·综　　述·

Non-invasive assessment of liver fibrosis

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1 Introduction

Liver fibrosis is a scar forming process which results from the repair and proliferation of liver tissue associated with increased deposition of extracellular matrix (ECM) in liver after diverse liver injuries. A liver biopsy remains the gold standard for assessing liver fibrosis. However, liver biopsy is an invasive procedure with several limitations. Direct serum markers are used for assessing the degree of liver fibrosis and guiding treatment in clinical practice. Serum HA levels were more clinically useful for diagnosing and predicting the absence or presence of significant fibrosis than mild and moderate fibrosis. The increase of serum PIIINP levels prompted active liver fibrosis in the early stage of CLD and reflected the activity of inflammation. Serum CIV levels significantly increased with the early progression of fibrogenesis and serum laminin has a strongly positive correlation with the degree of portal hypertension. Non-patented serum marker indexes are more used in clinical practice when patented. But none of these can exactly reflect liver fibrosis.

According to a great quantity of research, the complex constituents of liver fibrosis tissue including glycoproteins, collagens and proteoglycans had different formative mechanisms. On the other hand, liver fibrogenesis is a dynamic process with different rates of progression or regression. Serum HA levels were useful for both predicting and monitoring the progression of CLD, and significantly associated with the high stage of fibrosis and cirrhosis. In contrast, the early increase of serum PIIINP levels reflected the activity of inflammation better than the degree of liver fibrosis. Serum CIV and laminin had more advantages in assessing portal hypertension. So from the viewpoint of clinical significance, when used these serum markers individually, the accuracy for diagnosing liver fibrosis was limited. Pilette et al.[1] recommended the tests constitute of HA, laminin and PIIINP for the assessment of liver fibrosis in clinical practice. Later on, the same results were confirmed by others[2].

Among a total of 2 237 references, 14 validated biomarkers have been identified between 1991 and 2007 by Poynard et al.[3]. Nine markers were not patented: PGA index (Prothrombin, Gamma Glutamyl Transpeptidase, Apolipoprotein A1), AP index (Age platelet), Bonacini index, Pohl score, Forns index, APRI index (Aspartate aminotransferase/Platelets Ratio), MP3 index (MMP1, PIINP), FIB4, and Fibroindex. Five markers were patented: Fibrotest, FibroSpect II, ELF (Enhanced Liver Fibrosis), Fibrometer, and Hepascore (Table 1).

2 Ttransient elastography(FibroScan)

Transient elastography (FibroScan; Echosens, Paris, France) is a novel, non-invasive, and rapid technique to assess liver fibrosis by the measurement of liver stiffness. It can be performed easily at the bedside or in the outpatient department with immediate results and good reproducibility.

FibroScan is a machine to assess the degree of liver fibrosis based on pulsed elastography technology by ultrasound. FibroScan includes a probe, an electronic analysis system and a control unit installed on a personal computer. The probe consists of an ultrasonic transducer mounted on the axis of a vibrator. A vibration with mild amplitude and low frequency (50 Hz) generated by the piston's pulse propagated through the liver. The pulse generates an elastic wave referred as a shear wave which propagates through the tissue. The velocity of the shear wave was measured by ultrasound, and was directly related to the stiffness of the liver, in turn, it reflects the degree of fibrosis. The stiffer the tissue, the faster the shear wave propagates. The elasticity is measured in kilopascal (kPa). The method of measuring is as follows: firstly, make the patient in the supine position, then abduct right arm over the head and breath deeply to increase the intercostals space. Ultrasound can assist to locate the position of two intercostal spaces below the upper right liver lobe at the level of the anterior or middle axillary line. The area located by ultrasound should be at least 6cm thick and have no vascular structures. The depth of measurement is 2-5cm approximately. Ten successful measurements are usually performed in each patient. At the end, we can obtain the median of 10 measurements on the screen.

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In a normal adult, the mass of liver is about 1500g. A liver biopsy sample represents a parenchymal fraction of approximately 1/25,000-1/50,000, while FibroScan measures the volume that can be approximated by a cylinder of 20mm in height and 20mm in diameter, which represents about 1% of liver volume. So we can easily found that the detected volume of FibroScan is 500 times larger than that of liver biopsy. It is a reason why LSM would be less likely to be affected by sampling error than liver biopsy[4].

The most obvious advantage of transient elastography is reproducibility compared with liver biopsy. Sandrin et al.[5] described the intra- and inter-operator reproducibility of transient elastography in 106 patients with CHC. LSM were reproducible, operator-independent and well correlated to fibrosis stage. What is more, transient elastography is simple to be learnt and operated by only one person. LSM provided the median of 10 objective measurements that can reduce the interobserver variability.

Table1 Serum markers of liver fibrosis

Name of index (yea)	Components	Diseases
PGA(1991)	Prothrombin, GGT, APOA1	ALD
AP(1997)	Platelet, age	HCV
Bonacini(1997)	Platelet, ALT, AST,	HCV
Pohl(2001)	Platelet, AST	HCV
Fibrotest/Fibrosure(2001)	A2M, haptoglobin, APOA1, bilirubin, GGT, age, gender	HCV, HBV, ALD NAFLD, HIV
Forns(2002)	Platelet, cholesterol, age	HCV
APRI(2003)	Platelet, AST	HCV
MP3(2004)	PIIINP, MMP1	HCV
FibroSpectII(2004)	A2M, HA, TIMP1	HCV
ELF(2004)	HA, PIIINP, TIMP1	Mixed diseases
Fibrometer(2005)	Platelet, AST, A2M, HA, Prothrombin, age, gender	Mixed diseases
Hepascore(2005)	A2M, HA, GGT, age, gender	HCV
FIB-4(2006)	Platelet, AST, ALT, age	HCV/HIV
Fibroindex(2007)	Platelet, AST, gamma globulins	HCV

Table 1. The indexes were proposed from 1991 to 2007 in varied liver diseases for assessment of liver fibrosis, which included nine not patented indexes and five patented indexes. PGA: Prothrombin, Gamma Glutamyl Transpeptidase, Apolipoprotein A1; AP: Age platelet; APRI: Aspartate aminotransferase/Platelets Ratio; MP3: MMP1, PIINP; ELF: Enhanced Liver Fibrosis.

2.1 Diagnosis

2.1.1 Viral liver disease and cirrhosis CHC is one of the most important causes of chronic liver disease in the world, which can lead to liver fibrosis, cirrhosis, even hepatocellular carcinoma. Non-invasive assessment by transient elastography was regarded as a reliable tool to detect significant fibrosis or cirrhosis in patients with CHC. In addition, the study by Castéra et al.[6] assessed the performance of FibroScan, Fibrotest and APRI in 183 patients with CHC. The cut off values were 7.1 kPa, 9.5 kPa, and 12.5 kPa for F≥2, F≥3, and F=4, respectively. His analysis showed that the combing of FibroScan and Fibrotest for evaluating liver fibrosis could avoid liver biopsy in majority. The AUROC curve of the combining of FibroScan and Fibrotest (or FibroScan, Fibrotest and APRI) were 0.88 for F≥2, 0.95 for F≥3, and 0.95 for F = 4. So when the FibroScan and Fibrotest results were consistent, liver biopsy confirmed them in patients of 84% for F≥2, 95% for F≥3, and 94% for F = 4. On the other hand, Ioan Sporea et al.[7] found that a cut off value of 6.8 kPa is the one that best differentiates absence or mild fibrosis (F<2) from significant fibrosis (F≥2) with PPV of 98%, NPV of 30.1%, sensitivity of 59.6% and specificity of 93.3%.

LSM was well associated with fibrosis stage. FibroScan and Fibrotest have excellent utility for the identification of HCV related cirrhosis, but lower accuracy for earlier stages. According to Ziol et al.[8] optimal stiffness cut off values were 8.7 and 14.5 kPa for F≥2 and F=4, respectively. The AUROC curves were 0.79 for F≥2, 0.91 for F≥3, and 0.97 for F=4. Subsequently, Castéra[8] considered that Castera algorithm (FibroScan and Fibrotest) has lower accuracy than biopsy for significant fibrosis. Conversely, it has significantly higher accuracy than biopsy for cirrhosis. At multivariate analysis by Colletta et al.[10] showed that among HCV carriers with normal aminotransferases (NALT), FibroScan is superior to the Fibrotest in the non-invasive methods to detect progression of fibrosis, for which with the risk factors of excess alcohol con-

sumption in the past and high viral load.

Assessment of liver fibrosis by transient elastography in HIV/HCV co-infected patients is also a promising non-invasive method. For the diagnosis of cirrhosis, AUROC curves of LSM were significantly higher than those for platelet count, AST/ALT, APRI and FIB-4[11]. Then, performed by Gregory et al. with use of cut off values of ≥9.3 kPa for fibrosis and ≥12.3 kPa for cirrhosis, 79%-83% of participants were correctly classified by LSM compared with histologic methods, and the accuracy were higher among HIV uninfected participants than among HIV infected participants. In addition, NALT levels of patients co-infected with HIV/HCV also should be monitored closely.

Regarding HBV, LSM by transient elastography is a useful method. The study by Kim et al.[12] prospectively enrolled 200 patients with CHB. They considered that different LSM cut off values according to ALT level. In patients with ALT≤ULN, the cut off values for F≥2, F≥3, and F=4 were 6.0, 7.5, and 10.1 kPa, respectively, and in those with ALT >ULN and ≤2x ULN, they were 8.9, 11.0, and 15.5 kPa, respectively. On the other hand, FibroScan showed excellent performance for the diagnosis of cirrhosis. Ganne Carrié et al.[13] described the cut off values of 14.6 kPa in 1,007 patients, the optimal diagnosis accuracy were only 74% for PPV and 96% for NPV, respectively.

2.1.2 Complications of cirrhosis Recently, the relationship between LSM and the complications of cirrhosis such as variceal bleeding, hepatocellular carcinoma and ascites has been noted. Portal hypertension is the most common complication of cirrhosis. The best technique for the assessment of portal pressure is to measure HVPG. HVPG also correlated with the occurrence of complications of CLD. In the study by Vizzutti et al.[14], a significant positive correlation between LSM and HVPG was found. The AUROC for the prediction of HVPG≥10 and ≥12 mm Hg were 0.99 and 0.92, respectively. To detect clinically significant portal hypertension using LSM cut off values of 13.6 kPa, sensitivity was 97% and specificity was 92%, and severe portal hypertension using values of 17.6 kPa, sensitivity was 94% and specificity was 94%. In addition, LSM correlated positively with the presence of esophageal varices in patients with cirrhosis, but the correlation between LSM and esophageal varices size was not found. However, FibroScan cannot replace endoscopy for oesophageal varices screening. It may help to select patients for endoscopic screening varices in up to 60% patients with cirrhosis at cut off values of 19 kPa, with sensitivity of 91% and specificity of 60%, and it has similar effect in alcoholic patients[15].

2.1.3 Other diseases The study by Nguyen et al.[16] which included 103 alcoholic patients reported that FibroScan is more effective for assessment of liver fibrosis than other sever laboratory tests (Fibrotest, Fibrometer, Hepascore, APRI, PGA, PGAA and HA). With cut off values at 5.7, 6.3, 8.4, 15 and 47.3 kPa for F0, F1, F2, F3 and F4 by FibroScan, AUROC were 0.84, 0.91, 0.90 and 0.92, respectively. The AUROC ranged from 0.66 to 0.77 for F≥1, 0.54 to 0.82 for F≥2, 0.43 to 0.88 for F≥3 and 0.56 to 0.89 for F=4. On the other hand, NAFLD is one of the most common liver diseases, which has significant risk for developing complications such as liver cirrhosis, hepatocellular carcinoma and so on. In patients with NAFLD at cut off values of 7.9 kPa, the high NPV was 97% and the modest PPV was 52%, thus transient elastography is useful as a screening test to exclude advanced fibrosis[17]. In the last few years, assessing primary biliary cirrhosis (PBC) and primary sclerosing cholangitis (PSC) by transient elastography were reported by Corpechot et al.[18]. As a promising tool, optimal stiffness cut off values of transient elastography were 7.3, 9.8, and 17.3 kPa showed F≥2, F≥3 and F=4, respectively. The AUROC curves were 0.92 for F≥2, 0.95 for F≥3 and 0.96 for F=4.

Above studies investigated the ability of transient elastography to assess fibrosis compared to liver biopsy in several disease paradigms are as summarized in Table 2.

2.2 Factors influencing accuracy of transient elastography Transient elastography is a clinically useful non-invasive method for diagnosing liver fibrosis. However, it is expensive and its limitations have been described in a several well-conducted studies. Foucher et al.[19] investigated 200 patients with CLD with varying aetiology consecutively underwent transient elastography and liver biopsy and analyzed intraobserver and interobserver agreement with the intraclass correlation coefficient (ICC). They found that the overall interobserver agreement ICC was 0.98. Transient elastography reproducibility significantly reduced ICC associated with increased body mass index (BMI) (>25 kg/m^2), steatosis, and low staging grades (F<2). In addition, high BMI (>30 kg/m^2) could reduce the success rate of the measurement with FibroScan in patients with liver transplantation[20].

When patients have the same fibrosis staging, higher ALT levels tend to have higher LSM. The AUROC curve of LSM for cirrhosis was lower among patients who had ALT above the upper limit of normal, compared with that of patients with normal ALT levels. Coco et al.[21] identified 8.3 and 14 kPa as the fibrosis ≥F2 and cirrhosis cut off values respectively; their sensitivities were 85.2%/78.3%, specificities 90.7%/98.2%, PPV 93.9%/97.8%, NPV 78.8%/81.6%, and diagnostic accuracies 87.3%/88.2%. In patients with normal ALT either spontaneous or after antiviral therapy, liver stiffness was lower than that in patients with identical fibrosis stage. As a result, transient elastography might overestimate liver fibrosis when ALT is elevated.

Transient elastography also cannot be performed in ascitic patients because the interposed fluid blocks the progression of

the shear wave. Other contraindications are pregnancy and the presence of a cardiac pacemaker because there are no safety studies on the use of transient elastography in these conditions[22]. The study by Arena et al.[23] reported that the extent of necroinflammatory activity needs to be carefully considered in future studies that aimed at further validating transient elastography, particularly in patients with absent or low stage liver fibrosis (F0-F2). LSM does not represent a reliable instrument to detect the presence of advanced fibrosis and cirrhosis in patients who present clinical picture of acute hepatitis.

Table 2 Diagnostic accuracy and cut off values in which LSM was compared with histological fibrosis stage (METAVIR system)

reference	Patients	Stage (METAVIR)	Cut off	St (%)	Sp (%)	AUCROC
Castera[6]	HCV	F ≥ 2	7.1	67	89	0.83
		F ≥ 3	9.5	73	91	0.90
		F4	12.5	87	91	0.95
Ioan Sporea[7]	HCV	F ≥ 2	6.8	59.6	93.3	0.773
Ziol[8]	Mixed	F ≥ 2	8.74	55	84	0.79
		F ≥ 3	9.6	84	85	0.91
		F4	14.5	84	94	0.97
Colletta[10]	HCV	F ≥ 2	8.74	100	100	
de Ledinghen[11]	HCV/HIV	F ≥ 2	4.5	93	11	0.72
		F4	11.8	100	93	0.97
Marcellin[24]	HBV	F ≥ 2	7.2	70	83	0.81
		F ≥ 3	8.1	86	85	0.93
		F4	11	93	87	0.93
Ganne- Carrié[13]	Mixed	F4	14.6	79	95	0.95
Nguyen[16]	ALD	F ≥ 2	7.8	80	90.5	0.91
		F ≥ 3	11	85.7	84.2	0.90
		F4	19.5			0.92
Wong[17]	NAFLD	F ≥ 3	7.9			
Corpechot[18]	PBC / PSC	F ≥ 2	7.3	82	79	0.92
		F ≥ 3	9.8	89	90	0.95
		F4	17.3	87	95	0.96
Foucher[19]	Mixed	F ≥ 2	7.2	64	85	0.80
		F4	17.6	77	97	0.96
Coco[21]	HBV/HCV	F ≥ 2	8.3	85	91	0.93
		F4	14	78	98	0.96

Table 2. These authors proposed the cut off values in varied liver diseases in which LSM was compared with histological fibrosis stage (METAVIR system). The optimum diagnostic accuracy of transient elastography always depended on the maximal sums of sensitivity, specificity and the value of AUROC curves near 1. St: sensitivity; Sp: specificity.

3 Conclusion

Cirrhosis is an advanced stage of liver fibrosis which developed from CLD, and is one of the leading causes of death worldwide. Clinicians obtain the prognosis and clinical management of CLD information depending on liver fibrosis stages by histological assessment.

Transient elastography (FibroScan) provides high diagnostic accuracy for the detection of significant fibrosis and cirrhosis, but the values of stages before cirrhosis are less clear. In addition, FibroScan can evaluate portal hypertension and predict the presence of large esophageal varices in patients with cirrhosis, then help to select patients for endoscopic screening. However, many significant questions ane not resolved. Does FibroScan have values in patients with obese, significant ascites and steatosis? Which cut off values should be used for which indication?

In conclusion, we should not only focus on the alternatives to liver biopsy. The future probably may rely on the combination of these assessment methods, for example, liver biopsy and non-invasive methods to evaluate liver fibrosis.

参考文献：

[1] Plebani M, Burlina A. Biochemical markers of hepatic fibrosis[J]. Clin Biochem, 1991, 24(2): 219-239.

[2] Pilette C, Rousselet MR, Bedossa P, et al. Histopathological evaluation of liver fibrosis: quantitative image analysis VS semi-quantitative SCOreS[J]. J Hepatol, 1998, 28(4): 439-446.

[3] Poynard T, Morra R, Ingiliz P, et al. Assessment of liver fibrosis: noninvasive means[J]. Saudi J Gastroenterol, 2008, 14(4): 163-173.

[4] Bravo AA, heth G, Chopra A. Current concepts: liver biopsy[J]. NEngl J Med, 2001, 344(7): 495-500.

[5] Sandrin L, Fourquet B, Hasquenoph JM, et al. Transient elastography: a new noninvasive method for assessment of hepatic fibrosis[J]. Ultrasound Med Biol, 2003, 29(12): 1705-1713.

[6] Castéra L, Vergniol J, Foucher J, et al. Prospective comparison of transient elastography, fibrotest, APRI, and liver biopsy for the assessment of fibrosis in chronic hepatitis C[J]. Gastroenterology, 2005, 128(2): 343-350.

[7] Ioan S, Roxana S, Alexandra D, et al. Comparison of the liver stiffness measurement by transient elastography with the liver biopsy[J]. World J Gastroenterol, 2008, 14(42): 6513-6517.

[8] Ziol M, Handra-Luca A, Kettaneh A, et al. Noninvasive assessment of liver fibrosis by measurement of stiffness in patients with chronic hepatitis C[J]. Hepatology, 2005, 41(1): 48-54.

[9] Castéra L, Sebastiani G, Le Bail B, et al. Prospective comparison of two algorithms combining non-invasive methods for staging liver fibrosis in chronic hepatitis C[J]. J Hepatol, 2010, 52(2): 191-198.

[10] Colletta C, Smirne C, Fabris C, et al. Value of two noninvasive methods to detect progression of fibrosis among HCV carriers with normal aminotransferases[J]. Hepatology, 2005, 42(4): 838-845.

[11] de Lédinghen V, Douvin C, Kettaneh A, et al. Diagnosis of hepatic fibrosis and cirrhosis by transient elastography in HIV/hepatitis C virus-coinfected patients[J]. J Acquir Immune Defic Syndr, 2006, 41(2): 175-179.

[12] Kim SU, Kim do Y, Park JY, et al. How can we enhance the performance of liver stiffness measurement using FibroScan in diagnosing liver cirrhosis in patients with chronic hepatitis B[J]. J Clin Gastroenterol, 2010, 44(1): 66-71.

[13] 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[J]. Hepatology, 2006, 44(6): 1511-1517.

[14] Vizzutti F, Arena U, Romanelli RG, et al. Liver stiffness measurement predicts severe portal hypertension in patients with HCV related cirrhosis[J]. Hepatology, 2007, 45(11): 1290-1297.

[15] Kazemi F, Kettaneh A, N′kontchou G, et al. Liver stiffness measurement selects patients with cirrhosis at risk of bearing large oesophageal varices[J]. J Hepatol, 2006, 45(2): 230-235.

[16] Nguyen-Khac E, Chatelain D, Tramier B, et al. Assessment of asymptomatic liver fibrosis in alcoholic patients using FibroScan: prospective comparison with seven noninvasive laboratory tests[J]. Aliment Pharmacol Ther, 2008, 28(10): 1188-1198.

[17] Wong VW, Vergniol J, Wong GL, et al. Diagnosis of fibrosis and cirrhosis using liver stiffness measurement in nonalcoholic fatty liver disease[J]. Hepatology, 2010, 51(2): 454-462.

[18] Corpechot C, El Naggar A, Poujol-Robert A, et al. Assessment of biliary fibrosis by transient elastography in patients with PBC and PSC[J]. Hepatology, 2006, 43(5): 1118-1124.

[19] Foucher J, Castera L, Bernard PH, et al. Prevalence and factors associated with failure of liver stiffness measurement using FibroScan in a prospective study of 2114 examinations[J]. Eur J Gastroenterol Hepatol, 2006, 18(4): 411-412.

[20] Sánchez Antolin G, Garcia Pajares F, Vallecillo MA, et al. FibroScan evaluation of liver fibrosis in liver transplantation[J]. Transplant Proc, 2009, 41(3): 1044-1046.

[21] Coco B, Oliveri F, Maina AM, et al. Transient elastography: a new surrogate marker of liver fibrosis influenced by major changes of transaminases[J]. J Viral Hepatitis, 2007, 14(5): 360-369.

[22] Paolo DP, Silvia C. Is transient elastography a useful tool for screening liver disease[J]. World J Gastroenterol, 2009, 15(12): 1409-1414.

[23] Arena U, Vizzutti F, Corti G, et al. Acute viral hepatitis increases liver stiffness values measured by transient elastography[J]. Hepatology, 2008, 47(2): 380-384.

[24] Marcellin P, Ziol M, Bedossa P, et al. Non invasive assessment of liver fibrosis by stiffness measurement in patients with chronic hepatitis B[J]. Liver Int, 2009, 29(2): 242-247.

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