Liver stiffness measurement is a potent predictor of histological fibrosis regression after hepatitis C virus clearance

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Background Most of the studies on fibrosis regression prediction were based on noninvasive fibrosis markers and differ greatly. The ‘Beijing fibrosis classification’ can use histological results to classify fibrosis into progressive or ‘nonprogressive’ according to fibrotic septal morphology. We use this standard which served as the gold standard in order to find fibrosis regression predictors.

Aim To study the predictors of fibrosis regression after hepatitis C virus clearance according to histological fibrosis staging by the ‘Beijing fibrosis classification’.

Materials and methods This was a prospective cohort study. A total of 68 patients with advanced liver fibrosis or compensated cirrhosis who achieved sustained virological response were enrolled. Patients with the Ishak scores lower than 3 seemed to have fibrosis regression. The others were divided into the fibrosis progressive group and the nonprogressive group according to the ‘Beijing fibrosis classification’. Predictors of fibrosis regression were studied by logistic regression using baseline factors and the dynamic change in noninvasive fibrosis factors.

Results Eighteen patients were assigned to the progressive group, and the others were assigned to the nonprogressive group. The baseline liver stiffness measurements (LSMs) of the progressive and nonprogressive groups were 14.35 (11.3, 27.3) kPa and 11.3 (8.3, 14.2) kPa, respectively, \( P = 0.02 \). The baseline LSM was the only predictor of fibrosis progression. With a cutoff of 11.85 kPa, the AUC was 0.71 (0.5, 0.9), and the negative predictive value was 0.92.

Conclusions The baseline LSM was found to be the only predictor of fibrosis regression, 11.85 kPa is a possible ‘hepatic fibrosis return point’. Eur J Gastroenterol Hepatol 33: 547–554

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Liver stiffness measurements and laboratory tests

LSMs and controlled attenuation parameters were assessed by transient elastography (FibroScan-502, Echosens, Paris, France). Transient elastography is performed on a patient lying supine, with the right arm elevated to facilitate access to the right liver lobe. The tip of the probe is contacted to the intercostal skin with coupling gel in the 9th–11th intercostal space at the level where a liver biopsy would be performed. The final result of a transient elastography session can be regarded as valid if the following criteria are fulfilled: (1) a number of valid shots of at least 10; (2) a success rate (the ratio of valid shots to the total number of shots) above 60%; and (3) an interquartile range (IQR, reflecting the variability of measurements) less than 30% of the median LSMs value (IQR/M 60.30%) [12].

The lower limit of serum HCV-RNA was 15 IU/mL (Amplico; Roche Diagnostics, Manheim, Germany). Liver fibrosis was also evaluated by the aspartate transaminase-to-platelet ratio index (APRI) [and the fibrosis index based on the four factors (FIB-4)]. The serum markers of liver fibrosis were calculated according to the following formulas: APRI = (AST/upper limit of normal) x 100/PLT; FIB-4 = (age x AST)/(PLT x square root of ALT).

Statistical analysis

Statistical analyses were performed using SPSS 19.0 (SPSS, Chicago, Illinois, USA) and Stata 12.0 (StataCorp, College Station, Texas, USA). Categorical data are presented as numbers (percentages). Continuous variables are reported as the mean ± standard error or median (25th percentile/75th percentile). Patient characteristics were compared between progressive and nonprogressive fibrosis patients using χ² tests for categorical variables, t-tests for variables with normal distributions, and Mann–Whitney U tests for variables with nonnormal distributions. Logistic regression analysis was used for univariate and multivariate analyses. STATA 15 was used to calculate receiver operating characteristic (ROC) curves, and the accuracy of each diagnostic criterion was evaluated according to the area under each ROC curve (AUROC). We defined the cutoff values of LSMs of liver fibrosis based on the maximum ROC curve area and a negative predictive value (NPV) of 92%.

Ethics approval

The study was conducted in accordance with the Declaration of Helsinki. The study protocol was approved by the ethics committee of Beijing Youan Hospital. Written informed consent was provided by all patients. The study was registered with the Clinical Trials no. ChiCTR1900021376.

Results

Patient enrollment and characteristics

General cases

This group originated from the population infected with hepatitis C by apheresis blood transfusion in the 1990s in a county in Hebei province, China, with a total of 2892 patients. Among them, 550 patients had advanced liver fibrosis or cirrhosis. Seventy-three patients had liver biopsy
after achieving SVR, and 68 patients were finally enrolled (Table 1; Fig. 1). There were 18 patients (26.47%) in the cirrhosis group and 50 patients (73.53%) in the advanced fibrosis group. Genotyping was dominated by types 1b and 2a, with a few classified as untyped, and the main reason for the failure of typing was low viral load. *Analysis of treatment methods.* All patients who achieved SVR using PEG ± RBV entered the nonprogressive group, and patients who achieved SVR only with the DAA entered part of the progressive group and part of the nonprogressive group. There was a significant difference in the results of the two treatment methods *P* = 0.00.

This population had stable residence, the same route and time of infection, similar living environments, the initiation of treatment at the same time, and regular follow-up, with good comparability.

### Ishak modified histology activity index grading and staging system

In Fig. 2, patient 1 and patient 2 are two patients with cirrhosis, and patient 3 is a patient with advanced liver fibrosis; the patients were followed up for 30, 23, and 27 months, respectively. The pathological results were the Ishak fibrosis 6/histology activity index (HAI) 6, fibrosis 4/HAI 5, and fibrosis 3/HAI 2, respectively, for patient 1, patient 2, and patient 3. Overall, 66.7% of patients with cirrhosis after SVR still remained as Ishak stage 5–6 (cirrhotic stage), and 33.3% no longer had cirrhosis (Ishak stage 0–4). Of the 50 patients with advanced liver fibrosis, 12% progressed to cirrhosis after SVR (Ishak stage 5–6), 12% had no change (Ishak stage 4), and 76% had fibrosis reversal (Ishak stage 0–3). The proportion of fibrosis reversal was lower in patients with cirrhosis than in advanced liver fibrosis patients (Table 2).

### Beijing fibrosis classification

The observation of the fibrotic septal characteristics of the above images revealed that the fibrotic septa contained wide/broad, loosely aggregated collagen fibers with inflammation, as shown in Fig. 2, patient 1. Scars were thin, densely compacted stroma, and the septa could be fragmented and interrupted by hepatocytes, as shown in Fig. 2, patient 3. The dynamic changes of liver fibrosis in patients with hepatitis C after antiviral treatment are similar to those in patients with hepatitis B.

Classifying the patients according to the Beijing fibrosis classification, it can be seen (Table 2) that fibrosis progressed in 33.3% and did not progress in 66.7% of patients with cirrhosis; fibrosis progressed in 16.0% and did not progress in 84.0% of patients with advanced liver fibrosis after SVR. These findings also suggest that patients with cirrhosis have a lower proportion of liver fibrosis reversal.

### The Beijing fibrosis classification and clinical indicators

According to Beijing standards, all patients were divided into progressive group, indeterminate group, and regressive group (Supplementary Table 2, Supplemental digital content 1, [http://links.lww.com/WNR/A586](http://links.lww.com/WNR/A586)). In general,
the indeterminate fibrosis group will eventually enter the regressive group; therefore, the indeterminate and regressive groups were combined into the nonprogressive group for a total of 50 patients (see Table 2). The baseline data of the indeterminate and regressive groups were significantly higher than those of the nonprogression group only in LSMs, and there was no significant difference in ALB, APRI, and FIB-4 (Table 1). Follow-up clinical data showed that albumin was significantly lower in the progression group than in the nonprogression group (42.74 ± 5.04 vs 45.02 ± 2.93 g/L, \( P = 0.03 \)), LSM, APRI, and FIB-4 were all significantly higher in the nonprogression group (Table 3), suggesting that the progression group had more severe liver fibrosis and poorer liver function.

**Baseline parameters predict fibrosis regression**

Univariate analysis (Supplementary Table 3, Supplemental digital content 1, [http://links.lww.com/WNR/A586](http://links.lww.com/WNR/A586)) suggested that the baseline LSM was the only risk factor affecting fibrosis outcome. The cutoff was 11.85 kPa, and the AUROC (95% confidence interval) for predicting liver fibrosis progression was 0.708 (0.541–0.875), as shown in Fig. 3. When the baseline LSM was less than 11.85 kPa, the NPV was 92%, suggesting a 92% likelihood of the...
nonprogression of fibrosis after SVR in these patients (Fig. 3).

Dynamic changes in indicators predict fibrosis regression

After antiviral treatment, most patients’ LSMS rapidly decreased in both the progressive and nonprogressive groups, as shown in (Fig. 4). APRI, FIB-4, and other indicators showed the same decreasing trend (data not shown), and the degree and proportion of the decrease in LSM, FIB-4, and APRI did not predict the outcome of liver fibrosis (Supplementary Table 4, Supplemental digital content 1, http://links.lww.com/WNR/A586). The combination of ΔLSM and LSM was used to predict liver fibrosis, and the prediction model was $Y = 3.45 - 0.19 \times \text{LSM} + 3.04 \times \Delta \text{LSM}$, with an AUROC of 0.781 (0.633–0.929) (Fig. 3). Compared to the LSM indicator alone, $Z = 1.23$, $P = 0.27$, the results demonstrate that the prediction model is not better than the baseline LSM alone.

Discussion

This study, based on histology, found that the baseline LSM was the only predictor of fibrosis regression, and a baseline LSM less than 11.85 kPa could be used as a predictive factor for fibrosis regression, which can also be called the liver fibrosis returnable point. This finding is particularly important from a clinical point of view, given that it can have a potential impact in the follow-up strategies after SVR.

The available evidence shows that HCV eradication with both IFN-based and IFN-free therapies can improve fibrosis and portal hypertension [13–16]. However, few studies have used paired biopsies to evaluate the influencing factors of liver fibrosis regression. Poynard et al. [7] collected data on 3010 patients with paired biopsies before and after interferon-based therapy, with a mean follow-up of 20 months and an SVR rate of 36.3%. The factors influencing recovery from severe liver fibrosis after treatment were baseline liver fibrosis stage, SVR, age, BMI, mild baseline inflammation, and low viral load. Mauro et al. [6] performed a second liver puncture after antiviral therapy in 112 liver transplant patients with hepatitis C. The results indicated that 67% of patients had fibrosis reversal: seen in 43% of patients with cirrhosis and 72–85% of patients with other stages of liver fibrosis ($P = 0.002$), with pretreatment hepatic venous pressure gradient (HVPG) and LSM being the main predictive factors. The LSM irreversible point was 25.3 kPa. Pan et al. [15] performed paired biopsies in 15 patients with advanced liver fibrosis and cirrhosis, and 13 patients improved. The post-SVR liver biopsies of only four patients showed F1–F2, while 11 patients showed F3–F4, but this study did not analyze the influencing factors of liver fibrosis regression.

As mentioned earlier, both the morphological characteristics of fibrotic septa and the consistency of clinically noninvasive liver fibrosis indicators suggest that the Beijing fibrosis classification is suitable for assessing the dynamic changes in fibrosis after SVR for hepatitis C. The classic histological classification of liver fibrosis, such as Ishak

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**Table 2.** The Ishak score and Beijing score of compensated cirrhosis and advanced fibrosis patients after sustained virological response

| Histological evaluation | Ishak fibrosis score | Beijing fibrosis classification |
|-------------------------|----------------------|--------------------------------|
|                         | 0–2                  | 3                              | 4                              | 5–6                             |
| All patients, N (%)     | 23 (33.8)            | 19 (27.9)                      | 8 (11.8)                       | 18 (26.5)                       |
| Compensated cirrhosis, N=18 (%) | 2 (11.1) | 2 (11.1)                      | 2 (11.1)                       | 12 (66.7)                       |
| Advanced fibrosis, N=50 (%) | 21 (42.0) | 17 (34.0)                      | 6 (12.0)                       | 6 (12.0)                       |
|                         | 14 (20.6)            | 9 (13.2)                       | 45 (66.2)                      |

**Table 3.** Histological and clinical characteristics of the progressive group and the nonprogressive group classified according to the Beijing standard

| Characteristics          | Progressive | Nonprogressive | $P$ value |
|--------------------------|-------------|----------------|-----------|
| PLT (per nL), mean       | 156.15 ± 49.65 | 181.19 ± 57.43 | 0.15 |
| PT(S), mean              | 11.90 ± 0.49  | 11.69 ± 0.65  | 0.70 |
| INR, mean                | 1.06 ± 0.04   | 1.04 ± 0.06   | 0.67 |
| ALT (U/L), IQR           | 17.25 (12.98–19.53) | 17.80 (13.90–24.30) | 0.59 |
| AST, IQR                 | 23.95 (18.73–35.33) | 21.90 (19.70–26.60) | 0.40 |
| ALB (g/L)                 | 42.74 ± 5.04  | 45.02 ± 2.93  | 0.03 |
| CAP, IQR                 | 264.50 (238.00–285.50) | 241.50 (208.75–282.50) | 0.09 |
| LSM, IQR                 | 12.00 (8.40–18.50) | 7.80 (6.05–9.00) | 0.00 |
| APRI, mean               | 0.62 ± 0.44   | 0.41 ± 0.18   | 0.01 |
| FIB-4, mean              | 3.01 ± 1.54   | 1.99 ± 0.75   | 0.00 |
| Inflammation HAI score, n (%) | 0.00 | 0.00 | 0.00 |
| 0–3                      | 0 (0)         | 27 (50)       | 0.00 |
| 4–6                      | 11 (78.6)     | 25 (46.3)     |
| 7–9                      | 2 (3.7)       | 2 (3.7)       |
| ≥10                      | 3 (21.4)      | 0 (0)         |
| Ishak fibrosis score, n (%) | 0.00 | 0.00 | 0.00 |
| 0–2                      | 0 (0)         | 23 (42.6)     |
| 3                        | 3 (21.4)      | 16 (29.6)     |
| 4                        | 2 (14.3)      | 6 (11.1)      |
| 5                        | 3 (21.4)      | 7 (13.0)      |
| 6                        | 6 (42.9)      | 2 (3.7)       |

APRI, aspartate transaminase-to-platelet ratio index; CAP, controlled attenuation parameter; DAA, direct-acting antiviral agent; FIB-4, fibrosis index based on the four factors; HAI, histology activity index; HCV, hepatitis C virus; INR, international normalized ratio; IQR, interquartile range; PLT, blood platelet.

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staging, focuses on the severity of fibrosis rather than the dynamic changes of fibrosis. Some patients with cirrhosis in this cohort still had Ishak stages 5–6 after SVR, but it was shown that the fibrous tissue was degrading, as indicated by thin, densely compacted stroma, etc., suggesting that fibrosis was recovering and regressing. These patients are likely to have Ishak downstaging and cirrhosis reversal in the future. The Beijing fibrosis classification makes up for the short duration of clinical follow-up. Liver fibrosis was regressive but had not reached downstaging, suggesting that the Beijing fibrosis classification is a useful addition to Ishak staging.

Our results suggest a low proportion of liver fibrosis reversal in patients with cirrhosis and a high LSM at baseline, similar to other findings. Hedenstierna et al. [8] analyzed a total of 269 patients with advanced fibrosis or cirrhosis, as determined by Fibroscan, who were followed up for a mean of 7.7 years (range, 0–20 years) after treatment with interferon for hepatitis C. Twenty-one percent of the patients with cirrhosis who were followed for more than 10 years still had advanced fibrosis. Risk factors for persistent fibrosis were pretreatment cirrhosis, old age, and BMI. In the study of Lledó et al. [9], a total of 260 HCV patients were treated with DAAs and 246 patients achieved SVR, 57.2% of whom had advanced fibrosis or cirrhosis. At SVR12, 40% of patients had significant fibrosis regression. Multivariate analysis showed that only the baseline LSM was associated with liver fibrosis reversal. Mauro et al. [6] also found the baseline differences in LSM between patients with cirrhosis with (n = 14) and without fibrosis regression (n = 20) were also statistically significant: 17.1 kPa (13.0–21.6) vs 26.6 kPa (25.3–35.6; P = 0.003), respectively. They have proposed that in patients with compensated cirrhosis and HVPG >10 mmHg and LSM >21 kPa are predictive points at which liver fibrosis cannot be reversed. Our study showed that LSM was the only factor that predicted the reversal of liver fibrosis and that liver fibrosis did not progress in 92% of patients when LSM <11.85 kPa.

LSM often shows a rapid and significant decrease after DAA treatment, but this does not represent a reversal of liver fibrosis [17]. Although, in our study, the type of treatment (INF vs INF-free) was significantly different in the progressive group and the nonprogressive group, this association was no longer significant in the univariate and multivariate analysis. And in the progressive group, the histological inflammation and fibrosis score was significantly higher than in the nonprogressive group. Enomoto et al. [18] also found significant histological inflammation of unknown cause in some patients. Additionally, improvement in liver fibrosis was not evident in the short term after achieving a sustained virologic response to direct-acting antiviral treatment. The reasons are unknown, maybe the persistent inflammation after SVR cause fibrosis progression and the amount of specimens can be expanded to further clarify.

Our results also showed that neither the degree nor the proportion of LSM reduction was effective in predicting liver fibrosis reversal. In a study by Mauro et al. [6], the median reduction in LSM was 47 and 30% in the progressive and nonprogressive groups of liver fibrosis, respectively. The percentage decrease in LSM predicted liver fibrosis regression with an AUC of 0.653; a 50% decrease predicted liver fibrosis recovery with a positive predictive value of 77.8% and an NPV of 44% and was able to correctly distinguish only 55% of patients. Therefore, LSM dynamic changes are not good indicators of fibrosis regression.
Many studies have suggested that old age, obesity, diabetes, and other factors are associated with persistent liver inflammation and fibrosis after SVR for hepatitis C [8,19]. There were no similar findings in our study, which may be related to the small sample size.

There are certain shortcomings in this study. For example, the sample size was small, and the follow-up period was only 25 months. A longer follow-up and larger sample sizes are needed to further clarify this result in the future.

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All patients were informed in writing of the use of their data for clinical research purposes and accepted.

Conflicts of interest

There are no conflicts of interest.

References

1 Blach S, Zeuzem S, Manns M, Altraif I, Duberg AS, Muljono DH, Abaalkhail, F. Global prevalence and genotype distribution of hepatitis
C virus infection in 2015: a modelling study. *Lancet Gastroenterol Hepatol* 2017; 2:161–176.

2 Takano S, Yokosuka O, Imazeki F, Tagawa M, Omata M. Incidence of hepatocellular carcinoma in chronic hepatitis B and C: a prospective study of 251 patients. *Hepatology* 1995; 21:650–655.

3 Poynard T, Moussalli J, Munteanu M, Thabut D, Lebray P, Rudler M, et al.; FibroFrance-GHPS Group. Slow regression of liver fibrosis presumed by repeated biomarkers after virological cure in patients with chronic hepatitis C. *J Hepatol* 2013; 59:675–683.

4 Tachi Y, Hirai T, Miyata A, Ohara K, Iida T, Ishizu Y, et al. Progressive fibrosis significantly correlates with hepatocellular carcinoma in patients with a sustained virological response. *Hepatol Res* 2018; 12:544–551.

5 Mauro E, Crespo G, Montironi C, Londoño MC, Hernández-Gea V, Ruiz P, et al. Portal pressure and liver stiffness measurements in the prediction of HCC in CHC patients with curative antivirals. *Hepatol Int* 2018; 12:544–551.

6 Hedenstierna M, Nangarhari A, El-Sabini A, Weiland O, Aleman S. Cirrhosis, high age and high body mass index are risk factors for persisting advanced fibrosis after sustained virological response in chronic hepatitis C. *J Hepatol* 2018; 67:1683–1694.

7 Poyaud T, McHutchison J, Manns M, Trepo C, Lindsay K, Goodman Z, et al. Impact of pegylated interferon alfa-2b and ribavirin on liver fibrosis in patients with chronic hepatitis C. *Gastroenterology* 2002; 122:1303–1313.

8 Hedenstrierna M, Nangarhari A, El-Sabini A, Weiland O, Aleman S. Cirrhosis, high age and high body mass index are risk factors for persisting advanced fibrosis after sustained virological response in chronic hepatitis C. *J Viral Hepat* 2018; 25:802–810.

9 Lledó GM, Carrasco I, Beñitez-Gutiérrez LM, Arias A, Royuela A, Requena S, et al. Regression of liver fibrosis after curing chronic hepatitis C with oral antivirals in patients with and without HIV co-infection. *AIDS* 2018; 32:2347–2352.

10 van der Meer AJ, Berenguer M. Reversion of disease manifestations after HCV eradication. *J Hepatol* 2016; 65:595–5108.

11 Sun Y, Zhou J, Wang L, Wu X, Chen Y, Piao H, et al. New classification of liver biopsy assessment for fibrosis in chronic hepatitis B patients before and after treatment. *Hepatology* 2017; 65:1438–1450.

12 Castera L, Forns X, Alberti A. Non-invasive evaluation of liver fibrosis using transient elastography. *J Hepatol* 2008; 48:835–847.

13 D’Ambrosio R, Aghemo A, Rumi MG, Ronchi G, Donato MF, Paradis V, et al. A morphometric and immunohistochemical study to assess the benefit of a sustained virological response in hepatitis C virus patients with cirrhosis. *Hepatology* 2012; 56:532–543.

14 Rockey DC. Fibrosis reversal after hepatitis C virus elimination. *Curr Opin Gastroenterol* 2019; 35:137–144.

15 Pan JJ, Bao F, Du E, Skillin C, Frenette CT, Waalen J, et al. Morphometry confirms fibrosis regression from sustained virologic response to direct-acting antivirals for hepatitis C. *Hepatol Commun* 2018; 2:1320–1330.

16 Wei L, Huang YH. Long-term outcomes in patients with chronic hepatitis C in the current era of direct-acting antiviral agents. *Expert Rev Anti Infect Ther* 2019; 17:311–325.

17 European Association for the Study of the Liver. EASL-ALEH clinical practice guidelines: non-invasive tests for evaluation of liver disease severity and prognosis. *J Hepatol* 2015; 63:237–264.

18 Enomoto M, Ikura Y, Tamori A, Kozuka R, Motoyama H, Kawamura E, et al. Short-term histological evaluations after achieving a sustained virologic response to direct-acting antiviral treatment for chronic hepatitis C. *United Eur Gastroenterol J* 2018; 6:1391–1400.

19 Desbois AC, Cacoub P. Diabetes mellitus, insulin resistance and hepatitis C virus infection: a contemporary review. *World J Gastroenterol* 2017; 23:1697–1711.