, and soluble suppression of tumorigenicity 2 for assessing liver fibrosis in chronic liver diseases. Ann Lab Med. 2018;38:331–7.