FibroTest & FibroMax Scientific Publications

Key publications for 2013

FibroTest: a prognosis also in HBV
Non-invasive tests for fibrosis and liver stiffness predict 5-year survival of patients chronically infected with hepatitis B virus. Aliment Pharmacol Ther. 2013 Apr 5
Authors proposed to evaluate the 5-year prognostic value of FibroTest, liver stiffness by transient elastography, and other non-patented tests for liver fibrosis (APRI, FIB-4), and liver biopsy, in a prospective cohort of 600 patients with chronic hepatitis B. Survival was significantly decreased in patients diagnosed with severe fibrosis, whatever either by FibroTest or liver stiffness (P < 0.0001), or liver biopsy (P = 0.02). The 5-years overall survival as per FibroTest in patients without cirrhosis was 97%, but only 49% in patients with cirrhosis. FibroTest had a high significant prognostic value without differences with transient elastography but with superior applicability in decompensated patients with ascites. Noninvasive prognostic tools as FibroTest may help physicians to early assess prognosis and specific treatments, such as liver transplantation.

FibroTest compared to transient and supersonic elastography
The aim was to compare the applicability (the measurement of reliable results) and performances of SWE for the diagnosis of fibrosis as compared with FibroTest and liver stiffness by transient elastography applying a methodology without gold standard. FibroTest applicability was 98%, significantly higher than that of both imaging methods: SWE (90%) or transient elastography (91%). Despite a lower SWE applicability of FibroTest, it was greater than that of TE in patients with ascites. The best performances for early stages were for FibroTest; SWE had a low performance for discrimination between F0 and F1. Performances for the diagnosis of cirrhosis were similar between FibroTest, LSM and SWE when elastography.

FibroTest-ActiTest improved during maintenance therapy
EPIC-3 included patients with F2-F3 METAVIR fibrosis scores who failed retreatment and that were randomized to PegIFN alfa-2b (0.5 microg/kg/week) or observation for 36 months. Repeated blinded liver biopsies were obtained before retreatment and after maintenance therapy and concomitantly FibroTest and ActiTest. Using semi-quantitative paired biopsy, despite that maintenance therapy does not significantly improve fibrosis stage compared with observation, however, significantly more patients receiving PEG-IFN alfa-2b experienced an improvement in necroinflammatory activity as assessed by paired biopsies. Using continuous scores as FibroTest significantly more observed patients showed a worsening in fibrosis score compared with those receiving PEG-IFN alfa-2b. Similarly using ActiTest equivalence, more patients receiving PEG-IFN alfa-2b showed improvement in activity grade compared with the observation group. Both FibroTest and ActiTest were significantly improved during maintenance therapy of patients with baseline F2-F3 METAVIR fibrosis.

FibroMax utility in the general practice
A multicenter national study named « VARES » was entirely conducted in a family medicine setting within the Italian College of General Practitioners (GP). A total of 259 subjects were included by GPs, aged between 18 and 65 years old, with ultrasound and/or clinical features of non-alcoholic fatty liver disease (NAFLD) with a control group of 23 lean matched healthy subjects. According to FibroMax 70% had moderate to severe steatosis as per SteatoTest, 13% had severe fibrosis F3-F4 as per FibroTest and 6.6% inflammation as per NashTest. FibroTest significantly discriminate NAFLD patients with severe fibrosis F3-F4 from controls (no or minimal fibrosis). Multivariate analysis identified age older than 50 years, diabetes, elevated transaminases and waist circumference or obesity as highly significant independent factors for fibrosis. In severe fibrosis F3-F4 as per FibroTest patients, liver biopsy confirmed fibrosis. In conclusion, FibroMax emerges as a promising tool to evaluate liver disease staging with a key role of GPs in this respect.
### Key publications for 2013

#### Screening for liver fibrosis by using FibroScan and FibroTest in patients with diabetes.

**De Lédinghen, 2012**


The Bordeaux team has made a non-invasive detection of severe liver fibrosis using FibroTest and liver stiffness measurement (LSM) in 277 diabetic patients hospitalized. This study showed a high prevalence of severe fibrosis in patients hospitalized with diabetes, especially in patients aged 50 years or older with type 2 diabetes or with a history of diabetic foot.

#### Noninvasive assessment of hepatic fibrosis in Egyptian patients with chronic hepatitis C virus infection.

**Fouad, 2012**

*World J Gastroenterol.* 2012 Jun 21

This study evaluated the SteatoTest from the FibroMax panel of blood tests for the noninvasive diagnosis of steatosis in 44 patients with chronic HCV and liver biopsy. There was a significant positive correlation between the percentage of steatosis evaluated by SteatoTest and by liver biopsy ($r = 0.952, P = 0.0001$). The authors stressed the limits of ultrasound evaluation of steatosis as almost one third of patients with histological steatosis had no steatosis on ultrasound and almost half of patients without ultrasound steatosis had mild to moderate steatosis on biopsy. In conclusion, the authors recommend FibroMax for steatosis assessment along fibrosis and activity (by SteatoTest, FibroTest and ActiTest, respectively) in patients with HCV and steatosis.

#### Screening for liver fibrosis in a group of Mexican children. A multicenter study.

**Flores-Calderón, 2012**


68 children of mean age 10 years (1-17 years) with chronic liver disease, mainly steatosis, autoimmune hepatitis, biliary atresia and metabolic diseases were included with FibroTest, APRI and biopsy. The diagnostic value (AUROC) of FibroTest for advanced fibrosis (F3F4) was 0.90. Based on these results the authors propose the FibroTest to select patients for biopsy, fibrosis screening before clinical and biological signs and follow patients longitudinally.

#### Comparison of nine blood tests and transient elastography for liver fibrosis in chronic hepatitis C: the ANRS HCEP-23 study.

**Zarski, 2012**

*J Hepatol.* 2012 Jan

This ANRS HCEP-23 study compared the diagnostic accuracy of nine blood tests including FibroTest and transient elastography to assess liver fibrosis vs. liver biopsy, in untreated patients with chronic hepatitis C (CHC) in 19 French University hospitals. Main advantages of the study compared to previous validations were that the biopsy was reviewed by two experienced pathologists and performances were assessed using ROC curves corrected by Obuchowski's method in order to avoid spectrum bias effect. 22% patients had not applicable (failure or unreliable) elastography by Fibroscan results. Among nine noninvasive markers FibroTest AUROC was 0.84 being as accurate as the transient applicable elastography (AUROC 0.84) and two other noninvasive patented methods and significantly better than APRI, Fib-4 and Hyaluronate.

#### Validation of liver fibrosis biomarker (FibroTest) for assessing liver fibrosis progression: Proof of concept and first application in a large population.

**Poynard, 2012**


September 2012, the Journal of Hepatology Editor pick: 342.346 FibroTests in the world to study the factors of fibrosis progression to cirrhosis. The fastest growth is associated with alcoholic liver disease in men - a 10-year advanced risk compared to women - the slower for women with non-alcoholic fatty liver disease. Other factors associated with progression to cirrhosis are co-infection with HIV-HCV and residency in the Middle East or Eastern Europe.

#### Screening for liver fibrosis by using FibroScan and FibroTest in patients with diabetes.

**Poynard, 2012**

*Clin Gastroenterol Hepatol.* 2012;10:657-63.e7

We examined whether the performance of non-invasive tests such as FibroTest for the diagnosis of intermediate stages of fibrosis (F2 vs F1) results from the biopsy limits even for F2 vs F1. We analyzed 27,869 digitized images of virtual biopsies of different sizes (5 to 30mm) from surgical explants. The performance of the biopsy was worse for diagnosing F2 vs. F1 than for F1 vs F0 or F4 vs F3, even for samples of 30-mm. Contrary to what is often said, the recommendation to perform biopsy instead of a noninvasive marker for the diagnosis of intermediate stages of fibrosis is not scientifically validated.
Non-invasive tests for fibrosis and liver stiffness predict 5-year survival of patients chronically infected with hepatitis B virus

V. de Lédinghen*,†, J. Vergniol*, C. Barthe*, J. Foucher*‡, F. Chermak*, B. Le Bail†§, W. Merrouche* & P.-H. Bernard‡

*Centre d’Investigation de la Fibrose hépatique, Hôpital Haut-Lévêque, CHU Bordeaux, Pessac, France.
†INSERM U1053, Université Bordeaux Segalen, Bordeaux, France.
‡Service d’Hépato-Gastroentérologie, Hôpital Saint-André, CHU Bordeaux, Bordeaux, France.
§Service d’Anatomie-Pathologique, Hôpital Pellegrin, CHU Bordeaux, Bordeaux, France.

Correspondence to:
Dr V. de Lédinghen, Service d’Hépato-Gastroentérologie, Centre d’Investigation de la Fibrose hépatique, Hôpital Haut-Lévêque, 33604 Pessac, France.
E-mail: victor.deledinghen@chu-bordeaux.fr

Publication data
Submitted 7 December 2012
First decision 3 January 2013
Resubmitted 13 March 2013
Accepted 19 March 2013

SUMMARY

Background
Liver stiffness and non-invasive tests predict overall survival in chronic hepatitis C. However, in patients chronically infected with hepatitis B virus (HBV), only the association between liver stiffness and the risk of hepatocellular carcinoma has been published.

Aim
To evaluate the 5-year prognostic value of liver stiffness, non-invasive tests of liver fibrosis, and liver biopsy, to predict overall survival in chronic hepatitis B.

Methods
In a consecutive cohort, we prospectively assessed fibrosis, with liver stiffness, FibroTest, APRI, FIB-4 and liver biopsy (if indicated). We examined death and liver transplantation during a 5-year follow-up, and factors associated with overall survival.

Results
A total of 600 patients (men 64%, mean age 42 years, inactive carriers 36%) with chronic hepatitis B were included. At 5 years, 25 patients were dead (13 liver-related deaths) and four patients had liver transplantation. Overall survival was 94.1% and survival without liver-related death 96.3%. No liver-related death was observed in inactive carriers. Survival was significantly decreased in patients diagnosed with severe fibrosis, whatever the non-invasive method used (P < 0.0001), or liver biopsy (P = 0.02). Patients’ prognosis decreased as liver stiffness and FibroTest increased. In multivariate analysis, FibroTest and liver stiffness had the highest hazard ratio with survival. The association persisted after adjustment on age, necro-inflammatory histological activity presumed by ActiTest and treatment.

Conclusions
Liver stiffness measurement or FibroTest can predict survival in chronic HBV infection. Thus, these tools may help physicians to early assess prognosis and discuss specific treatments, such as liver transplantation.

Aliment Pharmacol Ther
Liver fibrosis evaluation using real-time shear wave elastography: Applicability and diagnostic performance using methods without a gold standard

Thierry Poynard¹,*, Mona Munteanu², Elena Luckina¹,³, Hugo Perazzo¹, Yen Ngo¹, Luca Royer¹, Larysa Fedchuk¹, Florence Sattonnet¹, Raluca Pais¹, Pascal Lebray¹, Marika Rudler¹, Dominique Thabut¹, Vlad Ratziu¹

¹Hepato-Gastroenterology, APHP UPMC Liver Center, Paris, France; ²Research Unit, BioPredictive, Paris, France; ³ANRS, Paris, France

Background & Aims: Real-time shear wave elastography (SWE) is a new two-dimensional transient elastography which had no assessment of factors associated with reliability, and had limited comparisons with other validated fibrosis biomarkers. The aim was to assess the applicability and performances of SWE for the diagnosis of fibrosis as compared with FibroTest (FT) and liver stiffness measurement (LSM) by transient elastography using two probes (TE-M and TE-XL).

Methods: Without a gold standard, the strength of concordance, discordance analysis and latent class analysis (LCM) were applied.

Results: 422 patients were included. The applicability of SWE (90.0%) was significantly lower than that of FT (97.9%; \(p<0.0001\)) and did not differ from those of TE-M (90.5%) and TE-XL (90.3%); it was higher though for SWE (86%) in 22 patients vs. 55% using TE-M (\(p=0.04\)). For the diagnosis of all fibrosis stages as presumed by FT, the performance of SWE was highly significant (Obuchowski measure \(0.807 \pm 0.013\) [in ± se]), but lower than those of TE-M (0.852; \(p=0.0007\)) and TE-XL (0.834; \(p=0.046\)). SWE had a low performance for discrimination between F0 and F1. For the diagnosis of cirrhosis using LSM, SWE specificities were all equal to 99%, and SWE sensitivities ranged from 0.47 to 0.64. For the diagnosis of non-cirrhotic stages, the results were heterogeneous.

Conclusions: The performance of SWE for the diagnosis of cirrhosis was similar to those of FT and TE. SWE applicability was lower than that of FT, but greater than that of TE in patients with ascites.

© 2013 European Association for the Study of the Liver. Published by Elsevier B.V. All rights reserved.

Keywords: Fibrosis; Obuchowski measure; Concordance; FibroTest; FibroSure; FibroScan; Aixplorer; Non-invasive methods.

Introduction

Liver fibrosis evaluation using real-time shear wave elastography (SWE) by Aixplorer™ is a new two-dimensional transient elastography technique [1], which has been used in few studies of liver disease, and only in patients with chronic hepatitis C [2,3].

Like transient elastography (TE) and acoustic radiation force imaging (ARFI), SWE evaluates the speed of a shear wave to provide a quantitative estimate of tissue stiffness. SWE has the advantage over TE of being able to image liver stiffness in real time because the shear waves are generated by ultrasound pushes. The SWE image, not limited to a single location, is also guided by a higher frame-rate B-mode image than TE and ARFI. SWE is providing a real-time quantitative map of liver tissue stiffness [1–3].

The applicability (failure rate and non-reliable rate) from these limited published studies of SWE in liver disease is unknown. Furthermore, no study has compared the applicability and performance of SWE for the diagnosis of fibrosis with the two most validated non-invasive biomarkers, the in vitro multivariate assay FibroTest (FT) and liver stiffness measurement based on TE using Fibroscan™ [4–6].

The first aim of this study was to estimate and compare the applicability of SWE with that of FT and the two Fibroscan probes (TE-M and TE-XL) in consecutive patients with chronic liver disease not restricted to chronic hepatitis C. We previously demonstrated that the applicability of fibrosis estimates directly impacts the performance of tests in an intention-to-diagnose analysis [7].

The second aim of this study was to compare the diagnostic performance of SWE to those of FT and the two Fibroscan probes. Liver biopsy is usually used as the reference when performing these comparisons, but this methodology has major limitations. Even a biopsy specimen 25 mm in length has more than 20% false positive or false negative results for fibrosis staging vs. large surgical biopsies [8], and there is a significant gray zone for intermediate stages [9]. Therefore classical estimates of diagnostic test accuracy (sensitivity, specificity, area under the ROC curves [AUROC] and predictive values) are false or very limited [10]. The magnitude of the impact of this error of biopsy is so great that AUROC determinations >0.90 for the diagnosis of advanced
Improved inflammatory activity with peginterferon alfa-2b maintenance therapy in non-cirrhotic prior non-responders: A randomized study

Thierry Poynard1,*, Jordi Bruix2, Eugene R. Schiff3, Moises Diago4, Thomas Berg5, Ricardo Moreno-Otero6, Andre C. Lyra7, Flair Carrilho8, Louis H. Griffel9,⇑, Navdeep Boparai9,⇑, Ruiyun Jiang9, Margaret Burroughs9, Clifford A. Brass9,⇑, Janice K. Albrecht9,⇑

1Service d’Hépato-Gastroenterologie, APHP-UPMC Paris Liver Center, Paris, France; 2Centro de Investigación Biomédica en Red de Enfermedades Hepáticas y Digestivas, Liver Unit, Hospital Clinic, University of Barcelona, Barcelona, Spain; 3Center for Liver Disease, University of Miami Miller School of Medicine, Miami, FL, USA; 4Hospital General Universitario de Valencia, Valencia, Spain; 5Klinik und Poliklinik für Gastroenterologie & Rheumatologie, Sektion Hepatologie, Universitätsklinikum Leipzig, Leipzig, Germany; 6Hospital Universitario de la Princesa (IIS-IP) and Centro de Investigación Biomédica en Red de Enfermedades Hepáticas y Digestivas (Instituto de Salud Carlos III), Madrid, Spain; 7Hospital Sao Rafael and Federal University of Bahia, Salvador, Brazil; 8Gastroenterology, University of Sao Paulo School of Medicine, Sao Paulo, Brazil; 9Schering-Plough Corporation, now Merck Sharp & Dohme Corp., a Subsidiary of Merck & Co., Inc., Whitehouse Station, NJ, USA

Background & Aims: Therapeutic options for patients failing hepatitis C retreatment are limited. EPIC1 included a prospective trial assessing long-term peginterferon alfa-2b (PegIFNα-2b) maintenance therapy in patients with METAVIR fibrosis scores (MFS) of F2 or F3 who previously failed hepatitis C retreatment. Methods: Patients with F2/F3 MFS who failed retreatment were randomized to PegIFNα-2b (0.5 μg/kg/week, n = 270) or observation (n = 270) for 36 months. Blinded liver biopsies obtained before retreatment and after maintenance therapy were evaluated using MFS and activity scores, and confirmatory testing was performed using FibroTest and ActiTest. Results: In total, 348 patients had paired biopsies: 192 patients receiving PegIFNα-2b and 11% of observation patients had improvement in MFS (p = 0.32). More PegIFNα-2b than observation patients had improvement in activity score (20% vs. 9%; p <0.001). Among patients treated for >2.5 years, improvement in MFS or activity score was more common with PegIFNα-2b than observation (21% vs. 14%, p = 0.08 and 26% vs. 10%, p <0.001). FibroTest and ActiTest evaluations indicated significant benefit associated with PegIFNα-2b in terms of reduced fibrosis progression and improved activity score. The safety profile of PegIFNα-2b was similar to previous studies. Conclusions: PegIFNα-2b did not significantly improve MFS estimated by biopsy compared with observation; however, activity scores were significantly improved and MFS trended toward increased improvement with treatment durations >2.5 years. Both FibroTest and ActiTest were significantly improved during maintenance therapy.

© 2012 European Association for the Study of the Liver. Published by Elsevier B.V. All rights reserved.

Introduction

Advancing liver disease is one of the most widely recognized factors affecting treatment outcomes for patients with chronic hepatitis C (CHC). Treatment of hepatitis C with peginterferon (PegIFN) plus ribavirin is contraindicated in patients with compensated cirrhosis, and rates of sustained virologic response (SVR) are typically low in those with bridging fibrosis or compensated cirrhosis. Thus, many patients with advanced liver disease fail initial treatment and become candidates for retreatment. The Evaluation of Peglntron in Control of Hepatitis C Cirrhosis (EPIC2) study was a large, prospective, multiphase clinical program that evaluated the retreatment of patients with moderate to severe fibrosis/cirrhosis using PegIFNα-2b plus ribavirin [1,2]. In this study, retreatment of patients with CHC infection with PegIFNα-2b plus ribavirin resulted in SVR rates of 21%, 16%, and 10% in genotype 1 patients with METAVIR F2, F3, and F4 disease, respectively [1]. Low rates of SVR among patients who fail repeated courses of PegIFNα plus ribavirin have led to the study of maintenance therapy as an approach to slow the histologic advancement of liver disease and delay the development of end-stage liver disease.
Utility of noninvasive methods for the characterization of nonalcoholic liver steatosis in the family practice. The “VARES” Italian multicenter study

Ignazio Grattagliano,* Enzo Ubaldi,* Luigi Napoli,* Carlo Fedele Marulli,* Cristina Nebiacolombo,* Carmelo Cottone,* Piero Portincasa**

* Italian College of General Practitioners, Florence, Italy.
** Department of Biomedical Sciences and Human Oncology, University of Bari, Italy.

ABSTRACT

The diagnostic utilities of ultrasonography (US), fatty liver index (FLI) and an algorithm of nine serum markers (Fibromax) were evaluated in family practice to noninvasively characterize patients with nonalcoholic fatty liver disease (NAFLD). A multicenter study was conducted by enrolling 259 consecutively observed patients (age 51 ± 10 years) with clinical and ultrasonographic features of NAFLD. Patients had mild (16.2%), moderate (69.9%), or severe (13.9%) liver steatosis and 60.2% had hypertransaminasemia. The percent of patients with overweight, obesity, diabetes, hypertension, and dyslipidemia were 42.7%, 46.5% (4.2% severe obesity), 24.7%, 40.9%, and 56.4%, respectively. Lean patients (10.8%) had normal transaminases in two-thirds of the cases. A multivariate logistic regression (including age > 50 yrs, BMI > 30 kg/m², HOMA > 3, and hypertransaminasemia) identified 12.3% of patients at risk for steatohepatitis. With a sensitivity of 50% and specificity of 94.7%, Fibromax identified 34 patients (13.1%) with likely advanced fibrosis and found that over 28% of patients with moderate (ultrasonographic) steatosis were likely to be carrying severe steatosis. Steatotest score was significantly associated with BMI, waist circumference, ALT, triglycerides, and FLI. Fibrotest correlated only with ALT. FLI identified 73.4% of patients as likely to be carrying a fatty liver.

In conclusion, NAFLD should be systematically searched and characterized in all patients with metabolic disturbances and cardiovascular risk. Asymptomatic subjects at risk also should be screened for NAFLD. Fibromax is a promising noninvasive diagnostic tool in family medicine for identifying patients at risk for NAFLD who require targeted follow-up.

Key words. Fatty liver. Fibromax. General practice. Liver fibrosis. Primary care.

INTRODUCTION

Nonalcoholic fatty liver disease (NAFLD) is a clinico-histopathological entity with histological features resembling alcohol-induced liver injury. NAFLD occurs in patients with negligible or negative history of alcohol consumption. Histologically, the spectrum of NAFLD ranges from fat accumulation in hepatocytes without concomitant inflammation or fibrosis (simple hepatic steatosis) to hepatic steatosis with a necroinflammatory component (steatohepatitis) with or without fibrosis (nonalcoholic steatohepatitis, NASH). NAFLD is an emerging problem in westernized societies with high impact on care utilization and costs and is frequently associated with the metabolic syndrome and cardiovascular diseases. Prevention and early identification of NASH is of key importance since, whereas “simple” steatosis is benign, NASH puts 5-8% of patients at risk of cryptogenic cirrhosis within 5 years.

The ultimate diagnosis of NASH is based on liver biopsy, an invasive procedure not free of complications and poorly accepted by patients. Liver biopsy, moreover, is not currently advisable for the large scale population and carries potential bias including sampling error and intra- and inter-observer discrepancies. Therefore, noninvasive tests are actively being investigated and are useful in family practice to select the subset of patients requiring further consultations.
Liver, Pancreas and Biliary Tract

Screening for liver fibrosis by using FibroScan® and FibroTest in patients with diabetes

Victor de Lédinghen a,b, ∗, Julien Vergniol a, Concepcion Gonzalez c, Juliette Foucher a, Elisa Maury c, Louise Chemineau a, Sandrine Villars a, Henri Gin c, Vincent Rigalleau c

a Service d'Hépato-Gastroentérologie, Hôpital Haut-Lévêque, Pessac, France
b INSERM U1053, Université Bordeaux Segalen, Bordeaux, France
c Service de Nutrition, Hôpital Haut-Lévêque, Pessac, France

ARTICLE INFO

Article history:
Received 27 July 2011
Accepted 10 December 2011
Available online xxx

Keywords:
Cirrhosis
Diabetes
Fibrosis
Liver stiffness measurement
Metabolic syndrome
NAFLD
Non-invasive markers

ABSTRACT

Background: Patients with diabetes are at risk for nonalcoholic fatty liver disease leading to cirrhosis. Existing guidelines do not advocate screening for liver related complications amongst persons with diabetes.

Aim: The aim of this prospective study was to identify patients with severe liver fibrosis amongst patients hospitalized for their diabetes, using non-invasive methods, and to evaluate factors associated with severe fibrosis.

Methods: Consecutive patients with type 1 or 2 diabetes had clinical, biological parameters and liver fibrosis evaluation. Severe fibrosis was predicted when FibroTest was >0.59 or liver stiffness >8.7 kPa.

Results: A total of 277 patients were evaluated (type 1 diabetes 52%). The prevalence of severe fibrosis was 15.5%. By univariate analysis, factors associated with severe fibrosis were age, type 2 diabetes, body mass index, metabolic syndrome, previous cardiovascular events, no retinopathy, past history of foot ulcer, and elevated alanine aminotransferase. By multivariate analysis, factors associated with severe fibrosis were age >50 years, type 2 diabetes, no retinopathy, and past history of foot ulcer.

Conclusion: This study showed an elevated prevalence of severe fibrosis in hospitalized diabetic patients, especially patients aged 50 years or older with type 2 diabetes, or with a past history of foot ulcer.

© 2011 Editrice Gastroenterologica Italiana S.r.l. Published by Elsevier Ltd. All rights reserved.

1. Introduction

Patients with diabetes are at risk for nonalcoholic fatty liver disease (NAFLD). NAFLD is an increasingly recognized disease state in which lipid accumulates in the liver in the absence of excessive alcohol consumption [1]. Although the majority of patients with NAFLD have simple steatosis only, it represents a spectrum of liver disease progressing through non-alcoholic steatohepatitis (NASH) and fibrosis, to cirrhosis and end stage liver failure. NAFLD is now recognized as the most common cause of chronic liver abnormalities in the United States—not unexpected given the current epidemic of obesity in this industrialized nation. Parallel to adult obesity, the prevalence of type 2 diabetes mellitus has risen to an unprecedented level over the past decade [2]. The negative impact of diabetes on the retinal, renal, nervous and cardiovascular systems is well recognized, but little is known about its effect on the liver [3,4]. Existing guidelines do not advocate screening for liver related complications amongst persons with diabetes, making the liver a potentially neglected target organ. Diabetes–associated liver disease has been described largely in uncontrolled patient series [5]. However, those reports did not assess the development of serious liver disease. In one large cohort study based on computerized national databases of the US Department of Veterans Affairs, the incidence of NAFLD was nearly two times higher amongst those with diabetes [6]. However, the study sample consisted almost entirely of older men requiring care in hospital, for which the duration of diabetes was unknown. Recently, Porepa et al. evaluated whether adults with newly diagnosed diabetes were at increased risk of serious liver disease [7]. They found that the incidence rate of serious liver disease was 8.19 per 10,000 person-years amongst those with newly diagnosed diabetes and 4.17 per 10,000 person-years amongst those without diabetes.

Alanine aminotransferase (ALT) level and ultrasonography are not adequate methods for the diagnosis of liver fibrosis, even as screening tests. In the past decade, non-invasive markers have been developed as an alternative for liver biopsy, to evaluate liver fibrosis. The most used and useful test is FibroTest (Biopredictive, Paris, France) [8,9]. Liver stiffness measurement using FibroScan...
Comparative diagnostic study of biomarkers using FibroMax™ and pathology for prediction of liver steatosis in patients with chronic hepatitis C virus infection: an Egyptian study

Ahmad Fouad1,*, Dina Sabry2, Rasha Ahmed1,*, Manal Kamal3,*, Sayed Abd Allah4,*, Samar Marzouk2,*, Mona Amin4,*, Rokaya Abd El Aziz4,*, Ahmad El Badri4,*, Hany Khattab5,*, Dina Helmy5,*.  

1Endemic Medicine Department,  
2Medical Biochemistry and Molecular Biology Department,  
3Clinical and Chemical Pathology Department,  
4Internal Medicine Department,  
5Pathology Department, Faculty of Medicine, Cairo University, Egypt  

*These authors contributed equally to this work

Background: Steatosis is common in patients with hepatitis C virus (HCV) infection and may be a major determinant of progression of liver injury. This study evaluated FibroMax™ for noninvasive diagnosis of steatosis in patients with chronic HCV.

Methods: This cross-sectional study included 44 patients naïve to treatment who were referred to our hepatology clinic for assessment of fitness for antiviral therapy. Chronic HCV infection was diagnosed by viral markers. Investigations included assessment of abdominal ultrasonography, liver biopsy, calculation of body mass index, and biomarker parameters in serum using FibroMax.

Results: Histopathology of liver biopsies showed steatosis in 30 of 44 (68%) patients. FibroMax results were positively correlated with viral load by quantitative polymerase chain reaction and histopathological findings. Body mass index was significantly higher in steatotic patients (P = 0.003) and was significantly associated with the results on FibroMax (P = 0.005).

Conclusion: FibroMax was correlated with histopathology and body mass index in patients with HCV. Abdominal ultrasonography could not be used as a single tool to diagnose steatosis with HCV. Steatosis is correlated with viral load, which suggests a direct viral effect. We recommend FibroMax assessment in a larger number of patients to assess its applicability in patients with HCV and steatosis.

Keywords: steatosis, hepatitis C virus, histopathology, FibroMax™

Introduction

Hepatic steatosis has a high prevalence worldwide, and has been found to be associated with several features, including diabetes, hyperlipidemia, obesity, insulin resistance, and viral hepatitis.1 Egypt has the largest epidemic of hepatitis C virus (HCV) in the world. The overall prevalence of people positive for antibodies to HCV in Egypt has been reported to be 14.7%.2

Steatosis is a frequent feature of HCV infection, and may be an important cofactor in both accelerating fibrosis and increasing liver necroinflammatory activity in chronic HCV infection. Several studies have suggested that steatosis induces resistance to combination treatment with interferon and ribavirin.3

One of the major clinical problems is how to evaluate steatosis in patients with HCV. Liver biopsy is still recommended by the current guidelines for management of the disorder.4 However, numerous studies have strongly suggested that liver biopsy has limitations, such as potential sampling error, the fact that it is invasive, costly,
Non-invasive markers of liver fibrosis in chronic liver disease in a group of Mexican children. A multicenter study

Judith Flores-Calderón,* Segundo Morán-Villota,* Guillermo Ramón-García,* Berenice González-Romano,* María del Carmen Bojórquez-Ramos,** Laura Cerdán-Silva,*** Pablo Hernández-Frias***

* Departamento de Gastroenterología y Anatomía Patológica, UMAE, Hospital de Pediatría, Centro Médico Nacional Siglo XXI, Instituto Mexicano del Seguro Social, Mexico City, Mexico. ** Departamento de Gastroenterología, UMAE, Hospital de Pediatría del Centro Médico Nacional de Occidente, Guadalajara, Jal., Instituto Mexicano del Seguro Social. *** Departamento de Gastroenterología. UMAE Dr. Gaudencio González Garza, Centro Médico Nacional La Raza, Instituto Mexicano del Seguro Social. Mexico City, Mexico.

ABSTRACT

Introduction. Identifying liver fibrosis is important to evaluate the severity of liver damage and to establish a prognosis. Utility of non-invasive markers of liver fibrosis has been proved in adults but there are few reports in children. The aim of this study was to evaluate Fibrotest® score and APRI suitability to identify children with liver fibrosis. Material and methods. 68 children with chronic liver disease requiring liver biopsy were prospectively included from three 3rd-level pediatric hospitals. The same pathologist evaluated all liver biopsies; fibrosis degree was determined by METAVIR score. Serum samples were obtained to determine Fibrotest® and APRI. AUROC were used to determine cut-off and differentiate between advanced fibrosis (METAVIR F3, F4) and no fibrosis (F0). Results. 68 biopsies were evaluated; METAVIR > F3 was identified in 26 (38%). Non invasive liver fibrosis markers to differentiate between advanced and no fibrosis were: Fibrotest® AUROC = 0.90 (95% CI 0.77-1.00) (cut-off value 0.35) sensitivity 88.00% (95% CI 68-96) and specificity 80% (95% CI 29-98); and for APRI AUROC = 0.97 (95% CI 0.92-1.00) (cut-off value 0.82), sensitivity 88% (95% CI 68-96) and specificity 100% (95% CI 46-100). Conclusion. These results suggest the utility of Fibrotest® and APRI to identify advanced fibrosis; they can be recommended to select patients for liver biopsy and during patient follow-up.

Key words. Liver fibrosis. APRI. Fibrotest®. Children.

INTRODUCTION

Chronic liver disease (CLD) in children varies with age of presentation and differences in their natural history; the main causes in newborn and young infant are idiopathic neonatal hepatitis, biliary atresia and metabolic disease. In older children and adolescents, the leading causes are autoimmune hepatitis, cryptogenic cirrhosis, biliary atresia post-Kasai status, primary sclerosing cholangitis, Wilson’s disease, chronic hepatitis B (HBV), hepatitis C (HCV), and non-alcoholic fatty liver disease (NAFLD).1,2 These different etiologic forms of CLD have a common histopathological pathway that is the formation and accumulation of fibrosis leading to the development of progressive distortion of hepatic architecture and cirrhosis. Natural history studies indicate that advanced fibrosis and cirrhosis in children at diagnosis are 69% in those with autoimmune hepatitis (AIH) type I, 38% in AIH type 2, 21% in Wilson disease, 25% in patients with HCV, 3.5% in children with HBV and 3% in cases with NAFLD.3-7 Progression may take years, particularly for those patients with chronic viral hepatitis or NAFLD who are well compensated without clinical or laboratory signs of cirrhosis, therefore, staging of hepatic fibrosis is of clinical importance for diagnostic assessment and to decide the need for immediate...
Comparison of nine blood tests and transient elastography for liver fibrosis in chronic hepatitis C: The ANRS HCEP-23 study

Jean-Pierre Zarski1,2,* Nathalie Sturm2,3, Jérôme Guechot4, Adeline Paris5, Elie-Serge Zafrani6, Tarik Asselah7, Renée-Claude Boisson8, Jean-Luc Bosson5, Dominique Guyader9, Jean-Charles Renversez10, Jean-Pierre Bronowicki11, Marie-Christine Gelineau12, Albert Tran13, Candice Trocmé14, Victor De Ledinghen15, Elisabeth Lasnier4, Armelle Poujol-Robert16, Frédéric Ziegler17, Marc Bourlière18, Hélène Voitot19, Dominique Larrey20, Maria Alessandra Rosenthal-Alieri21, Isabelle Fouchard Hubert22, François Bailly23, Michel Vaubourdolle4, The ANRS HCEP 23 Fibrostar Group

1Clinique Universitaire d’Hépato-Gastroentérologie – Pôle DIGIDUNE-CHU de Grenoble, France; 2Unité INSERM/UJF U823 IAPC Institut Albert Bonniot-Grenoble, France; 3Département d’Anatomie et Cytologie Pathologiques-Pôle de Biologie, CHU de Grenoble, France; 4Laboratoire de Biochimie A – Hôpital Saint Antoine, AP-HP, Paris, France; 5Centre d’Investigation Clinique INSERM CIC003, CHU Grenoble, Grenoble F-38043, France; 6Département de Pathologie, Hôpital Henri Mondor, Assistance Publique-Hôpitaux de Paris, Université Paris Est, Créteil, Val de Marne, France; 7Service d’Hépato-Gastroentérologie, Hôpital Beaujon, AP-HP, Clichy, France; 8Laboratoire de Biochimie, Centre hospitalier Lyon Sud, Hospices Civils de Lyon, France; 9Service d’Hépatologie CHU de Rennes, France; 10Département de Biologie Intégrée, Pôle de Biologie, CHU de Grenoble, France; 11Service d’Hépato-Gastroentérologie, CHU de Nancy, France; 12Laboratoire de Biochimie, Hôtel Dieu, Hospices Civils de Lyon, France; 13Hépatologie, Hôpital l’Arche 2, Nice, France; 14GREPI TIMC-IMAG UMR CNRS 5525, Université de Grenoble/ Laboratoire d’Enzymologie, Dépt. de Biologie et Pathologie de la Cellule, Pôle de Biologie, CHU de Grenoble, France; 15Service d’Hépato-Gastroentérologie, CHU de Bordeaux, France; 16Service d’Hépato-Gastroentérologie, Hôpital St Antoine, AP-HP, Paris, France; 17Laboratoire de Biochimie, Institut de Biologie Clinique, EA4311, IHU, IF23/CHU, Rouen, France; 18Service d’Hépato-Gastroentérologie, Hôpital St Joseph, Marseille, France; 19Laboratoire de Biochimie, Hôpital Beaujon, AP-HP, Clichy, France; 20Service d’Hépato-Gastroentérologie, CHU de Montpellier, France; 21Laboratoire d’Immunologie, CHU de Nice, France; 22Service d’Hépato-Gastroentérologie, CHU d’Angers, France; 23Service d’Hépato-Gastroentérologie, Hotel Dieu-Lyon, France

Keywords: Chronic hepatitis C; Liver fibrosis; Surrogate markers; Transient elastography; Blood tests.

Background & Aims: Blood tests and transient elastography (Fibroscan™) have been developed as alternatives to liver biopsy. This ANRS HCEP-23 study compared the diagnostic accuracy of nine blood tests and transient elastography (Fibroscan™) to assess liver fibrosis, vs. liver biopsy, in untreated patients with chronic hepatitis C (CHC).

Methods: This was a multicentre prospective independent study in 19 French University hospitals of consecutive adult patients having simultaneous liver biopsy, biochemical blood tests (performed in a centralized laboratory) and Fibroscan™. Two experienced pathologists independently reviewed the liver biopsies (mean length = 25 ± 8.4 mm). Performance was assessed using ROC curves corrected by Obuchowski’s method.

Results: Fibroscan™ was not interpretable in 113 (22%) patients. In the 382 patients having both blood tests and interpretable Fibroscan™, Fibroscan™ performed similarly to the best blood tests for the diagnosis of significant fibrosis and cirrhosis. Obuchowski’s measure showed Fibrometer® (0.86), Fibrotest® (0.84), Hepascore® (0.84), and interpretable Fibroscan™ (0.84) to be the most accurate tests. The combination of Fibrotest®, Fibrometer®, or Hepascore® with Fibroscan™ or Apri increases the percentage of well classified patients from 70–73% to 80–83% for significant fibrosis, but for cirrhosis a combination offers no improvement. For the 436 patients having all the blood tests, AUROC’s ranged from 0.82 (Fibrometer®) to 0.75 (Hyaluronate) for significant fibrosis, and from 0.89 (Fibrometer® and Hepascore®) to 0.83 (FIB-4) for cirrhosis.

Conclusions: Contrarily to blood tests, performance of Fibroscan™ was reduced due to uninterpretable results. Fibrotest®, interpretable Fibroscan™, Fibrometer®, and Hepascore® perform best and similarly for diagnosis of significant fibrosis and cirrhosis.

© 2011 European Association for the Study of the Liver. Published by Elsevier B.V. All rights reserved.
Background & Aims: Time-dependent statistics have been used to assess liver fibrosis progression (LFP) in liver diseases from birth to first biopsy, in a limited number of patients. Non-invasive biomarkers such as FibroTest (FT) should allow the estimation of LFP on larger populations. We aimed at validating this concept by comparing LFP using FT vs. biopsy (P1) and then at applying the non-invasive method to a large population (P2).

Methods: In P1, LFP was assessed using biopsy and FT in 2472 untreated patients: 770 with chronic hepatitis C, 723 with hepatitis B, 761 with non-alcoholic fatty liver disease (NAFLD), and 218 with alcoholic fatty liver disease (ALD). In P2, 342,346 interpretable FT prospectively measured were used. LFP was estimated using transition rates (cumulative hazard rate) to cirrhosis (F4) or to minimal fibrosis (>F0).

Results: In P1, there was a significant concordance between FT and biopsy estimates of hazards with intraclass correlation (ICC) = 0.961 (95% CI 0.948–0.970) and 0.899 (95% CI 0.835–0.969) for F4 and >F0, respectively. This concordance persisted according to the disease and the gender. The more rapid LFP to F4 (biopsy/FT) was observed for men with ALD (1.44/1.62), and the slower for women with NAFLD (0.09/0.02).

In P2, the LFP started to increase for men at the age of 30 years. The cumulative fibrosis progression rate to minimal fibrosis in women crossed the “man curve” around the age of 80 years. The following factors were associated with LFP to F4 (all p < 0.0001): male gender (Relative Risk = 3.29), HIV co-infection (2.33), and residency in Middle East (2.67) or Eastern Europe (2.15).

Conclusions: Validated biomarkers such as FibroTest should allow powerful analysis of fibrosis progression in chronic liver diseases and better identification of risk factors.

© 2012 European Association for the Study of the Liver. Published by Elsevier B.V. All rights reserved.

Keywords: FibroTest; FibroSure; Fibrosis progression rates; Risk factors of fibrosis; Biopsy; Normal transaminases; HIV co-infection.

Introduction

Mortality associated with the four common liver diseases, chronic hepatitis B (CHB), chronic hepatitis C (CHC), non-alcoholic (NAFLD), and alcoholic steatosis (ALD), is almost always due to the development of cirrhosis, the final stage of fibrosis progression, and its complications [1,2]. Estimate of liver fibrosis progression (LFP) represents an important surrogate end point that may facilitate treatment decisions by clarifying the vulnerability of an individual patient to progression. The concept of LFP estimation was first developed and applied to patients with CHC [3]. In these patients, this concept has allowed literature observed discordances on the natural history of hepatitis C to be explained, and the epidemic to be reconstructed, and it has become apparent that age at infection and gender greatly influence the risk of developing chronic infection and progressing to fibrosis [3–6]. Even if the impact of age and gender was initially identified with simple modeling [3], LFP in CHC have been mostly assessed with Markov models using transition rates between fibrosis stages estimated using liver biopsy. If the exposure time was known, one biopsy was used to estimate the transition rate between normal liver and first stage (minimal fibrosis); two biopsies were otherwise necessary [6,7]. As liver biopsy has a risk of 3/1000 severe adverse events and 1/10,000 mortality rate, the total number of patients included in studies of LFP was rather limited.

For the other common liver diseases (CHB, NAFLD, and ALD), the exposure time is even more difficult to assess, and repeated biopsies were less often performed, explaining why very few studies of LFP have been published compared to CHC [8].

In order to circumvent the variability due to the exposure time and to use only one biopsy, in 2003, we proposed to use time-dependent statistics to assess and compare LFP from birth to first biopsy in chronic liver diseases [8].

Due to biopsy limitation (sampling error, inter-observers variability, cost, adverse events), several non-invasive fibrosis biomarkers have been validated since 2001 [9]. The diagnostic and prognostic performances of FibroTest have been extensively validated in CHC, CHB, ALD [10–12], and, more recently, NAFLD [13]. The aim of the present study was to demonstrate that validated non-invasive biomarkers such as FibroTest can be used as an alternative to liver biopsy for LFP assessment.
Liver Biopsy Analysis Has a Low Level of Performance for Diagnosis of Intermediate Stages of Fibrosis

**BACKGROUND & AIMS:** There is controversy about the performance of noninvasive tests such as FibroTest in diagnosing intermediate stages of fibrosis. We investigated whether this controversy results from limitations of biopsy analysis for intermediate-stage fibrosis and inappropriate determination of the standard area under the receiver-operator characteristic curve (AUROC).

**METHODS:** To determine whether biopsy has a lower diagnostic performance for fibrosis stage F2 (few septa) vs F1 (fibrosis without septa), compared with its performance for F1 vs F0 or F4 vs F3, we determined the fibrotic areas of large surgical samples collected from 20 consecutive patients with chronic liver disease or normal liver tissue that surrounded tumors. We analyzed digitized images of 27,869 virtual biopsies of increasing length and also analyzed data from 6,500 patients with interpretable FibroTest results who also underwent biopsy analysis.

**RESULTS:** The overall performance of biopsy analysis (by Obuchowski measure) increased with biopsy length from 0.885 for 5-mm to 0.912 for 30-mm samples (P < .0001). The performance of biopsy was lower for the diagnosis of F2 vs F1 samples (weighted AUROC [wAUROC] = 0.505) than for F1 vs F0 (wAUROC = 0.773; 53% difference; P < .0001) or F4 vs F3 (wAUROC = 0.700; 39% difference; P < .0001), even when 30-mm biopsy samples were used. The performance of FibroTest was also lower for the diagnosis of F2 vs F1 samples (wAUROC = 0.512) than for F1 vs F0 samples (wAUROC = 0.626; 22% difference; P < .0001) or F4 vs F3 (wAUROC = 0.628; 23% difference; P < .0001). However, the FibroTest had smaller percentage differences among wAUROC values than biopsy.

**CONCLUSIONS:** Biopsy has a low level of diagnostic performance for fibrosis stages F2 and F1. The recommendation for biopsy analysis, instead of a validated biomarker panel such as FibroTest, for the diagnosis of intermediate stages of fibrosis is therefore misleading.

**Keywords:**

---

**Abbreviations used in this paper:** AF, area of fibrosis; ALD, alcoholic liver disease; AUROC, area under the receiver-operator characteristic curve; CHB, chronic hepatitis B chronic hepatitis C; HBV, hepatitis B virus; HCV, hepatitis C virus; NAFLD, nonalcoholic fatty liver disease; sauroC, standard area under the receiver-operator characteristic curve; wAUROC, weighted area under the receiver-operator characteristic curve.

© 2012 by the AGA Institute
1542-3565/36.00
doi:10.1016/j.cgh.2012.01.023