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The usefulness of liver stiffness measurement using FibroScan in chronic hepatitis C in South Korea: A multicenter, prospective study

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Abstract

Aim: We investigated the accuracy of liver stiffness measurement (LSM) in chronic hepatitis C (CHC) in a multicenter, prospective study in South Korea.

Methods: Between June 2005 and July 2009, 91 CHC patients without a previous history of antiviral treatment, clinical evidences of cirrhosis, coinfection with other viruses, and heavy alcohol consumption and with alanine aminotransferase (ALT) ≤ 5x upper limit of normal, total bilirubin ≤ 1.5 mg/dL, sufficient liver biopsy quality (≥ 15 mm and more than six portal tracts), interquartile range to median liver stiffness (LS) value ratio ≤ 0.21, and more than 10 valid measurements, were recruited. The Batts and Ludwig scoring system was used for histologic assessment. Age–platelet index (API), aspartate aminotransferase (AST)–to–platelet ratio index (APRI), and age–spleen–platelet ratio index (ASPRI) were calculated. Area under the receiver operating characteristic curve (AUROC) was used to evaluate the performance of LSM and other noninvasive models.

Results: The mean age was 47.9 years, and the mean LS value was 7.7 kPa (44 men and 47 women). LS value was highly correlated to the fibrosis stages ($r = 0.835$, $P < 0.001$). The AUROCs of LSM were 0.909 for ≥ F2, 0.993 for ≥ F3, and 0.970 for F = 4 and were superior to those of API (0.72, 0.858, and 0.948, respectively), APRI (0.780, 0.887, and 0.904, respectively), and ASPRI (0.713, 0.862, and 0.957, respectively). The optimal cutoff LS values were 6.2 kPa for ≥ F2, 7.7 kPa for ≥ F3, and 11.0 kPa for F = 4.

Conclusions: Our data suggest that LSM can accurately assess liver fibrosis in patients with CHC and be applied in South Korea.

Introduction

The prognosis and management of chronic hepatitis C (CHC) are highly dependent on the extent of liver fibrosis.[1] Until recently, liver biopsy (LB) has been considered the only diagnostic method for the assessment of liver fibrosis. However, it is an invasive and painful procedure with potential life-threatening complications, including bleeding, perforation of other organs, and death, which sometimes limit its acceptance and repetition.[2] Thus, researchers have focused on seeking alternative noninvasive tools to establish information on the extent of fibrosis. Recently, liver stiffness measurement (LSM) using FibroScan (EchoSens, Paris, France), which is a rapid, noninvasive, and reproducible method, has been demonstrated to be a reliable tool for assessing liver fibrosis in patients with chronic hepatitis B (CHB) and CHC.[3–5] FibroScan generates an elastic wave using a vibrator applied to the intercostal spaces at the level of the right lobe of the liver and measures the propagation velocity of the shear wave, which is directly related to liver stiffness (LS).[6]

Initially, because LSM was first designed in France, most investigations on LSM were conducted in Europe where CHC is prevalent. Accordingly, much data on the usefulness of LSM in assessing liver fibrosis in patients with CHC has been accumulated. Recently, the applicability of LSM to patients with CHB has also been tested in several Asian studies.[7,8] However, the performance of LSM in an Asian population with CHC has not been validated yet, although some Japanese studies reported the usefulness of LSM for assessing liver fibrosis in patients with CHC.[9,10] Furthermore, because recent studies revealed that higher body mass index (BMI) and metabolic syndrome might increase

LS values,[11] data on the performance of LSM in Asian populations with generally lower BMI and lower prevalence of metabolic syndrome compared to those of Europe is crucial to confirm the applicability of LSM to Asian populations. Thus, in this multicenter and prospective study, we investigated the usefulness of LSM for patients with CHC in South Korea.

Methods

Patients

From June 2005 to July 2009, a total of 128 patients with CHC who underwent LB and LSM were recruited prospectively and consecutively for this study. The indication of LB was the assessment of the severity of liver fibrosis and inflammation before antiviral treatment. All patients had CHC infection, defined by detectable serum anti-hepatitis C virus (HCV) antibodies and HCV-RNA. The study population consisted of 86 patients from Shinchon Severance Hospital of Yonsei University College of Medicine, Seoul, South Korea, 15 from Gangnam Severance Hospital of Yonsei University College of Medicine, Seoul, South Korea, 19 from Ajou University Hospital of Ajou University School of Medicine, Suwon, South Korea, 6 from Hallym University Hospital of Hallym University College of Medicine, Chuncheon, South Korea, and 2 from Saint Vincent's Hospital of Catholic University School of Medicine, Suwon, South Korea.

No patient had evidence of decompensated liver cirrhosis such as history of variceal bleeding, severe ascites, hepatic encephalopathy, or Child-Pugh class B/C at the time of LB and LSM. Exclusion criteria were as follows: (i) previous antiviral treatment before LB, (ii) evidence of liver cirrhosis (platelet count < 100 000/uL and ultrasonographic findings suggestive of cirrhosis such as blunted or nodular liver edge accompanied by splenomegaly (> 12 cm), or presence of esophageal or gastric varices),[12] (iii) elevated alanine aminotransferase (ALT) more than 5 times upper limit of normal (ULN),[13–15] (iv) elevated total bilirubin more than 1.5 mg/dL,[16,17] (v) insufficient LB quality (LB length < 15 mm or less than six portal tracts),[18] (vi) coinfection with hepatitis B, hepatitis D, or human immunodeficiency virus, (vii) alcohol ingestion in excess of 40 g/day for more than five years, (viii) an interquartile range (IQR) to median LS value ratio (IQR/M) more than 0.21,[19] (ix) less than 10 valid measurement of LS values, (x) different day between LB and LSM, or (xi) right-sided heart failure.[20] With these exclusion criteria, thirty-seven patients were excluded. Ultimately, ninety-one patients were enrolled for statistical analysis. The study protocol was in accordance with the ethical guidelines of the 1975 Declaration of Helsinki. Written informed consent was obtained from each participant or responsible family members after the possible complications of LB had been fully explained. This study was also approved by the independent institutional review board of each hospital.

Besides the demographic data, blood parameters such as aspartate aminotransferase (AST), ALT, albumin, total bilirubin, prothrombin time, platelet count were collected on the same day with LB and LSM. The AST and ALT level were measured using an automated chemistry analyzer (Hitachii 7600, Tokyo, Japan), and 40 IU/mL was defined as the ULN of AST and ALT, respectively.

Evaluation of liver histology

Liver biopsy specimens were fixed in formalin and paraffin embedded. Four-micrometer-thick sections were stained with hematoxylin and eosin (H&E) and Masson trichrome. All liver tissue samples were evaluated by an experienced hepatopathologist of each center who was blinded to patients' clinical data including LSM results. Liver biopsy specimens unsuitable for fibrosis assessment (LB length < 15 mm or less than six portal tracts) were excluded from analysis.

Liver histology was evaluated semiquantitatively according to the Batts and Ludwig scoring system. Fibrosis was staged on a 0 to 4 scale: F0, no fibrosis; F1, portal fibrosis without septa; F2, portal fibrosis and a few septa; F3, numerous septa without cirrhosis; and F4, cirrhosis. Activity was graded as A0, none; A1, minimal; A2, mild; A3, moderate; and A4, severe activity. Steatosis was categorized by visual assessment as S0 (nonsignificant, < 5%), S1 (mild, 5–33%), S2 (moderate, 34–66%), and S3 (severe, > 66% of hepatocytes with fat deposits).[21,22]

Liver stiffness measurement

The LSM was performed once on the same day with LB by a single physician blinded to patients' clinical data according to previously described methods.[23,24] The success rate was calculated as the number of valid measurements divided by the total number of measurements. The results were expressed as kilopascals. The IQR was defined as an index of intrinsic variability of LSM corresponding to the interval of LSM results containing 50% of the valid measurements between the 25th and 75th percentiles. The median value was considered representative of the elastic modulus of the liver. Only procedures with at least 10 valid measurements and IQR/M < 0.21 were considered reliable, irrespective of success rate of LSM shots.[19]

Other noninvasive models for assessing liver fibrosis

The age–platelet index (API), AST to platelet ratio index (APRI), and age–spleen–platelet ratio index (ASPRI) were also evaluated to compare with LSM. The API, APRI, and ASPRI were calculated according to the previous studies by Poynard, Wai, and Kim *et al.*, respectively (Table 1).[25–27]

Statistical analysis

The patient characteristics are given as the mean ± SD or *n* (%) as appropriate. Variables were compared using the independent *t*-test and χ^2 test. Spearman's correlation analysis (*r*) was calculated to evaluate the correlation between LS values and other clinical and laboratory variables. Binary logistic regression analysis was used to identify the factors to predict the discordance between LSM and LB results. The diagnostic performance of LSM and other noninvasive models were assessed by the areas under the receiver operating characteristic (AUROC) curves. The optimal cutoff LS values were determined to maximize the sum of sensitivity (Se) and specificity (Sp) and corresponding positive predictive values (PPV), negative predictive values (NPV), negative likelihood ratio, and positive likelihood ratio were computed for these cutoff LS

Table 1 Simple fibrosis tests

Fibrosis test	Calculation method
API	Age (years): < 30 = 0; 30–39 = 1; 40–49 = 2; 50–59 = 3; 60–69 = 4; ≥ 70 = 5 Platelet count (10^9/L): ≥ 225 = 0; 200–224 = 1; 175–199 = 2; 150–174 = 3; 125–149 = 4; < 125 = 5 API is the sum of the above (possible value 0–10)
APRI	[(AST/ULN)/platelet count (10^9/L)] × 100
SPRI	Spleen size (cm)/platelet count (10^9/L) × 100
ASPRI	Age (years): < 30 = 0; 30–39 = 1; 40–49 = 2; 50–59 = 3; 60–69 = 4; ≥ 70 = 5 ASPRI is the sum of age and SPRI

API, age–platelet index; APRI, aspartate aminotransferase-to-platelet ratio index; ASPRI, age–spleen–platelet ratio index; AST, aspartate aminotransferase; SPRI, Spleen-to-platelet ratio index; ULN, upper limit of normal.

Table 2 Baseline characteristics ($n = 91$)

Variables	Enrolled patients ($n = 91$)	Excluded patients ($n = 37$)	*P*-value
Male gender	44 (48.4)	26 (70.3)	0.031
Age (years)	47.9 ± 11.6	50.4 ± 12.8	0.301
Genotype			0.053
I	45 (49.5)	9 (31.0)	
II	43 (47.3)	18 (62.1)	
III	3 (3.3)	2 (6.9)	
Body mass index (kg/m^2)	22.9 ± 2.9	24.6 ± 2.5	0.003
ALT (IU/L)	40.2 ± 20.9	94.5 ± 73.5	< 0.001
Albumin (g/dL)	4.4 ± 0.4	4.2 ± 0.5	0.062
Total bilirubin (mg/dL)	0.7 ± 0.4	1.3 ± 1.6	0.049
Prothrombin time (INR)	0.97 ± 0.08	1.03 ± 0.13	0.008
Platelet count (10^3/mm^3)	202 ± 61	166 ± 67	0.005
Spleen size (cm)	9.3 ± 1.3	10.9 ± 1.9	0.001
API	4.1 ± 2.4	5.3 ± 2.6	0.015
APRI	0.6 ± 0.5	2.1 ± 3.3	0.008
ASPRI	7.3 ± 2.9	11.3 ± 6.9	0.017
Liver stiffness value (kPa)	7.7 ± 6.9	15.9 ± 14.9	0.002
Success rate (%)	94.8 ± 10.4	95.8 ± 8.2	0.644

Variables were expressed as mean ± standard deviation (SD), or *n* (%). ALT, alanine aminotransferase; API, age–platelet index; APRI, aspartate aminotransferase to platelet ratio index; ASPRI, age–spleen–platelet ratio index; INR, international normalized ratio.

values. A two-sided *P*-value of < 0.05 was considered significant and all statistical analyses were performed using SPSS 12.0 (SPSS, Chicago, IL, USA).

Results

Baseline characteristics

The baseline characteristics of all patients who were enrolled and excluded at the time of LSM and LB are summarized in Table 2. The proportion of males, BMI, ALT, total bilirubin, prothrombin time, spleen size, LS values of 37 excluded patients were significantly higher than 91 enrolled patients, whereas platelet count was significantly lower in excluded patients (all $P < 0.05$). Age, albumin, and the proportion of HCV genotype were not significantly different between the two groups (all $P > 0.05$). Of the 37 excluded patients, 3 had a previous antiviral treatment before LB, 4 had IQR/M ≥ 0.21, 5 showed clinical evidences of liver cirrhosis, 9 had ALT ≥ 5x ULN, 2 showed total bilirubin ≥ 1.5 mg/dL, 9 had insufficient LB quality, and 1 had mixed etiology of liver cirrhosis. One patient with ALT ≥ 5x ULN and insufficient LB quality, one with clinical evidences of liver cirrhosis and insufficient LB quality, and two with mixed etiology of liver cirrhosis and ALT ≥ 5x ULN were also excluded.

The mean age, BMI, ALT, and LS values of all enrolled patients (44 men and 47 women) were 47.9 ± 11.6 years, 22.9 ± 2.9 kg/m^2, 40.2 ± 20.9 IU/L, and 7.7 ± 6.9 kPa, respectively. Fifty-three (58.2%) and 38 (41.8%) patients had normal ALT level and ULN < ALT ≤ 5x ULN, respectively.

Table 3 Liver histology ($n = 91$)

	Fibrosis	Activity		Steatosis	
Stage	*N*	Grade	*n* (%)	Category	*n* (%)
0	3 (3.3)	0	3 (100.0)	0	3 (100.0)
1	38 (41.7)	1	4 (10.5)	0	4 (100.0)
		2	28 (73.7)	0	26 (92.9)
				1	2 (7.1)
		3	6 (15.8)	0	6 (100.0)
		4	–	–	–
2	29 (31.9)	1	1 (3.4)	0	1 (100.0)
		2	21 (72.4)	0	18 (85.7)
				1	2 (9.5)
				2	1 (4.8)
		3	7 (24.2)	0	7 (100.0)
		4	–	–	–
3	12 (13.2)	1	1 (8.3)	0	1 (100.0)
		2	5 (41.7)	0	4 (80.0)
				1	1 (20.0)
		3	4 (33.3)	0	3 (75.0)
				1	1 (25.0)
		4	2 (16.7)	0	2 (100.0)
4	9 (9.9)	1	–	–	–
		2	3 (33.3)	0	2 (66.7)
				1	1 (33.3)
		3	5 (55.6)	0	4 (80.0)
				1	1 (20.0)
		4	1 (11.1)	0	1 (100.0)

Variables are expressed as *n* (%).

Liver histology

Distribution for fibrosis stage, activity grade, and steatosis of all patients are presented in Table 3. Three patients (3.3%) were classified as F0 fibrosis stage (without fibrosis), 38 (41.7%) as F1, 29 (31.9%) as F2, 12 (13.2%) as F3, and 9 (9.9%) as F4. Fifty (55.0%) and 21 (23.1%) patients had ≥ F2 and ≥ F3 fibrosis stage, respectively. Eighty-two (90.1%) patients showed S0 (non-significant) and eight (8.8%) showed S1 (mild) steatosis. Only one patient (1.1%) showed S2 steatosis (moderate). The median LS

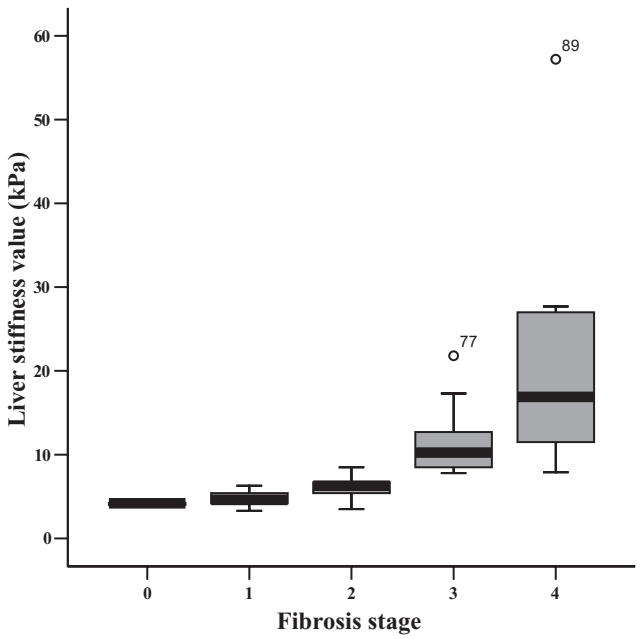

Figure 1 Liver stiffness values for each fibrosis stage. The *top* and *bottom* of the boxes are the first and third quartiles, respectively. Accordingly, the length of box plot represents the interquartile range within which 50% of the values were located. The lines through the middle of boxes indicate the median values. The median liver stiffness value was 4.2 kPa (range, 3.8–4.3 kPa) in F0 fibrosis stage, 4.6 kPa (range, 3.3–6.3 kPa) in F1, 6.3 kPa (range, 3.5–8.5 kPa) in F2, 10.3 kPa (range, 7.8–21.8 kPa) in F3, and 16.9 kPa (range, 7.9–57.2 kPa) in F4. kPa, kilopascal.

value was 4.2 kPa (range, 3.8–4.3 kPa) for patients with F0 fibrosis stage, 4.6 kPa (range, 3.3–6.3 kPa) for F1, 6.3 kPa (range, 3.5–8.5 kPa) for F2, 10.3 kPa (range, 7.8–21.8 kPa) for F3, and 16.9 kPa (range, 7.9–57.2 kPa) for F4 (Fig. 1).

Correlation between LS values and other variables

Liver stiffness values were positively correlated to fibrosis stages, ALT, activity grade, spleen size, prothrombin time, and age, whereas platelet count and albumin were negatively correlated to LS values (all $P < 0.05$) (Table 4). Among these variables, fibrosis stage showed the highest correlation to LS value ($r = 0.835$, $P < 0.001$). Steatosis and BMI were not correlated to LS values ($r = 0.104$; $P = 0.791$ and $r = 0.077$; $P = 0.468$, respectively).

Different performance of LSM according to alanine aminotransferase level

Although our study included patients with ALT level less than 5x ULN to exclude unreliable LSM examinations, we still found a significant correlation between ALT and LSM. Thus, we investigated the ALT effects on LS values by stratifying all patients into two groups (normal ALT group [$n = 53$] and abnormal ALT group [$n = 38$, ULN < ALT ≤ 5x ULN]).

Table 4 Correlation between liver stiffness values and other variables (Spearman's correlation analysis)

Variables	Correlation coefficient	*P*-value
Fibrosis stage	0.835	< 0.001
ALT (IU/L)	0.455	< 0.001
Activity grade	0.441	< 0.001
Platelet count (10^3/mm^3)	–0.411	< 0.001
Spleen size (cm)	0.383	0.001
Albumin (g/dL)	–0.317	0.002
Prothrombin time (INR)	0.259	0.013
Age (years)	0.282	0.007
Total bilirubin (mg/dL)	0.115	0.277
Steatosis category	0.104	0.791
Gender	–0.077	0.468
Body mass index (kg/m^2)	0.077	0.468
HCV RNA (IU/mL)	0.005	0.965

ALT, alanine aminotransferase; HCV, hepatitis C virus; INR, international normalized ratio.

Table 5 The comparison of diagnostic performance between LSM and other noninvasive models

		AUROC	*P*-value	95% confidence interval
≥ F2	LSM	0.909	< 0.001	0.844–0.974
(*n* = 50, 55.0%)	API	0.715	0.001	0.602–0.828
	APRI	0.780	< 0.001	0.676–0.885
	ASPRI	0.713	0.002	0.599–0.828
≥ F3	LSM	0.993	< 0.001	0.982–1.000
(*n* = 21, 23.1%)	API	0.858	< 0.001	0.731–0.986
	APRI	0.887	< 0.001	0.789–0.986
	ASPRI	0.862	< 0.001	0.730–0.994
F = 4	LSM	0.970	< 0.001	0.933–1.000
(*n* = 9, 9.9%)	API	0.948	< 0.001	0.893–1.000
	APRI	0.904	0.001	0.776–1.000
	ASPRI	0.957	< 0.001	0.906–1.000

API, age–platelet index; APRI, aspartate aminotransferase to platelet ratio index; ASPRI, age–spleen–platelet ratio index; AUROC, area under the receiver operating characteristic curve; LSM, liver stiffness measurement.

The performance to predict ≥ F2 and F4 fibrosis stage were slightly higher in normal ALT group than abnormal ALT group (AUROC = 0.920 for ≥ F2 and 0.960 for F4 in normal ALT group *vs* AUROC = 0.902 for ≥ F2 and 0.943 for F4 in abnormal ALT group), although statistical significance between AUROC was not identified between two groups (all $P > 0.05$).

Diagnostic performance of LSM and other noninvasive models

Table 5 and Figure 2 show the diagnostic performance and corresponding receiver operating characteristic curves of LSM and other noninvasive models for predicting ≥ F2, ≥ F3, and F4 fibrosis stage. Although LSM and other models significantly predicted ≥ F2, ≥ F3, and F4 fibrosis stage, LSM performed better than API, APRI, and ASPRI (AUROC = 0.909 *vs* 0.715, 0.780, and

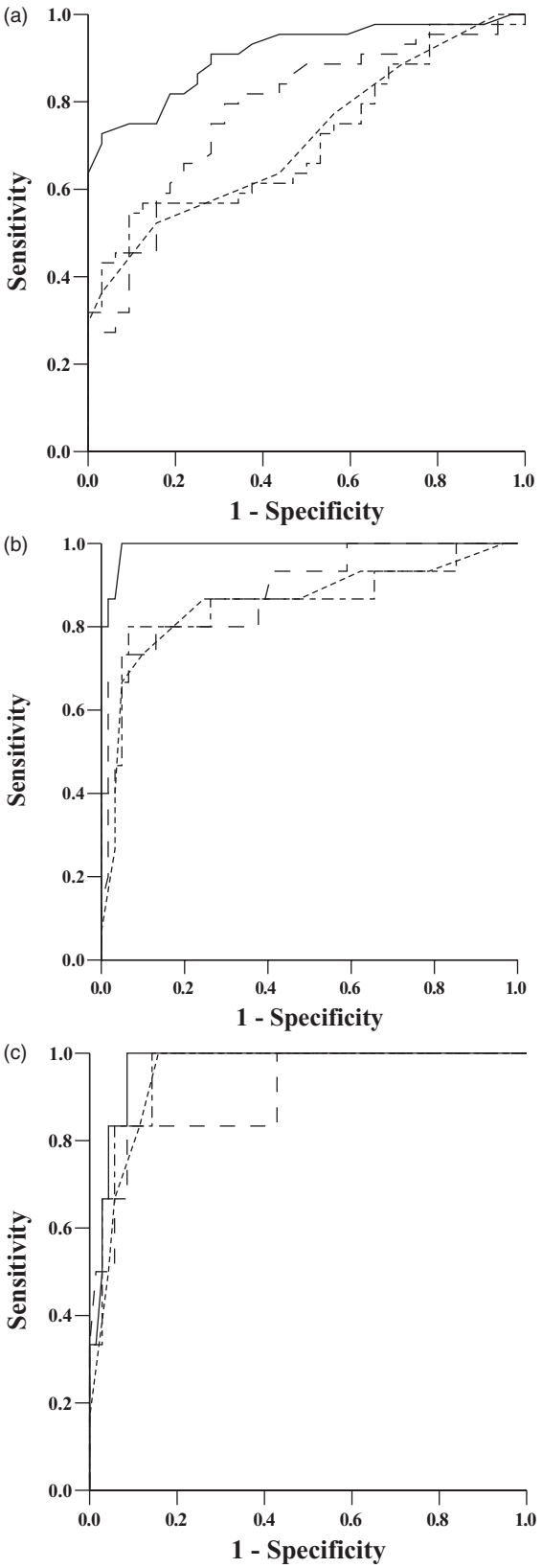

Figure 2 The receiver operating characteristic curves of LSM and other noninvasive models. (a) ≥ F2 (b) ≥ F3, and (c) F = 4. LSM performed better than API, APRI, and ASPRI (AUROC = 0.909 *vs* 0.715, 0.780, and 0.713 for ≥ F2; 0.993 *vs* 0.858, 0.887, and 0.862 for ≥ F3; 0.970 *vs* 0.948, 0.904, and 0.957 for cirrhosis, respectively). LSM, liver stiffness measurement; API, age–platelet index; APRI, aspartate aminotransferase to platelet ratio index; ASPRI, age–spleen–platelet ratio index; AUROC, area under the receiver operating characteristic curve. —, LSM; - - - - -, API; – –, APRI; ---, ASPRI.

Table 6 Cutoff value and diagnostic indexes of liver stiffness measurement ($n = 91$)

	≥ F2	≥ F3	F = 4
Number of patients, *n* (%)	50 (55.0)	21 (23.1)	9 (9.9)
†Cutoff value (kPa)	6.2	7.7	11.0
Sensitivity (%)	76.0	100.0	77.8
Specificity (%)	97.5	95.7	93.9
Positive predictive value (%)	97.4	87.5	58.3
Negative predictive value (%)	80.0	100.0	97.5
Negative likelihood ratio	0.3	0.0	0.2
Positive likelihood ratio	30.4	23.3	12.8
Correctly predicted (%)	85.7	96.7	92.3

†Cutoff value was calculated to maximize the sum of sensitivity and specificity.

0.713 for ≥ F2; 0.993 *vs* 0.858, 0.887, and 0.862 for ≥ F3; 0.970 *vs* 0.948, 0.904, and 0.957 for cirrhosis, respectively).

Optimal cutoff LS values and corresponding diagnostic indexes

The optimal cutoff LS values and corresponding diagnostic indexes are listed in Table 6. The cutoff LS values were 6.2 kPa for ≥ F2 (Se 76.0%, Sp 97.5%, PPV 97.4%, and NPV 80.0%), 7.7 kPa for ≥ F3 (Se 100.0%, Sp 95.7%, PPV 87.5%, and NPV 100.0%), and 11.0 kPa for F4 fibrosis stage (Se 77.8%, Sp 93.9%, PPV 58.3%, and NPV 97.5%).

Independent predictors for ≥ F2 and F4 fibrosis stage

In univariate binary regression analysis, age ($P = 0.041$), ALT ($P = 0.019$), platelet count ($P = 0.003$), spleen size ($P = 0.024$), and LS values ($P < 0.001$) predicted significantly ≥ F2 fibrosis stage. However, LS values were the single independent predictor for ≥ F2 fibrosis stage in multivariate analysis ($P < 0.001$, hazard ratio 4.481, 95% confidence interval 2.071–9.696). Similarly, age ($P = 0.005$), AST ($P = 0.089$), albumin ($P = 0.030$), prothrombin time ($P = 0.001$), platelet count ($P = 0.001$), spleen size ($P = 0.039$), and LS values ($P < 0.001$) were the significant predictors for F4 fibrosis stage, but LS values remained as the only independent predictor for F4 fibrosis stage ($P = 0.033$, hazard ratio 1.270, 95% confidence interval 1.020–1.583).

Disagreement between LB and LSM in patients with cirrhosis

When we set 11.0 kPa as the optimal cutoff LS value for F4 fibrosis stage according to the results of this study, disagreement was observed in seven (7.7%) out of 91 patients and LB specimens of them were reevaluated. For two patients with F4 fibrosis stage and LS value < 11.0 kPa (8.7 and 10.1 kPa, respectively), predominantly micronodular cirrhosis and severe periseptal activity were noted. For five patients with less than F4 fibrosis stage and LS value ≥ 11.0 kPa, F3 was the only fibrosis stage (100.0%) and the activity grade was 4 in three patients (60.0%). The ALT level of 5 patients with F3 and LS values ≥ 11.0 kPa was higher than the other 7 with F3 and LS values < 11.0 kPa without statistical significance (76.0 ± 35.8 IU/L *vs* 46.9 ± 19.0 IU/L, $P = 0.095$).

When we compared patients with consistent results between LB and LSM ($n = 84$) to those with inconsistency ($n = 7$), ALT level was lower (37.9 ± 17.8 IU/L *vs* 68.9 ± 33.3 IU/L; $P < 0.001$) and activity grade 4 was less prevalent (0% *vs* 40%; $P = 0.004$) in patients with consistent results between LB and LSM.

Discussion

Liver biopsy has been regarded as the 'gold standard' for assessing the severity of liver disease.[2] However, because LB is an invasive procedure with potential risk of life-threatening compications,[28] its value has been questioned. Furthermore, the accuracy of LB has been challenged due to its sampling errors and intra- and inter-observer variability that may lead to over- or under-estimation of fibrosis stage.[18,29]

As an alternative to LB, LSM was recently introduced for the noninvasive measurement of liver elasticity which has a significant correlation to liver fibrosis.[3,4,30] Until recently, most studies on the usefulness of LSM were performed in Europe.[3,4] However, because higher BMI and metabolic syndrome were revealed as significant factors which might increase LS values,[11] the validation of LSM for Asian populations with CHC showing lower BMI and lower prevalence of metabolic syndrome than Europeans is still needed. This validation will be also a critical prerequisite for the extended application of LSM to Asian countries.

In this study, we compared the performance of LSM with other simply available noninvasive models such as API, APRI, and ASPRI. Although all noninvasive models predicted liver fibrosis significantly, LSM showed the higher performance (AUROC = 0.909 for ≥ F2, 0.993 for ≥ F3, and 0.970 for F = 4, respectively) than noninvasive models. Furthermore, AUROC values of LSM were higher than those of previous studies. Castera *et al*.[3] reported that AUROC values of LSM were 0.83 for ≥ F2, 0.90 for ≥ F3, and 0.95 for F = 4, and Ziol *et al*.[4] reported that AUROC values were 0.79 for ≥ F2, 0.91 for ≥ F3, and 0.97 for F = 4. Corresponding cutoff LS values of our study (6.2 kPa for ≥ F2, 7.7 kPa for ≥ F3, and 11.0 kPa for F = 4) were slightly lower than those of previous study (7.1 kPa for ≥ F2, 9.5 kPa for ≥ F3, and 12.5 kPa for F = 4 in Castera *et al*.;[3] 8.8 kPa for ≥ F2, 9.6 kPa for ≥ F3, and 14.6 kPa for F = 4 in Ziol *et al*.[4]).

To date, several confounders of LSM such as high ALT and IQR/M have been clarified. Thus, we adopted strict criteria such as ALT level and IQR/*M*-values for enhanced LSM performance. Because high ALT level was the most important confounder of LSM in previous studies,[7,31] we excluded patients with ALT > 5x ULN. This exclusion criteria of ALT level has begun to be applied to other recent studies.[32] A previous study revealed that LSM was significantly influenced by even a small increase of ALT level.[33] Furthermore, ALT level showed a significant correlation to LSM in our study despite excluding patients with ALT > 5x ULN. Therefore, we further tested whether the performance of LSM could be affected according to ALT level. Although there was no significant difference, we found that more strict exclusion of patients with high ALT level had a potential to enhance the performance of LSM. This suggests that mild or moderate ALT elevation could still affect LSM to a certain extent. Furthermore, because IQR/M was proved as a factor of overestimated liver fibrosis,[19] we also excluded patients showing IQR/M > 0.21. These strict exclusion criteria using ALT and IQR/M can explain in part the superior AUROC values and lower cutoff LS values for predicting each fibrosis stage in our study, compared to previous studies.[3,4]

The BMI of our study population was only 22.9 ± 2.9 kg/m^2 which was much lower than that of European studies.[3,4] Although it was revealed that high BMI has an independent effect on LSM,[11] BMI seemed to have no correlation to LS values in our study population. However, the effects of high BMI should be further evaluated the considering lower proportion of high BMI (BMI ≥ 27.5 kg/m^2 in only four patients [4.4%]) and small sample size of our study. Among all patients before exclusion ($n = 128$), only two patients (1.6%) showed a success rate of LSM of less than 60% (45% and 59%, respectively) and were not excluded by our exclusion criteria. Furthermore, no one showed less than 10 valid measurements of LS values. The lower proportion of patients with success rate less than 60% and no patient with less than 10 valid measurements can be explained by the lower BMI of our study population.[34] Also, lower BMI can explain in part the lower cutoff LS values together with strict exclusion criteria of ALT level and IQR/*M*-value.[11]

We found a significant positive correlation between LS values and fibrosis stages, whereas platelet count and albumin were negatively correlated to LS values, which were similar to our previous study.[23] However, LS values were selected as the only independent predictor for ≥ F2 and F4 fibrosis stages in binary logistic regression analysis. The impact of fibrosis-adjusted steatosis on LSM could not be evaluated because all patients except one with moderate steatosis (S2) showed nonsignificant (S0) or mild steatosis (S1). To date, the effects of steatosis on LSM have still been inconclusive.[35–40] Although metabolic syndrome and its components did not seem to influence LS values in our study (all $P > 0.05$, data not shown), the influence of metabolic syndome should be validated further with large number of patients with chronic liver disease, because the portion of patients with metabolic syndrome in our study was extremely small ($n = 3$, 3.3%) and the influence of metabolic syndrome was confirmed initially in healthy subjects.

When we reevaluated the discrepancy between LSM and LB results focusing on F4 fibrosis stage, disagreement was observed in seven (7.7%) out of 91 patients. If we set 11.0 kPa for cirrhosis, five patients without cirrhosis showed LS value ≥ 11.0 kPa. All of these patients (100%) showed F3 fibrosis stage, and the activity grade was 4 in three patients (60.0%). It might mean that high activity grade increased the LS values above the cutoff indicating cirrhosis. In contrast, two patients with cirrhosis and LS

value < 11.0 kPa showed predominantly micronodular cirrhosis and severe periseptal activity histologically. However, we could not find any possible explanation for this discrepancy. In a further analysis to identify significant predictors for inconsistent results between LB and LSM, ALT ($P < 0.001$) and activity ($P = 0.004$) was selected. These results confirmed again the confounding effects of necroinflammatory activity and corresponding high ALT on LSM.

There were several limitations of the study. The interpretation of LB samples by one hepatopathologist of each center was accepted in our study. Thus, it can be a limitation of our study because of potential interobserver variability among pathologists.[41] To minimize this bias, we tried to increase the quality of LB samples by excluding those with insufficient length and number of portal tracts. The uneven distribution of patients in each fibrosis stage, espiecally low prevalence of F0 stage, might raise another issue of spectrum bias, which might influence AUROC values and corresponding cutoff LS values. Finally, although antiviral treatment was recommended for Asian patients with ≥ F1 fibrosis stage,[42] we could not obtain the optimal cutoff values for ≥ F1 fibrosis because the prevalence of patients with F0 fibrosis was extremely low ($n = 3$, 3.3%).

In conclusion, LSM showed high diagnostic accuracy in assessing liver fibrosis in patients with CHC and can be applied in South Korea.

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