Journal of Viral Hepatitis, 2009, **16,** 249–258 doi:10.1111/j.1365-2893.2009.01088.x

Noninvasive diagnosis of liver fibrosis in patients with HIV infection and HCV/HBV co-infection

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Received September 2008; accepted for publication October 2008

SUMMARY. The measurement of fibrosis stage critically affects the identification of the progression of liver disease, the establishment of a prognosis and therapeutic decision making. Liver biopsy has been the single, most useful method to determine the degree of liver fibrosis (LF), but with recognized limitations, mainly associated with its invasiveness. In recent years, alternative noninvasive methods have been developed, including imaging methods, such as transient elastometry, and assays based on serum biomarkers. This article reviews the available studies evaluating the value of various noninvasive methods for the assessment of LF in patients with HIV-infection and HBV/HCV co-infection, and makes recommendations on how to best use and combine them in clinical practice.

Keywords: elastometry, HIV/HCV co-infection, liver biopsy, liver fibrosis, serum markers.

INTRODUCTION

In patients co-infected with HIV and hepatotropic viruses, particularly in those highly immunosuppressed, chronic liver disease is now a leading cause of hospitalization and mortality [1–3]. In some countries, up to 50% of all patients with HIV are also co-infected with HCV, and about 5–7% are co-infected with HBV [4,5].

Fibrosis is a consequence of the liver's excessive healing response, an excessive response only triggered by chronic injury. Liver fibrosis (LF) is characterized by the abnormal accumulation of extracellular matrix (ECM), mostly collagen, accumulations leading to the disorganization of the liver's normal tissue architecture and, consequently, functional loss. The accurate measurement of the stage of fibrosis improves the clinician's capability to determine the progression of liver disease, reach a prognosis, and make therapeutic decisions. These fibrosis measurements are especially useful for patients co-infected with HIV/HCV, not only because of the more rapid progression of fibrosis in co-infected compared with monoinfected patients, and the overall weaker response to anti-HCV therapy in co-infected patients, but also because of the potential interactions between antiretroviral and anti-HCV drugs [6–9].

Liver biopsy, the gold standard for LF staging, was widely recommended for the assessment of the need-to-treat co-infected patients [5]. However, the limitations of liver biopsy include its invasive nature and possible complications, inadequate biopsy size, intra- and inter-observer variability, tissue fragmentation, high cost, and its low acceptance by most patients [10–12]. These limitations drove the development of noninvasive procedures to stage LF. These procedures are currently divided into two main categories: imaging methods, such as transient elastometry (TE) [13] and assays based on serum biomarkers [14]. The potential advantages of these noninvasive methods include that they are better tolerated by patients and easier for

Abbreviations: ECM, extracellular matrix; GGT, gamma glutamyltranspeptidase; HA, hyaluronic acid; LF, liver fibrosis; LS, liver stiffness; MRI, magnetic resonance imaging; NPV, negative predictive value; PPV, positive predictive value; TIMP-1, tissue inhibitor of metalloproteinase 1; TE, transient elastometry.

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clinicians, may be repeated periodically, and are less costly. However, these new noninvasive diagnostic techniques lack clear indications for their use in the clinical management of patients co-infected with HIV and hepatotropic viruses and require the establishment of recommendations for the optimization of their application in various clinical scenarios.

The FIT Foundation is a nonprofit organization that integrates physicians devoted to HIV care as well as people living with HIV/AIDS. The main goal is to discuss and update aspects of antiretroviral treatment and related issues. Recently, FIT organized a multidisciplinary meeting, attended by specialists with extensive experience in the treatment of patients co-infected with HIV/HCV/HBV (HIV-specialists, hepatologists, pathologists and community representatives), to set recommendations for the optimization of these new methods for the diagnosis and staging of the LF in patients with both HIV and chronic liver disease caused by either HCV or HBV who are seen in different clinical settings. The present document summarizes the conclusions of the meeting.

IMPORTANCE OF LIVER FIBROSIS STAGING

For patients with both HCV infection and co-infection with HIV, knowledge of their stage of LF assists in the prediction of their disease progression and also permits a more targeted monitoring through both periodic fibrogastroscopy for the detection of gastric oesophageal varices and imaging procedures to screen for hepatocellular carcinoma in patients with cirrhosis. Accurate fibrosis staging also helps determine the urgency of initiating HCV antiviral therapy, predict therapeutic response, and, in patients with mild or advanced LF, staging provides longitudinal information. As a result of the higher prevalence of severe hepatotoxicity in patients with advanced LF, fibrosis staging of patients with co-infection assists the selection of antiretroviral therapy.

The usefulness of LF staging has been challenged when deciding on the urgency of initiating peginterferon and ribavirin therapy, because of the timeline for the development of new HCV drugs such as HCV polymerase and protease inhibitors. Although these drugs, even in their early stages of development, inhibit HCV replication they have not yet been studied in patients co-infected with HIV and the establishment of their usefulness for this population may take time. Also unknown, among many others, is both the impact of these drugs on resistance mutations and their interaction with antiretrovirals.

In summary, LF staging is considered of the utmost importance both for physicians and patients. The fibrosis stage provides the physician important information for the taking of therapeutic decisions as well as for other management issues. For patients, LF constitutes the best prognostic predictor of liver disease, as well as a useful criterion with which to determine their suitability for antiviral therapy.

LIVER BIOPSY

Liver biopsy has been considered the gold standard for the diagnosis of liver damage (necro-inflammation and fibrosis) associated with chronic viral hepatitis. However, it is an invasive technique, with well established contraindications, that requires the patient's cooperation [15]. The most common adverse effects of liver biopsy are pain in 30% of patients (20% moderate, and significant pain that requires intravenous analgesics or opiates in 3%); and vasovagal syndrome in 2%. Other adverse effects include serious complications, such as hemoperitoneum, pneumothorax, bile peritonitis, and puncture of hollow viscera in 0.57%. More than 90% of these complications appear within 2–24 h, with hemobilia being the complication that appears late. Although mortality is decreasing, it is not negligible, occurring in 1 out of 10 000 to 12 000 procedures [16].

Although liver biopsy provides a direct measure of LF, its main drawbacks are sampling errors and inter-observer variability. Some years ago, a study analysing the inconsistencies between the results of two laparoscopic biopsies – taken from both hepatic lobes with an assessment by two independent pathologists – showed that 33% of patients undergoing biopsy had differences in their fibrosis stage ≥1 – between their right and left hepatic lobes, and up to 10% of biopsies had significant variation in the interpretation of their findings. Two additional studies, demonstrated the importance of the variability in results associated with the size of the liver biopsy core. Regev *et al.* concluded that 65–75% of the cores between 1.5 and 2.5 cm long showed the same histological data as surgical specimens [12]. Colloredo *et al.* found that a smaller size of hepatic core led to a higher prevalence of moderate stages of fibrosis to the detriment of more advanced stages [17].

In addition to fibrosis measurement, liver biopsy is the only method that provides information on both necrotic and inflammatory activity, as well as steatosis, and biopsy is also the only method that can identify the concomitant causes of liver disease. The usefulness of measuring the degree of inflammation and necrosis is based on the correlation with the progression of fibrosis, as measured by units of fibrosis per year. If liver biopsy shows mild necro-inflammation, then the progression of fibrosis over the next 5 years will be low [18]. Although it may be important from this perspective, no clinical decisions are taken based on these histological findings. With few exceptions, management decisions are largely based on fibrosis staging with little impact of necroinflammatory activity or the degree of steatosis. Similarly, most reports have shown the low frequency of significant parenchymal diseases that are diagnosed by liver biopsies performed for the assessment of chronic hepatitis.

In conclusion, liver biopsy remains the method of choice for the assessment of LF, and is still the gold standard for assessing the efficacy of noninvasive methods for predicting fibrosis stage. The main disadvantages of liver biopsy are the

morbidity associated with the procedure and its low acceptance by patients. Although liver biopsy can provide additional histological information, these findings usually have little impact on the clinical management of patients.

SERUM MARKERS

The ideal features of a serum marker to be used as an alternative to liver biopsy would include the following: a marker should be liver-specific, independent of metabolic alterations, easy to perform, and minimally influenced by impaired urinary and biliary excretion. These ideal markers also should reflect fibrosis in all types of chronic liver disease, correlate with matrix content, be sensitive enough to discriminate between different stages of fibrosis from chronic hepatitis to cirrhosis and they must also reflect the response to successful antifibrotic therapy [19].

Two types of serum markers of LF are currently in use: (a) indirect markers reflecting alterations in hepatic function but that do not directly reflect ECM metabolism, such as platelet count, coagulation studies and assessment of aminotransferases (Table 1); and (b) direct markers reflecting qualitative and quantitative changes in ECM macromolecules, some of which reflect fibrogenesis and others fibrolysis (Table 2). Yet to date, the utility of these markers has been evaluated mostly with HCV-monoinfected patients and have received less attention by clinicians treating co-infected populations.

Indirect serum markers of liver fibrosis

Forn's index

Forn's index discriminates between patients with and without significant LF. In a study analysing a cohort of 476 consecutive untreated patients (estimation group, $n = 351$, validation group, $n = 125$) with chronic hepatitis C who underwent a liver biopsy [20], the authors constructed both a model and a scoring system combining the following: age, gamma glutamyltranspeptidase (GGT), cholesterol and platelet count. The area under the ROC curve (AUROC) was 0.86 for the estimation group and 0.81 for the group validating the presence of significant fibrosis (>F2). Two cut-off values were chosen to identify absence (<4.2) and presence (>6.9) of significant fibrosis. In the validation group, using the lower cut-off, presence of significant fibrosis could be excluded with high accuracy [negative predictive value (NPV) 96%]. Using the higher cut-off score the positive predictive value (PPV) was 66%. The authors concluded that this index accurately predicts the absence of significant fibrosis and might render liver biopsy unnecessary in more than one-third of patients with chronic hepatitis C. In addition, the Forn's index has been validated for patients with HIV infection [21], although a recent study including 272 patients with co-infection – from a single centre – showed Forn's index to be less accurate for the identification of significant fibrosis [22].

Table 1 Indirect serological markers for liver fibrosis

Classification	Marker name
Routine clinical measurements	AST/ALT ratio
	Platelet count
	GGT
	Prothrombin index
	PGA index
	Total bilirubin
	Albumin
Acute-phase protein	Alpha2-macroglobulin
	Haptoglobin
	Apolipoprotein A1

GGT, gamma glutamyl transpeptidase; PGA, prothrombin time, gamma glutamyl transpeptidose and serum apolipoprotein A1.

Table 2 Direct serological markers of liver fibrosis

Classification	Marker name	Remarks
ECM component	HA	Glycosaminoglycan
	PIIINP	Collagen propeptide
	Procollagen I	Collagen propeptide
	Collagen IV	
	Laminin	Glycoprotein
ECM regulatory enzymes	YKL-40	Chitinase
	MMPs	Degradation enzymes
	TIMPs	Inhibitors of ECM degradation enzymes (MMPs)

ECM, extracellular matrix; PIIINP, procollagen III N terminal peptide; HA, hyaluronic acid; MMPs, matrix metalloproteinases; TIMPs, tissue inhibitors of metalloproteinases.

Fibrotest

Fibrotest assesses the following: alpha2 macroglobulin, alpha2 globulin (or haptoglobin), gamma globulin, apolipoprotein A1, GGT and total bilirubin [23]. Results from each test are formulated to determine three categories of fibrosis: mild (METAVIR F 0-1), significant (METAVIR F2-4) and indeterminate fibrosis. The analysis was performed during an initial 1-year period of 205 patients and then tested for a second 1-year period with 134 patients. The areas (SD) under the ROC curves did not differ ($P = 0.44$) neither for the first [0.836 (0.430)] nor for the second-year groups [0.870 (0.340)]. Detection of significant fibrosis – F2 or greater – had a 75% sensitivity and an 85% specificity. Correct identification of disease as either mild or severe was made in 46% of patients, overall.

Although Fibrotest's score has been validated in other hepatitis C cohorts, an independent study did not achieve the same results [24]. Fibrotest has been evaluated in the population with HIV/HCV co-infection. In a study of 130 patients, a multivariate analysis using this five marker index resulted in an AUROC of 0.856. Its PPV was 86% for scores greater than 0.60, with an NPV of 93% for scores of 0.20 or less. The authors concluded that in patients with HIV/HCV co-infection, Fibrotest accurately predicts significant fibrosis, and may substantially reduce the need for liver biopsy [25].

APRI

The APRI test refers to the AST to platelet ratio index [26]. This model was developed to predict both significant fibrosis (Ishak score ≥ 3) and cirrhosis (Ishak score 5 or 6) in 270 consecutive patients that were HCV treatment-naïve, and underwent liver biopsy. The AUROC for APRI for the prediction of significant fibrosis and cirrhosis in the training set were 0.80 and 0.89, respectively. In the validation set, the AUROC for APRI for the prediction of significant fibrosis and cirrhosis were 0.88 and 0.94, respectively. Based on the ROC, 2 cut-off points were chosen to predict the absence (≤0.50) or presence (>1.50) of significant fibrosis. The PPV and NPV of an APRI of 0.50 were 64% and 90%, and the corresponding values for an APRI of 1.50 were 91% and 65%, respectively. Similarly, two cut-off points were chosen to predict the absence (≤1.00) or presence (>2.00) of cirrhosis. For the prediction of cirrhosis, the PPV and NPV of an APRI of 1.00 were 35% and 100%, and the corresponding values for APRI of 2.00 were 65% and 95%, respectively. The authors concluded that in patients with chronic hepatitis C, the APRI test can identify with a high degree of accuracy both significant fibrosis and cirrhosis. The APRI test has also been validated by independent groups in co-infected patients [21,27–32].

In one of these studies, APRI performed worse showing an AUROC of 0.71 [27]. The results of another study suggest that different cut-offs may be needed for the APRI in co-infected patients [32,33].

FIB-4

In patients with HIV-HCV, FIB-4 uses routine laboratory tests to predict liver histology, as can be observed in the APRICOT trial, a multi-centre study of HCV therapy in patients with HIV/HCV co-infection [28]. By multivariate logistic regression analysis, a simple index was developed including age, AST, ALT and platelet count. The AUROC of the index was 0.765 for differentiation between Ishak stage 0–3 and 4–6. At a cut-off of <1.45, the NPV to exclude advanced fibrosis (stage 4–6) was 90% with a sensitivity of 70%. A cut-off of >3.25 had a PPV of 65% and a specificity of 97%. The authors concluded that FIB-4 can accurately predict hepatic fibrosis and may reduce the need for liver biopsy in most patients with HIV/HCV co-infection. The test, however, has not been validated to predict F2, a cut-off value for LF with significant clinical utility.

HGM1 and HGM2

HGM1 and HGM2, two simple models based on routine laboratory data, were developed in order to discriminate between mild or no fibrosis and significant fibrosis and also to predict advanced fibrosis in a consecutive series of 296 patients co-infected with HIV/HCV (training set = 226, validation set = 70) [34]. By multivariate logistic regression analysis, the authors developed the HGM-1 index to predict significant fibrosis and the HGM-2 index to predict advanced fibrosis. HGM-1 was based on platelet count, AST and fasting serum glucose. HGM-2 was based on platelet count, INR, ALP and AST. The AUROCs of the HGM-1 index for the estimation group and the validation group were 0.807 and 0.712, respectively. Two cut-off values were chosen to identify mild or no fibrosis (<0.316) and significant fibrosis (>0.848). At a cut-off of 0.316 the NPV to exclude significant fibrosis was 54.5% with a sensitivity of 89.1%. The cut-off of 0.848 had a PPV of 93.3% and a specificity of 95.8%. The AUROCs of the HGM-2 index for both the estimation group and the validation group were 0.844 and 0.815, respectively. Two cut-off values were chosen to identify absence ($P < 0.138$) and presence ($P > 0.598$) of advanced fibrosis. At a cut-off of 0.138, the NPV to exclude advanced fibrosis was 92.3% with a sensitivity of 89.5%. The cut-off of 0.598 had a PPV of 64.3% and a specificity of 90.2%. In patients co-infected with HIV/HCV, the authors concluded that HGM-2 accurately predicts advanced fibrosis, but that HGM-1 was less accurate at predicting the absence of significant fibrosis. In this study, HGM-1 and HGM-2 compared favourably with the results of Forn's index, APRI and FIB-4.

Direct markers of liver fibrosis

The most commonly reported direct markers of ECM turnover are listed in Table 2. As mentioned before, none of them are liver-specific and all are affected by changes in metabolism and excretion. The characteristics of individual markers are summarized in Table 3. We will focus on the combination of assays that are used in clinical practice.

FibroSpect

In a large retrospective cohort study of 696 patients with HCV, McHutchinson and his group evaluated the FibroSpect assay [35]. The assay involves three parameters: hyaluronic acid (HA), tissue inhibitor of metalloproteinase 1 (TIMP-1) and alfa2-macroglobulin. All patients could be evaluated by FibroSpect with no indeterminate values, which is an advantage of this assay. The three-marker panel reliably differentiated patients with chronic hepatitis C with moderate/severe fibrosis from those with no/mild fibrosis (combined AUROC = 0.831), although an accurate delineation between stages was not possible.

Table 3 Characteristics of selected individual direct markers of fibrosis

Marker	Comment	Reference
HA	Hyaluronic acid is a large glycosaminoglycan found in most body tissues. In the liver it is produced by HSC and serves as an integral component of the ECM. Numerous studies have found a good correlation between levels of HA and severity of liver disease. The clinical utility of HA lies in its excellent ability to exclude cirrhosis. The test is less useful, however, for differentiating earlier stages of fibrosis.	[24–28]
Type IV collagen and PIIINP	Both type IV collagen and PIIINP have been found to be good predictors of fibrosis in patients with HCV although their abilities to differentiate fibrosis score 4 from fibrosis score 0–3 are inferior to HA	[26,29,30]
MMPs and TIMPs	MMPs are proteolytic enzymes secreted by HSC and are involved in the destruction of ECM. TIMPs block this process. Increased levels of MMPs and TIMPs have been found to correlate well with cirrhosis. One report indicates that MMPs levels can determine the stage of inflammation but not the grade of fibrosis.	[31–33]
Laminin	A basement membrane glycoprotein, that has been found to perform well in detecting significant fibrosis from HCV	[34]
YKL-40	A glycoprotein found in cartilage and liver involved in tissue remodelling and degradation of ECM. Preliminary results are promising, but more data are needed for this new marker	[30]

ECM, extracellular matrix; PIIINP, procollagen III N terminal peptide; HA, hyaluronic acid; MMPs, matrix metalloproteinases; TIMPs, tissue inhibitors of metalloproteinases.

European liver fibrosis assay

The European liver fibrosis group reported a novel assay in an international multi-centre cohort of 1021 patients with hepatitis C, NAFLD and alcoholic liver disease [36]. An algorithm was developed utilizing age, HA, PIIINP and TIMP 1. For significant fibrosis the AUROC was 0.804. The sensitivity for the detection of Scheuer stage 3 or 4 fibrosis was 90% at a threshold score of 0.102, yielding an NPV of 92%. The algorithm achieved a similar level of sensitivity and specificity when compared with the scoring of three different pathologists, providing evidence that it could be used with similar accuracy in different settings.

SHASTA

In a cohort of 95 patients co-infected with HIV/HCV, this biomarker assay was developed utilizing HA, AST and albumin (SHASTA). For fibrosis stage F3–F6, the AUROC was 0.878. As with other biomarker assays, the SHASTA Index accurately staged mild and advanced fibrosis [27].

Hepascore

Hepascore is a model of four serum markers (bilirubin, GGT, HA, alpha2-macroglobulin) plus age and sex developed at time of liver biopsy in 117 untreated patients with hepatitis C (training set) and validated in 104 patients from other institutions. Both multivariate logistic regression and AUROC analyses were used to create a predictive model for significant fibrosis (METAVIR F2, F3 and F4), advanced fibrosis (F3 and F4) and cirrhosis (F4). The model produced AUROCs of 0.85, 0.96 and 0.94 for significant fibrosis (METAVIR F2, F3 and F4); advanced fibrosis (F3 and F4) and cirrhosis, respectively. Among the validation set, the AUROC for significant fibrosis, advanced fibrosis and cirrhosis were 0.82, 0.90 and 0.89, respectively [37].

Clinical utilization of serum markers

Currently, multiple different noninvasive indexes of hepatic fibrosis probably have the same diagnostic ability. No single marker fulfils the requirements of an ideal serum marker, and a combination may have to be used particularly in patients whose initial evaluation with a simple test results in a score that falls within the indeterminate zone. They can effectively identify patients with advanced liver disease but perform less well in patients with mild disease and cannot accurately discriminate between individual stages of fibrosis. In patients with HIV–HCV co-infection, these markers may be useful in the evaluation of fibrosis. However, improvement in their diagnostic accuracy is required before they can completely supplant liver biopsies. Prospective studies are

required to determine the potential utility of the marker panel in guiding treatment decisions and following-up disease progression.

TRANSIENT ELASTOGRAPHY (FIBROSCAN®)

The ultrasound waves spreading through a physical environment allows the measurement of LF. The speed of transmission of these ultrasounds through a specific environment varies with the density and the elasticity of this environment. Thus, the speed of transmission can be calculated using a probe which emits and receives ultrasound waves. Using a physical equation, the hepatic elasticity can be computed based on the transmission speed, in such a way that the higher the speed, the lower the elasticity and the higher the stiffness, the opposite quality to elasticity [38]. There is a strong correlation between stiffness and LF. Consequently, we can estimate the degree of LF based on liver stiffness (LS) [14]. That is the basis of transient elastography, a name that may not be proper. In fact, the procedure does not obtain an image, but a measurement. Therefore, TE could be a more appropriate denomination.

Correlation between LF and LS in patients with chronic hepatitis C

A number of studies have shown that LS measured by TE correlates with LF in patients mono-infected with HCV ($r = 0.73$) [39]. The AUROC of LS for the presence of significant LF (F2–F4) is 0.82 (95% CI 0.74–0.88), according to a recent systematic review of four studies [40]. For cirrhosis, this figure is even higher: 0.95 (0.87–0.99) [40]. The PPV and the NPV have changed from one study to another, depending on the cut-off values chosen. For 7.1 kPa, the PPV was 95% and NPV 48% [40]. A cut-off of 12.5 kPa showed a PPV of 77% and NPV of 95% for the diagnosis of cirrhosis.

The correlation between LS and LF and the performance of TE in patients with HIV and HCV co-infection are similar to those found in subjects mono-infected with HCV (Fig. 1) [41]. The AUROC for F2–F4 is 0.87 (95% CI 0.84–0.93), and for F4, 0.95 (0.92–0.99). A cut-off of 7.2 kPa has a PPV 88% and a NPV of 75%. For cirrhosis, the cut-off of 14.6 kPa yields a PPV of 86% and a NPV of 94% [41].

The main limitation of TE is its relatively poor ability to discriminate between mild (F1) and moderate-to-advanced LF (F2–F3). This is a major issue, as therapeutic decisions in patients with F1 and in those with F2–F3 are often different. Using 7.2 kPa as cut-off value, 24% of patients with lower LS, and, therefore classified as F0–F1, show LF ≥ F2 on biopsy; 8% of the former subjects actually had F3 [41]. In the case that this figure is used in the selection of patients for HCV treatment, a substantial proportion of subjects with moderate-to-severe LF would remain untreated. Moreover, 17% of patients with LS ≥ 7.2 kPa show F0 or F1 on biopsy

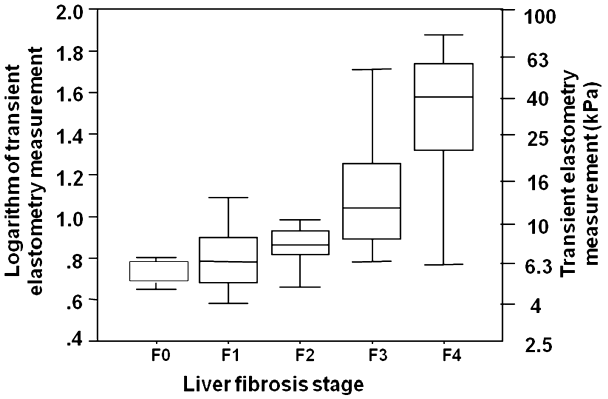

Fig. 1 Distribution of transient elastometry measurements according to liver fibrosis stages ($n = 169$) (T-$\beta = 0.64$; $P < 0.001$) [37].

[41]. However, if the decision in this case was to start treatment, the mistake would be less relevant.

Use of TE to determine HCV therapy for patients with HIV co-infection

It is a common practice to restrict HCV therapy for patients with HIV co-infection to those who have LF ≥ F2. This recommendation is included in some international guidelines, particularly for patients with predictors of nonresponse, such as genotype 1, genotype 4, or those with a high plasma HCV RNA load [42,43]. It is therefore critical to know if a patient has LF ≥ F2. Given the above-stated limitations of TE, there has been a search for new cut-off values which allow the reliable identification of at least a substantial portion of patients with and without such a degree of LF. This topic has been studied in 197 patients in whom TE and liver biopsy had been carried out less than 1 year apart. Patients were randomly assigned to an elaboration group ($n = 99$) or to a validation group ($n = 98$). Using ROC curves, the values 6 and 9 kPa were selected. A measurement equal to or lower than 6 kPa was observed in 31% of the total population. This cut-off value yielded a NPV of 90% for LF ≥ F2 and 100% for LF ≥ F3 in the validation group. LS ≥ 9 kPa was found in 41% of the total population. This figure provided a PPV of 87% for LF ≥ F2 and a NPV of 100% for F0 in the validation group. In subjects with LS ≥ 9 kPa therapy for HCV could be prescribed, but no patient without LF would undergo treatment. In those with a LS < 6 kPa therapy could be deferred and sequential measurements of LS carried out. Thus, no patient with advanced LF would remain untreated. If the LF progressed, the progression would be detected by subsequent TE examinations [44].

The major drawback for these cut-offs is that as much as 28% of the patients show a LS from 6.1 to 8.9 kPa, results that would be considered indeterminate. In these cases, blood markers of fibrosis and the virologic characteristics of

the patient – genotype and plasma HCV RNA load – should be taken into account in reaching a therapeutic decision. Ultimately, some of these patients would require a biopsy. Accordingly, liver biopsy would still be needed in 19% of candidates with HIV/HCV co-infection to decide on hepatitis C therapy [44].

Factors affecting TE results

Obesity is a major limitation in the performance of TE. Because subcutaneous fat interferes with the spreading of ultrasound waves, it is much more difficult for TE to be successfully carried out in obese patients. TE may even be impossible in the extremely overweight [45,46]. Technical improvements in the probes will hopefully overcome this problem in the future. Other factors that reduce the success rate of TE examinations are lack of examiner experience and the age of the patient, making examinations more difficult in older patients [45]. Likewise, ascites or pleural effusion makes TE difficult or impossible, as fluid interposes between the chest wall and the liver.

Little is known about factors influencing the reliability of measurements obtained by TE. Intra-exam variability, for example, caused by probe placement on different sites of the chest wall has not been studied. The following are also unknown: inter-examiner variability, the impact of the examiner's experience and that of the presence of liver steatosis. A high degree of liver inflammation reduces the concordance of TE and biopsy [46]. Finally, it remains unclear if the dispersion and the success rates of the measurements obtained influence the reliability of TE results.

Other uses of TE: TE in other liver diseases

TE has also been proven to be useful for the diagnosis of cirrhosis caused by other than the hepatitis C aetiologies in patients without HIV infection, which include the following: hepatitis B, alcohol, or non alcoholic steatohepatitis. The yielding of the procedure seems to be slightly poorer in hepatitis B, but it is still very high [AUC (95% CI) = 0.90 (0.77–0.96)] [47]. Perhaps, higher cut-off values, around 20 kPa, should be used in patients with alcoholic cirrhosis and non alcoholic steatohepatitis [48].

An LS measurement with TE provides some data that liver biopsy does not. It has been reported that RH strongly correlates with both the Child-Pugh score [39] and the hepatic venous pressure gradient [49]. TE can therefore be useful to predict the presence of oesophageal varices, particularly those with high risk for bleeding, which should receive prophylactic therapy [50]. It has been found that a cut-off point of 21.6 kPa has a NPV of 100% for varices F2–F3 or F1 with red wale signs [51]. This cut-off point can be used to select patients to undergo endoscopic screening of varices. We could thus avoid endoscopic examinations from which a therapeutic decision would not be made. Likewise, it is possible that the LS value is associated with the risk for hepatocellular carcinoma [39], but this issue requires further clarification.

Balance of benefits and limitations of TE

TE is more representative than biopsy, because it explores a hepatic area 100-fold larger than biopsy. It also is a noninvasive procedure, not hazardous or troublesome for patients and, consequently, very well tolerated and suitable for sequential determinations. TE can be used in liver diseases other than chronic hepatitis C. LS provides information that is not obtained from biopsy, such as data on liver function, portal hypertension or the presence of oesophageal varices.

Conversely, TE does not provide information on the heterogeneity of fibrosis throughout the liver. Likewise, the reproducibility of TE is not absolute, a drawback shared with biopsy. Unlike histological examination, it does not give data about liver inflammation or necrosis. Discrimination between F1 and F2–F3 may be difficult with TE and its performance in overweight patients is low. TE should be suitable for monitoring fibrosis changes, but this issue has not been proven to date. The equipment currently used is expensive to purchase and, but especially, to maintain, although it is cost-effective, compared with biopsy. Hopefully, costs will decrease, as alternatives to the devices presently used are marketed. In addition, TE is not yet available in some countries with a large population of co-infected patients, such as the USA.

MAGNETIC RESONANCE IMAGING

Magnetic resonance imaging (MRI) enhanced with superparamagnetic iron oxide or double enhanced spoiled gradient echo sequences has been evaluated for the detection of hepatic fibrosis. Preliminary results have shown good accuracy [52], as well as a good correlation with the Child-Pugh score in a small number of patients with cirrhosis [53]. Although MRI is a promising technique for these patients, no recommendation can yet be made because of the following: the number of patients studied has been too small, the studies have not been validated independently, and the cut-offs have not been defined.

FIBRO-CT

Recent, preliminary data of the optical digital analysis of CT images suggest that they can be effective in determining both the stage and distribution of LF in patients with chronic hepatitis C. The ROC curve to diagnose significant fibrosis using Fibro-CT was 0.83. Interestingly, the correlation between LF and Fibro-CT values was higher in patients with homogeneous distribution of fibrosis [54].

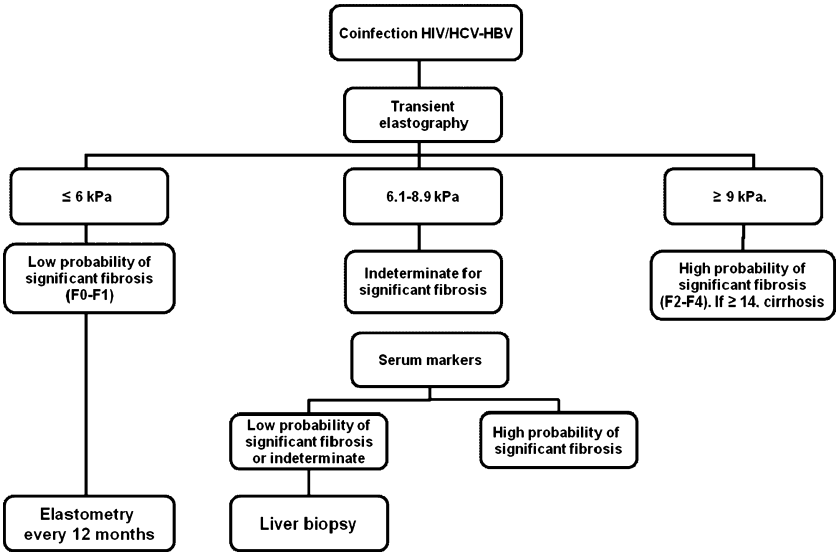

Fig. 2 Algorithm for evaluation of liver fibrosis in HIV co-infected patients.

RECOMMENDATIONS

With the available information on the advantages and disadvantages of liver biopsy *vs* alternative methods (serum markers, transient elastography), given the current status of anti-hepatitis therapies, the following recommendations may be established in order to optimize the indication for these tools in various clinical situations involving patients infected with HIV (Fig. 2):

1. All HIV-infected patients with chronic infection by HCV or HBV must have initial staging of their LF. This initial assessment of fibrosis is useful for making decisions regarding therapy for HIV and hepatitis, as well as for prognostic and other management aspects.
2. In patients co-infected with HCV, transient elastography is considered the method of choice for the initial evaluation and the follow-up of LF (there is not enough information for patients with HBV co-infection). These patients may be classified, according to the results obtained, into those with either a low probability of significant fibrosis (≤6 kPa, equivalent to F0-F1 in the Metavir score) or those with high probability of significant fibrosis (≥9 kPa, equivalent to F2-F4 in the Metavir score). If a measurement ≥14 kPa is obtained, the probability of cirrhosis is high.
3. A transient elastography measurement >6 and <9 kPa is considered indeterminate, and should be further evaluated by additional methods. It is considered that serum markers should be used first in this situation. If serum markers show a high probability of significant fibrosis, then no further evaluation would be needed at that time. If serum markers show a low probability of significant fibrosis or give indeterminate results, liver biopsy should be performed in order to confirm the fibrosis stage.
4. These recommendations do not intend to establish the criteria for when to initiate therapy for chronic hepatitis. Physicians make their therapeutic decisions based on multiple factors, of which the degree of LF may be only one. The proposed algorithm may be of help for the universal assessment of fibrosis, given the possibility of re-evaluating patients periodically.
5. Patients with a high probability of cirrhosis, based on noninvasive methods, must be followed-up on a regular basis to prevent complications, and the development of hepatocellular carcinoma or for evaluation as candidates for transplantation.
6. In patients with a low probability of significant fibrosis, based on noninvasive methods, new evaluations of LF must be made at least annually.

ACKNOWLEDGEMENT

We are thankful to David Peck for editorial assistance.

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