European Young Hepatologists Workshop

IC-HEP
International Coalition of Hepatology Education Providers

This IC-HEP activity is supported by an educational grant from Gilead Sciences, Inc.

WORKSHOP 1: Non-invasive methods for the management of cirrhosis

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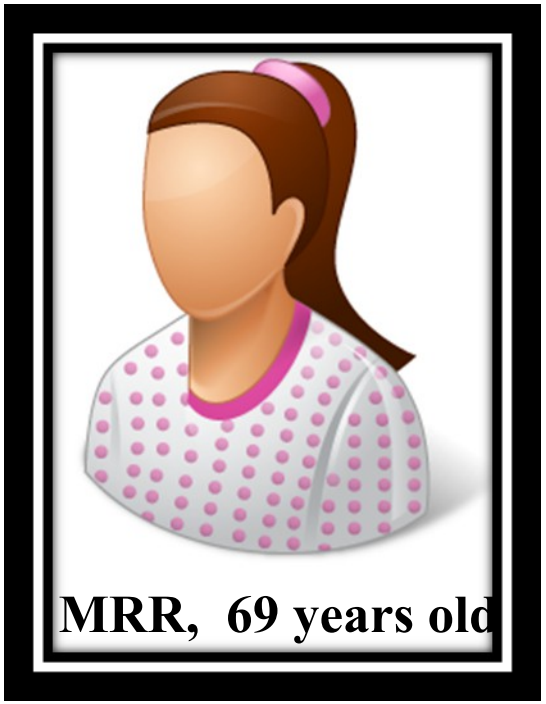
Moulin de Vernègues, France

CLINICAL SETTING



- Mother deceased because of liver cancer
- Type II Diabetes, obesity, osteoporosis

- Asymptomatic
- AST 47 ALT 98 ALP 56 Bi 1.7 mg/dL

- HBV positive: HBsAg+; HBcAb+; HBeAg-
- DNA 2657 IU/mL

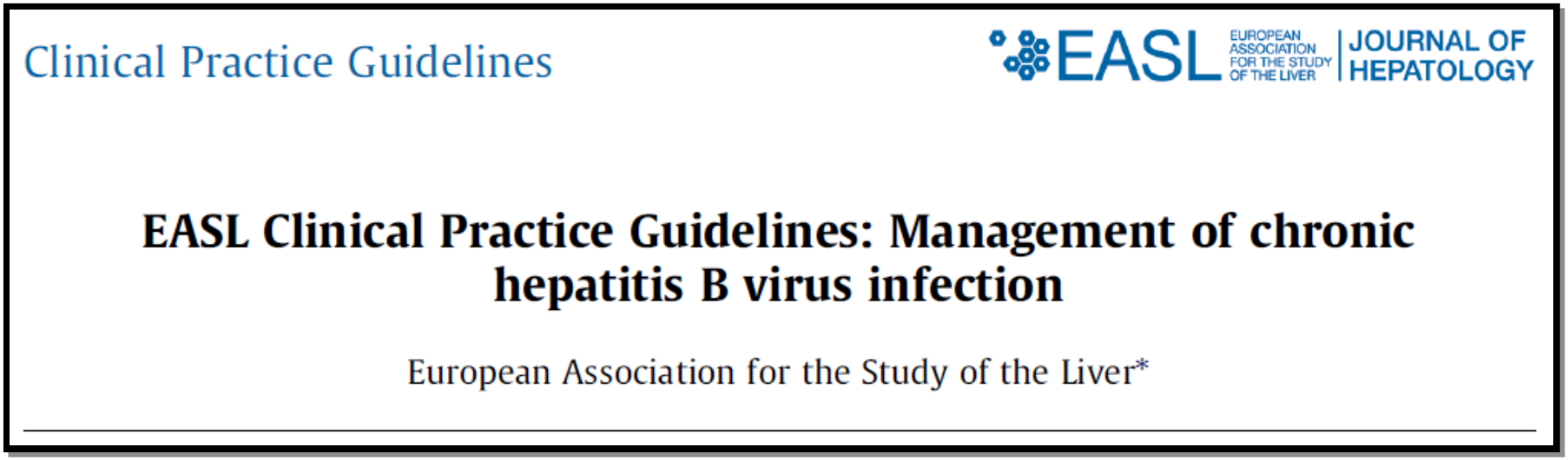
- Abd. Ultrasound: normal sized liver with surface nodularity. No signs of PH.

QUESTION 1

¿What is the clinical relevance of liver fibrosis in this patient?

1) Fibrosis stage impacts on prognosis but does not influence therapeutic decisions.
2) Fibrosis stage provides indication for antiviral therapy but does not impact on prognosis.
3) Fibrosis stage impacts on prognosis and guide therapeutic decisions.
4) Fibrosis stage is irrelevant, and no further evaluation is needed.

EVALUATION OF FIBROSIS IN HBV

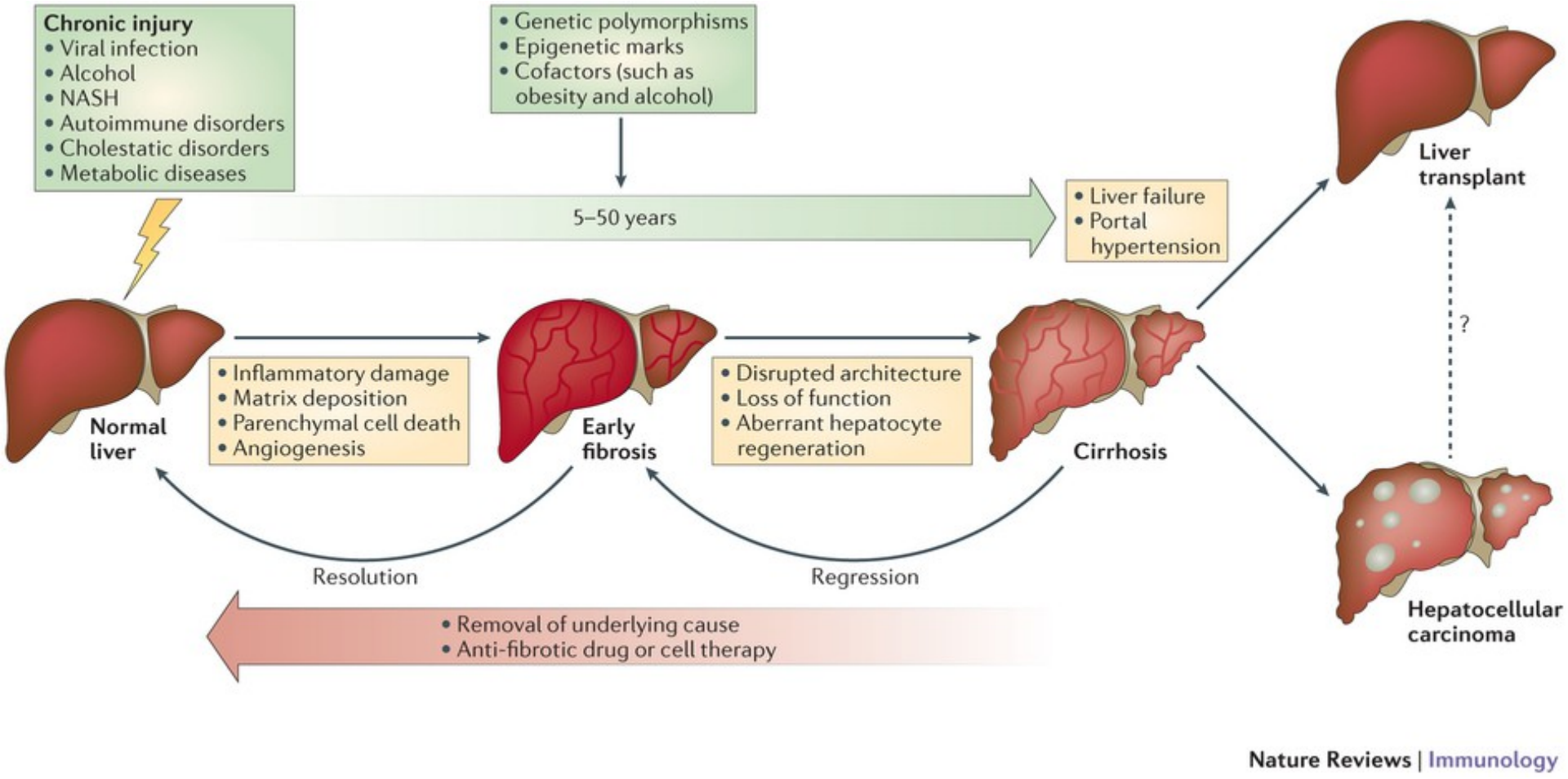


Patients should be considered for treatment when they have HBV DNA >2000 IU/mL, elevated ALT and **at least moderate fibrosis** assessed by liver biopsy (or validated non-invasive markers)

Grade of recommendation A1

EASL clinical practice guidelines: Management of chronic hepatitis B virus infection. *J Hepatol* (2012)

EVALUATION OF FIBROSIS IN HBV



Pellicoro et al. *Nat Rev Immunol* (2014)

QUESTION 2

In our patient, ¿Which method would you advise for the assessment of liver fibrosis?

1) Liver biopsy (METAVIR system).
2) Fibrotest®.
3) Transient elastography (Fibroscan®).
4) Enhanced Liver Fibrosis Test (ELF).

CONSIDERATIONS IN FIBROSIS EVALUATION

- Liver biopsy is an imperfect gold standard.
- Many serum and imaging techniques are available to evaluate liver fibrosis.
- A valid non-invasive liver fibrosis test should be accurate, reproducible, easy to perform and cost-effective.
- Etiology-specific validation is needed before implementation in clinical practice.

LIVER BIOPSY: COMPLICATIONS

TRANSJUGULAR LIVER BIOPSY

62 series, n=7469

Complications	No. of complications	(%) of biopsies
Total	529	(7.1)
Major	42	(0.6)
Large hepatic hematoma	4	(0.05)
Intraperitoneal haemorrhage	15	(0.2)
Inferior vena cava perforation	1	(0.01)
Renal vein perforation	1	(0.01)
Ventricular arrythmia	4	(0.05)
Pneumothorax	4	(0.05)
Respiratory arrest	1	(0.01)
Not specified	12	(0.2)
Deaths	8	(0.1)
Intraperitoneal haemorrhage	5	(0.06)
Ventricular arrythmia	3	(0.04)

Kalambokis et al. *J Hepatol* (2007)

SERUM LIVER FIBROSIS TESTS

Biomarkers	Etiologies	Year	Patients (n)	F≥2 (%)	F4 (%)	Cut-offs	AUROC	Se (%)	Sp (%)	CC (%)
FibroTest® [21]	HCV	2001	339	80		>0.48	0.87	75	85	46
Forns Index [22]	HCV	2002	476	26		<4.2 >6.9	0.81	30-94	51-95	45
APRI [23]	HCV	2003	270	50		≤0.5 >1.5	0.80	41-91	47-95	44
					17	<1.0 ≥2.0	0.89	57-89	75-93	72
FibroSpectII® [24]	HCV	2004	696	52		>0.36	0.83	77	73	75
MP3 [25]	HCV	2004	194	45		<0.3 >0.4	0.82	35-65	85-96	n.a.
FPI [26]	HCV	2005	302	48		≤0.2 ≥0.8	0.77	42-85	48-98	40-49
Hepascore® [27]	HCV	2005	211	57		≥0.5	0.82	63	89	92
					16	>0.84	0.89	71	89	n.a.
Lok index [28]	HCV	2005	1141		38	<0.2 ≥0.5	0.81	40-98	53-99	52
GUCI [29]	HCV	2005	179		12	>0.1	0.85	80	70	n.a.
ViraHep-C [30]	HCV	2006	398	37		≤0.22 >0.55	0.83	51-90	54-90	52
Fibroindex [31]	HCV	2007	360	50		≤1.25 ≥2.25	0.83	30-40	97-97	35
FIB-4 [32]	HCV	2007	830		17*	<1.45 >3.25	0.85	38-74	81-98	68
HALT-C model [33]	HCV	2008	512		38	<0.2 ≥0.5	0.81	47-88	45-92	48
Hui Score [36]	HBV	2005	235	25		≤0.15 >0.5	0.79	37-88	50-88	49
Zeng score [37]	HBV	2005	372	58		<3.0 >8.7	0.77	40-98	28-90	35
SHASTA [38]	HIV-HCV	2005	95	27		<0.3 >0.8	0.87	15-88	72-100	42
FIB-4 [39]	HIV-HCV	2006	832		22*	<1.45 >3.25	0.76	70	97	62
ELF® [34]	Mixed	2004	1021/496**	40		0.102	0.78	87	51	n.a.
		2005			12	n.a.	0.89	n.a.	n.a.	n.a.
Fibrometer® [35]	Mixed	2007	598/503**	56		n.a.	0.89	80	84	82
NFS [40]	NAFLD	2008	733		27*	<-1.455 >0.676	0.82	43-77	97-97	68
BARD score [41]	NAFLD		669		30*	≥2	0.81	n.a.	n.a.	n.a.

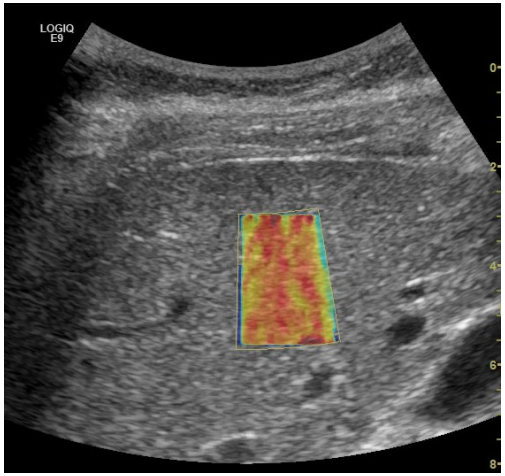
EASL-ALEH clinical practice guidelines: Non invasive tests for evaluation of liver disease
J Hepatol (2015)

IMAGING-BASED FIBROSIS TESTS

FibroScan®

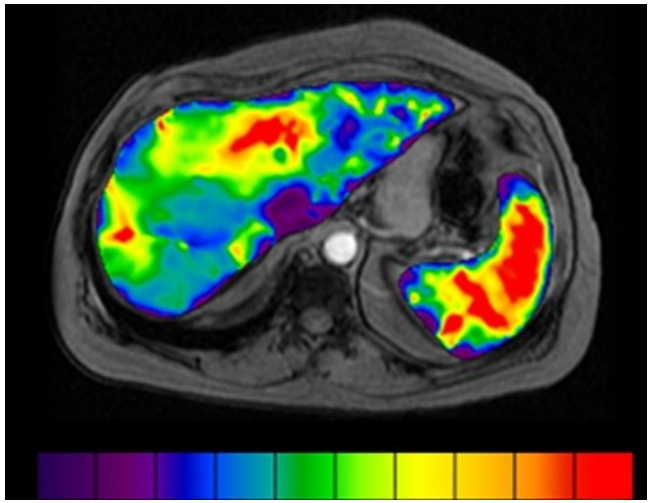
pSWE/ARFI

SIEMENS

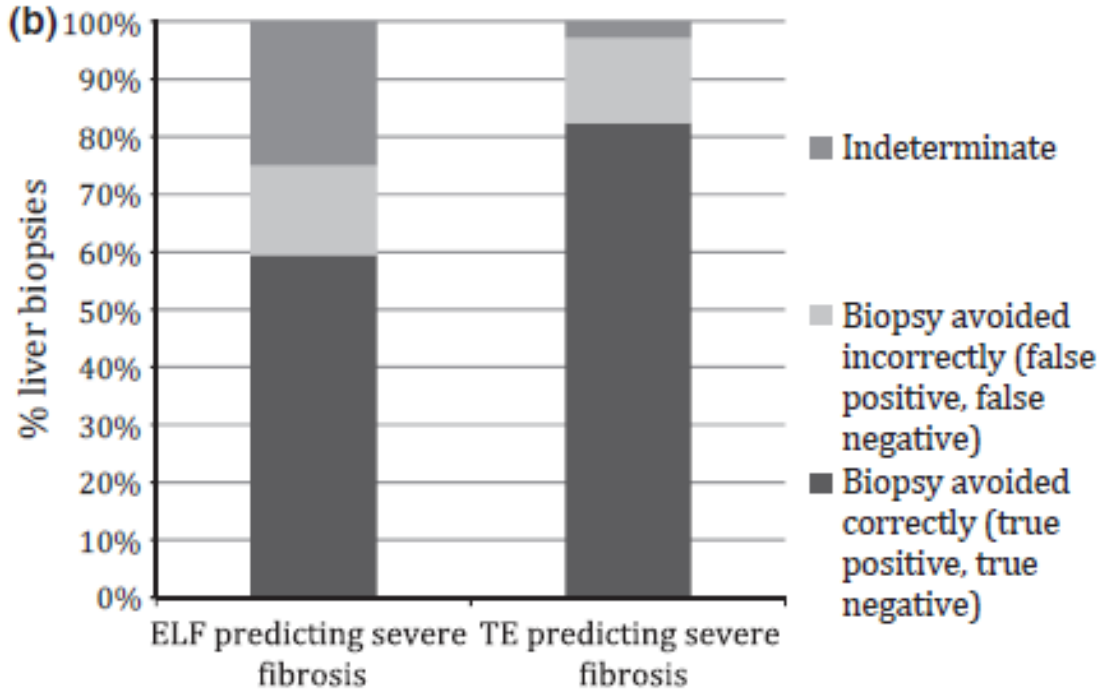


2D-SWE

MRE
Magnetic Resonance Elastography



TRANSIENT ELASTOGRAPHY VS ELF



	ELF score (n = 182)	TE (kPa) (n = 182)	
Fibrosis stage	Adjusted AUROC	Adjusted AUROC	P value*
0–2 *vs* 3,4	0.83	0.94	<0.01
0–3 *vs* 4	0.86	0.96	<0.01

Trembling et al. *J Viral Hepat* (2014)

RECOMMENDATIONS IN HEPATITIS B

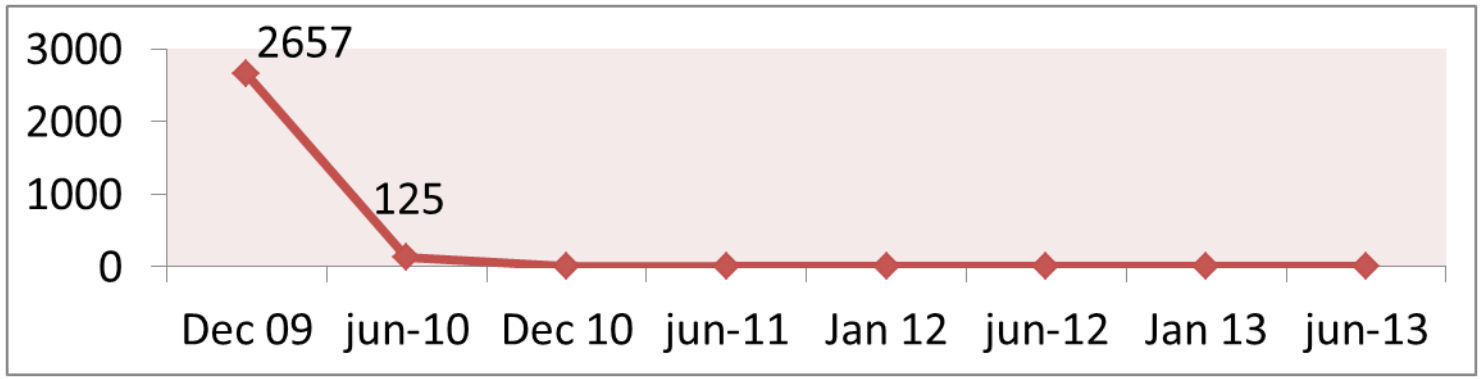
- TE can be considered the non-invasive standard for the measurement of LS (**A1**)
- Although alternative techniques, such as pSWE/ARFI or 2D-SWE seem to overcome limitations of TE, their quality criteria for correct interpretation are not yet well defined (**A1**)
- MR elastography is currently too costly and time-consuming for routine clinical practice use and seems more suited for research purposes (**A1**)
- TE has better prediction for advanced liver fibrosis and cirrhosis than serum biomarkers in chronic hepatitis B (**B1**)
- TE is best used to determine liver fibrosis in hepatitis B patients with active viraemia (HBV DNA >2000 IU/ml) but normal ALT (**A1**)

EASL-ALEH clinical practice guidelines: Non invasive tests for evaluation of liver disease
J Hepatol (2015)

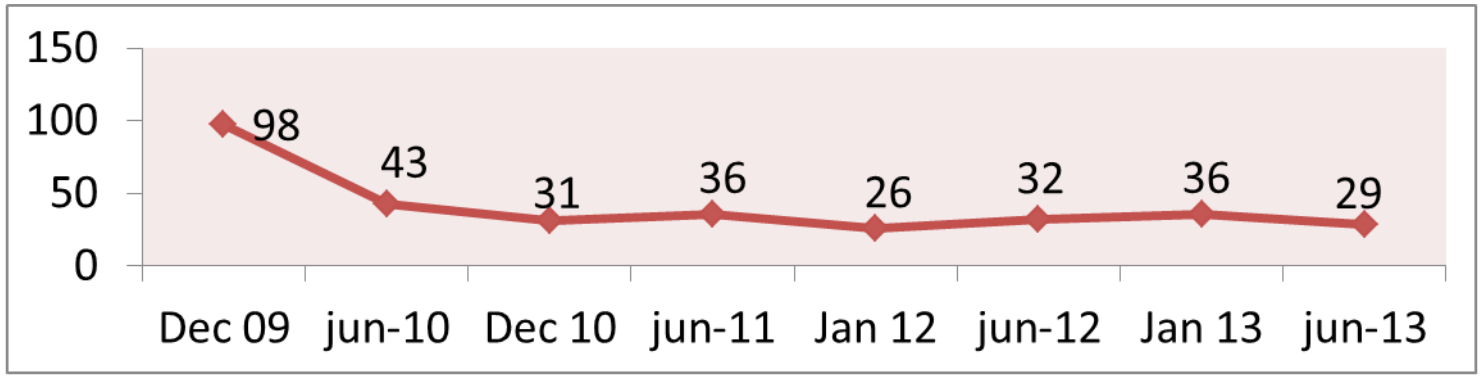
TREATMENT AND SURVEILLANCE

- Fibroscan showed liver stiffness of 18 Kpa (F4)
- The patient started Tenofovir 245 mg once daily
- Clinical and ultrasound surveillance was recommended every 6 months.

DNA (IU/mL)



ALT (IU)



QUESTION 3

Within F4 stage, ¿Which one of the following options indicates poor prognosis?

1) Liver stiffness assessed by transient elastography.
2) Spleen stiffness assessed by transient elastography.
3) Collagen proportional area.
4) Virological response (ie. undetectable HBV DNA at 24 weeks during treatment).
5) All options are correct.

COLLAGEN PROPORTIONAL AREA

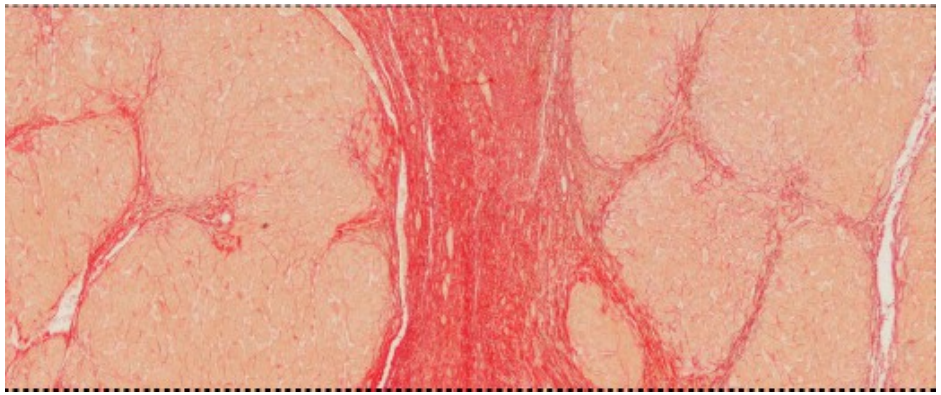
Research Article

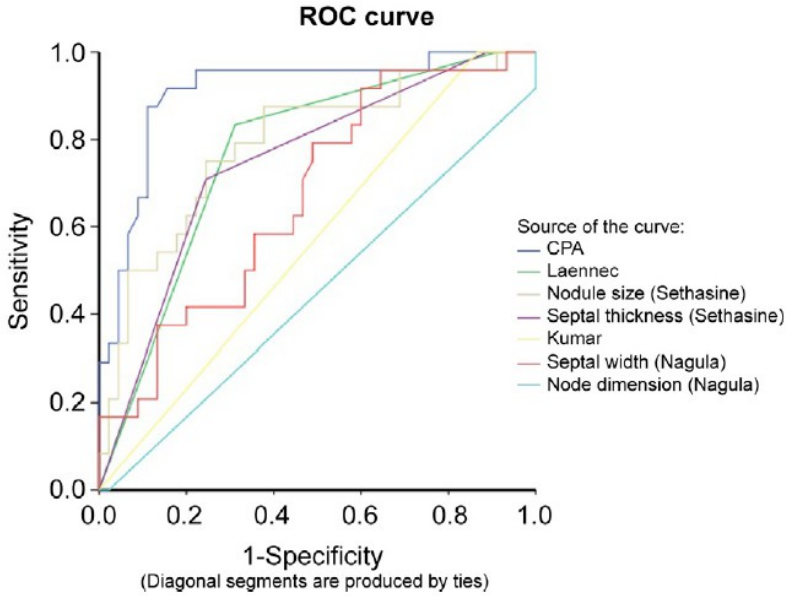
EASL EUROPEAN ASSOCIATION FOR THE STUDY OF THE LIVER | JOURNAL OF HEPATOLOGY

Collagen proportionate area is superior to other histological methods for sub-classifying cirrhosis and determining prognosis

Emmanuel Tsochatzis[1], Sara Bruno[2], Graziella Isgro[1], Andrew Hall[2], Eleni Theocharidou[1], Pinelopi Manousou[2], Amar P. Dhillon[2], Andrew K. Burroughs[1,*,†], Tu Vinh Luong[2,*,†]

[1]*The Royal Free Sheila Sherlock Liver Centre, Royal Free Hospital and UCL Institute for Liver and Digestive Health, London, UK;* [2]*Department of Histopathology, UCL Medical School, Royal Free Campus, UK*





Tsochatzis et al. *J Hepatol* (2014)

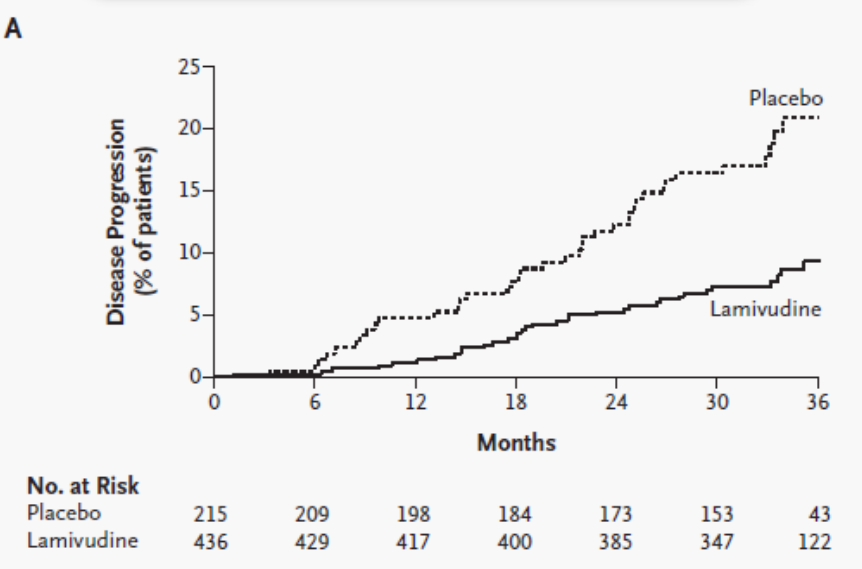
VIROLOGICAL RESPONSE

The NEW ENGLAND JOURNAL of MEDICINE

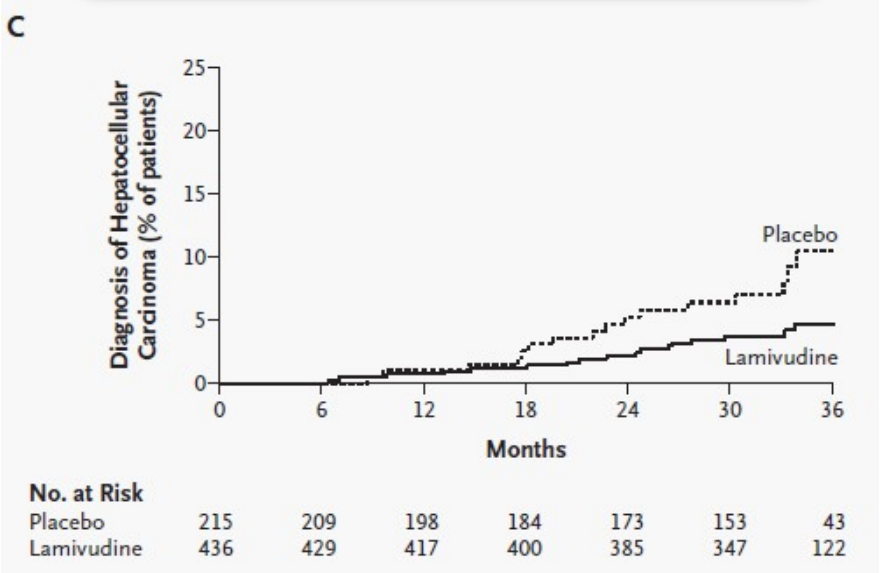
ORIGINAL ARTICLE

Lamivudine for Patients with Chronic Hepatitis B and Advanced Liver Disease

HEPATIC DECOMPENSATION

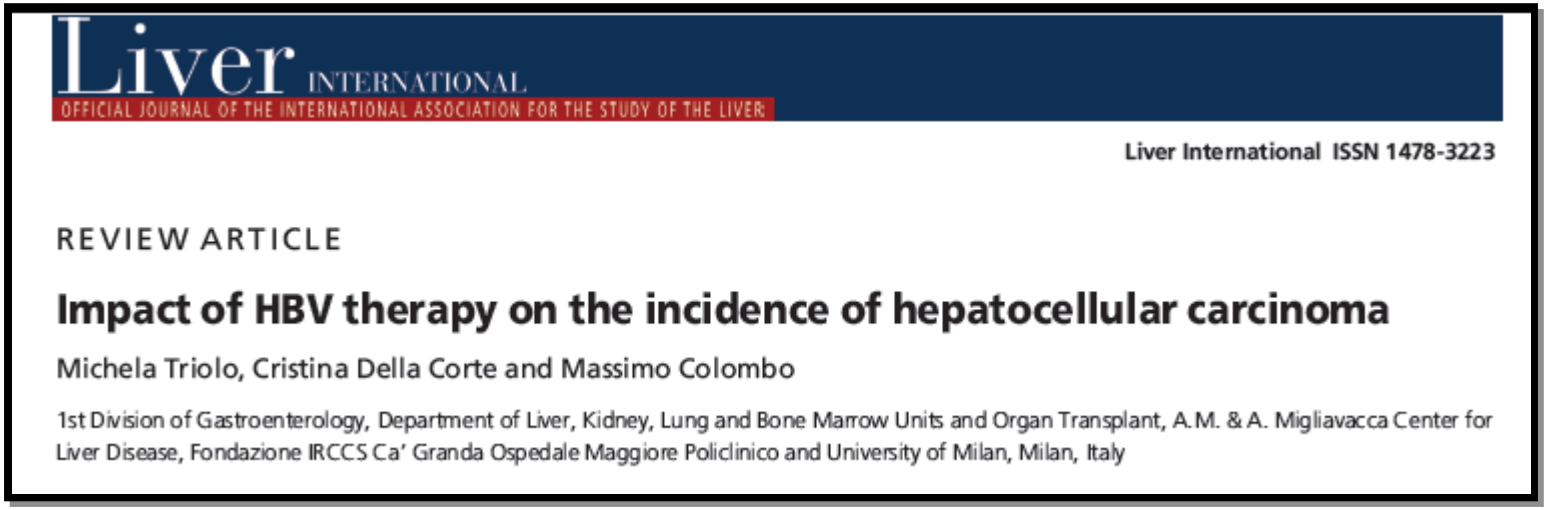


HEPATOCELLULAR CARCINOMA



Liaw et al. *NEJM* (2004)

VIROLOGICAL RESPONSE: HCC



Liver INTERNATIONAL
OFFICIAL JOURNAL OF THE INTERNATIONAL ASSOCIATION FOR THE STUDY OF THE LIVER

Liver International ISSN 1478-3223

REVIEW ARTICLE

Impact of HBV therapy on the incidence of hepatocellular carcinoma

Michela Triolo, Cristina Della Corte and Massimo Colombo

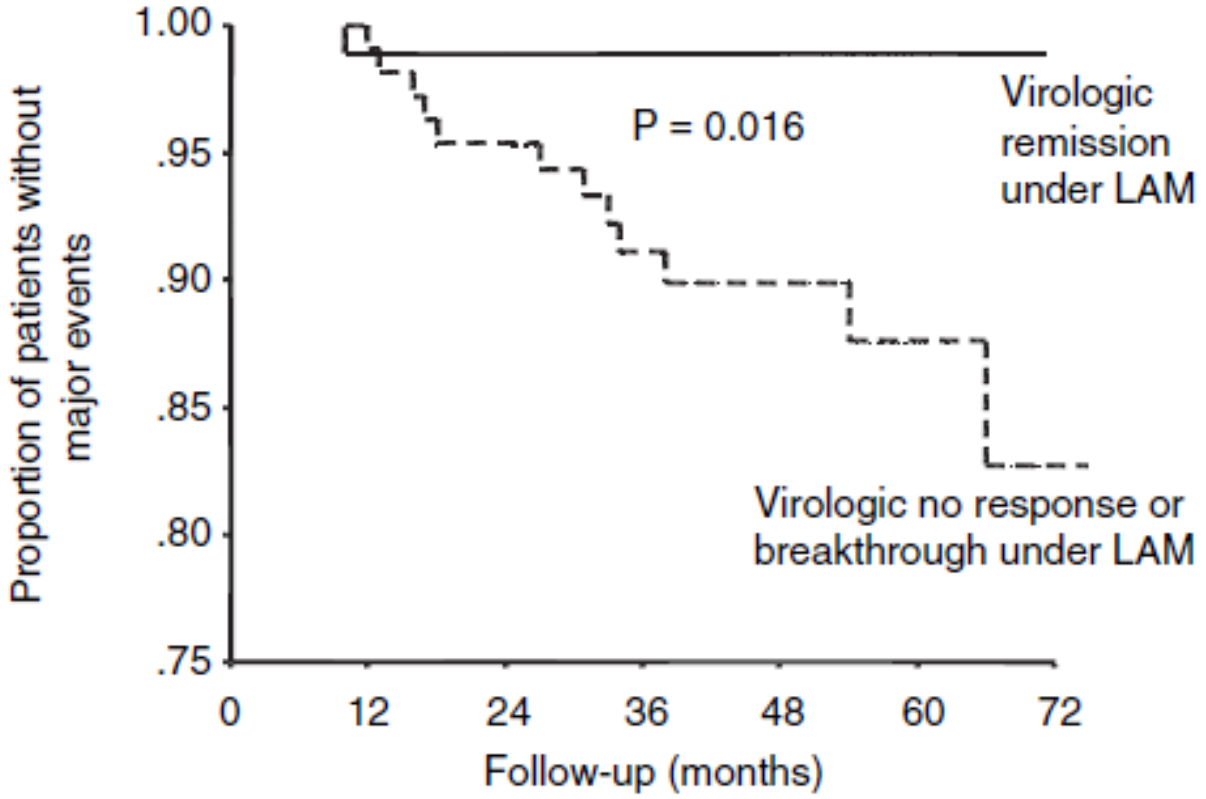
1st Division of Gastroenterology, Department of Liver, Kidney, Lung and Bone Marrow Units and Organ Transplant, A.M. & A. Migliavacca Center for Liver Disease, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico and University of Milan, Milan, Italy

"Successful treatment of chronic hepatitis B can decrease the risk of HCC"

"Patients should still receive monitoring"

Triolo et al. *Liver Int* (2014)

VIROLOGICAL RESPONSE: HBeAg -



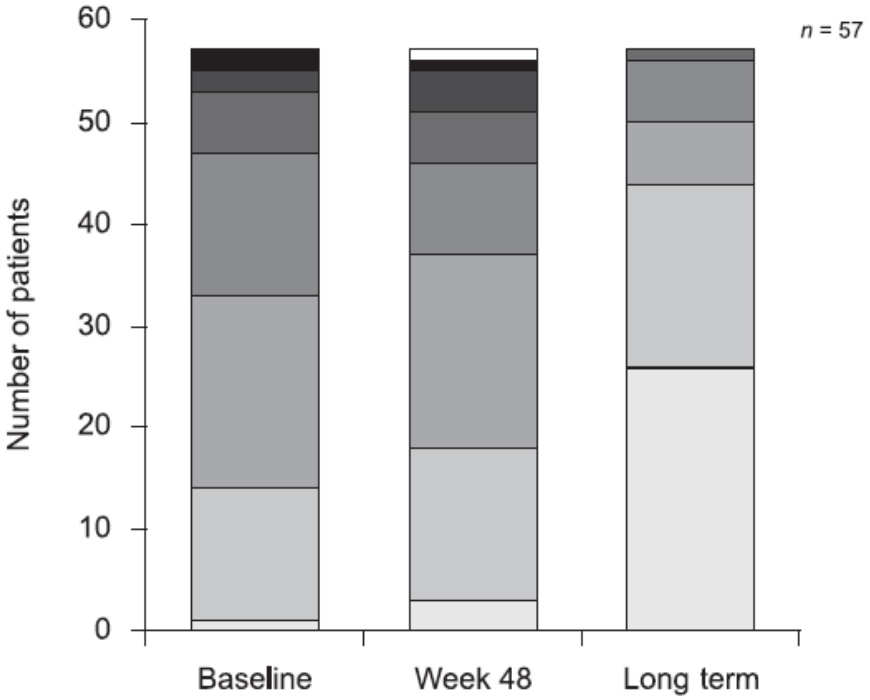
Patients at risk

Vir. remission	92	91	83	54	26	10	4
No response or breakthrough	109	109 (108*)	103 (102*)	78 (77*)	59 (58*)	35 (35*)	17 (16*)

Papatheoridis et al. *Hepatology* (2005)

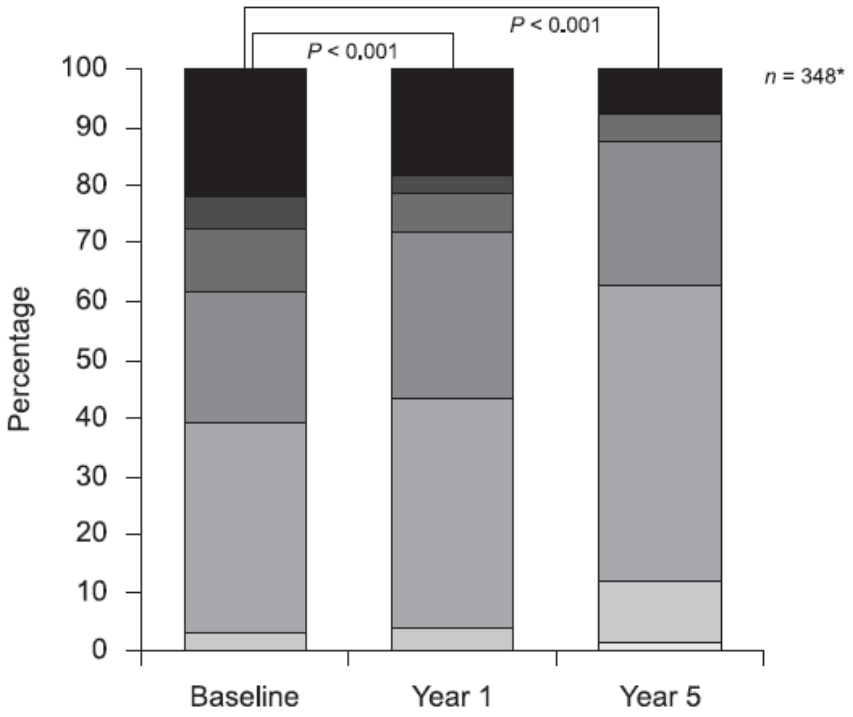
VIROLOGICAL RESPONSE: FIBROSIS REGRESSION

ENTECAVIR



Chang et al. *Hepatology* (2010)

TENOFOVIR



Marcellin et al. Lancet (2013)

ISHAK fibrosis score 1 2 3 4 5 6

P. Marcellin and T. Asselah. *J Gastroenterol Hepatol* (2013)

LIVER STIFFNESS

PORTAL HIPERTENSION
HPVG>10mmHg

Authors, [Ref.]	Patients (n)	Etiologies	Study design	Prevalence of clinically significant portal hyperten-sion (%)	Cut-offs HVPG ≥10 mmHg (kPa)		AUC	Se (%)	Sp (%)	PPV (%)	NPV (%)	+LR	-LR
Carrion *et al.*, [35]	124	HCV-LT	Pro. mono.	21	8.7*	-	0.92	90	81	81	90	4.7	0.12
Vizzutti *et al.*, [36]	61	HCV	Pro. mono.	77	13.6	17.6**	0.99 0.92	97 94	92 81	97 86	92 91	13.7 4.9	0.02 0.08
Sanchez-Condé *et al.*, [39]	38	HIV-HCV	Pro. mono.	74	14.0	23.0**	0.80 0.80	93 83	50 67	84 79	71 71	3.5 2.5	0.62 0.49
Lemoine *et al.*, [38]	44 48	HCV Alcohol	Retro. mono.	77 83	20.5 34.9		0.76 0.94	63 90	70 88	88 97	35 64	2.1 7.5	0.53 0.13
Bureau *et al.*, [37]	150	CLD	Pro. mono.	51	21.0		0.94	90	93	93	91	12.8	0.10

Castera et al. *J Hepatol* (2012)

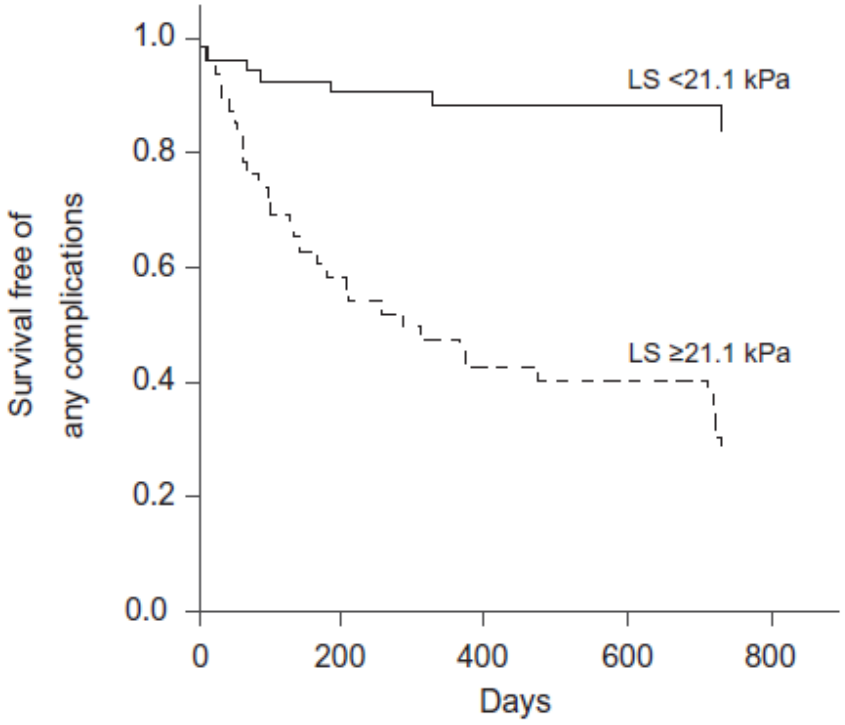
LIVER STIFFNESS

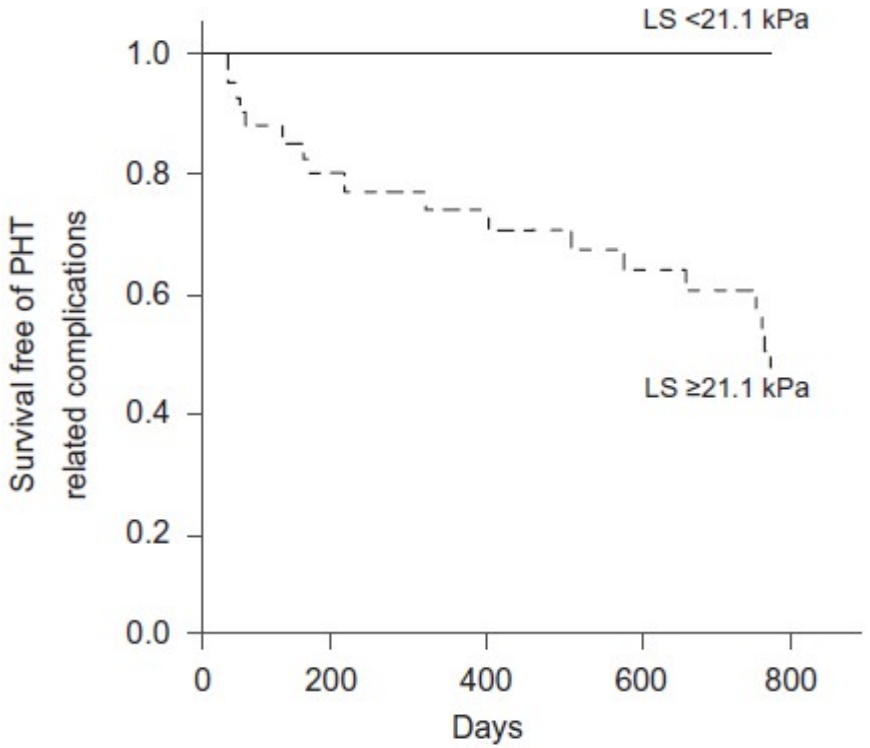
OESOPHAGEAL VARICES

Authors, [Ref.]	Patients (n)	Etiologies	Study design	Child-Pugh A (%)	End point	Prevalence OV (%)	Cut-offs (kPa)	AUC	Se (%)	Sp (%)	PPV (%)	NPV (%)	+LR	-LR	Saved endoscopy (%)
Kazemi *et al.*, [45]	165	CLD	Retro. mono.	n.a.	OV LOV	45 28	13.9 19.0	0.83 0.84	95 91	43 60	57 48	91 95	1.7 2.3	0.13 0.14	66 69
Vizzutti *et al.*, [36]	47	HCV	Pro. mono.	60	OV	66	17.6	0.76	90	43	77	66	1.6	0.23	74
Pritchett *et al.*, [48]	211	CLD	Retro. mono.	n.a.	OV LOV	n.a. 37	19.5 19.8	0.74 0.76	76 91	66 56	56 91	82 55	2.2 2.1	0.36 0.16	n.a. 69
Bureau *et al.*, [37]	89	CLD	Pro. mono.	34	OV LOV	72 48	21.1 29.3	0.85 0.76	84 81	71 61			2.9 2.1	0.22 0.31	81 71
Castera *et al.*, [46]	70	HCV	Retro. mono.	100	OV LOV	36 19	21.5 30.5	0.84 0.87	76 77	78 85	68 56	84 94	3.5 5.1	0.31 0.27	73 79
Pineda, *et al.*, [47]	102	HIV-HCV	Pro. multi.	76	CROV*	13	21.0	0.71	100	32	25	100	1.5	0.0	44
Nguyen et al. [49]	183 58 103	CLD HCV/HBV Alcohol	Retro. mono.	63	LOV	22 17 25	48.0 19.8 47.2	0.76 0.73 0.77	73 89 85	73 55 64	44 27 44	90 97 93	2.7 2.0 2.4	0.37 0.20 0.23	73 60 69
Malik *et al.*, [50]	124	CLD	Retro. mono.	n.a.	OV	51	20.0	0.85	n.a.	n.a.	80	75	n.a.	n.a.	n.a.

Castera et al. *J Hepatol* (2012)

LIVER STIFFNESS





Robic et al. *J Hepatol* (2011)

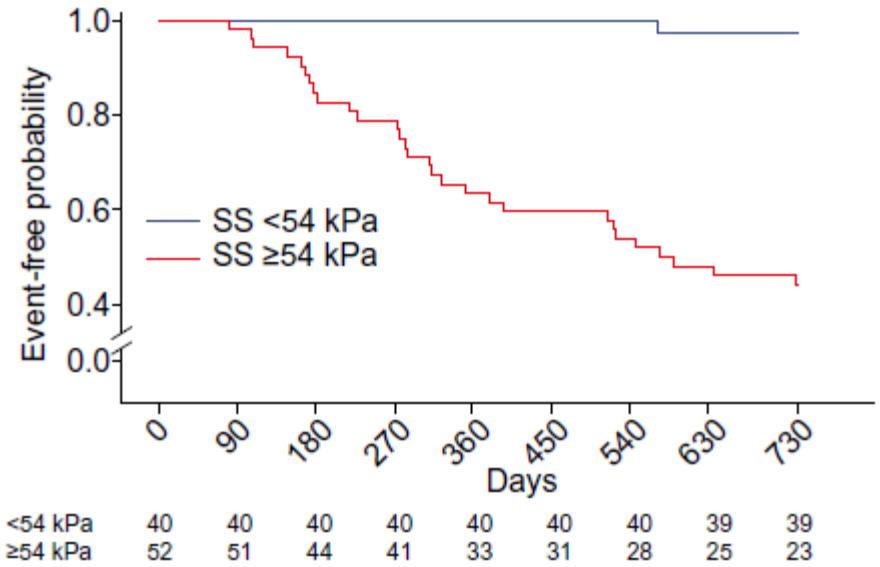
SPLEEN STIFFNESS

Research Article

EASL EUROPEAN ASSOCIATION FOR THE STUDY OF THE LIVER | JOURNAL OF HEPATOLOGY

Spleen stiffness measurement can predict clinical complications in compensated HCV-related cirrhosis: A prospective study

Antonio Colecchia[1,*], Agostino Colli[2], Giovanni Casazza[3], Daniele Mandolesi[1], Ramona Schiumerini[1], Letizia Bacchi Reggiani[4], Giovanni Marasco[1], Martina Taddia[1], Andrea Lisotti[1], Giuseppe Mazzella[1], Anna Rita Di Biase[5], Rita Golfieri[1], Massimo Pinzani[6], Davide Festi[1]



Colecchia et al. *J Hepatol* (2014)

SURVEILLANCE (II)

• In October 2013 the patient had peripheral leg edemas and her GP prescribed furosemide 40 mg bid.

• One week later the patient was admitted in the hepatology unit having insomnia, confusion, distended abdomen and renal dysfx.

•Abd Ultrasound: Portal vein patent (Vm=6cm/sec) + global ascites + mildly enlarged spleen (diameter 141 mm).

• Upper GI: no indirect signs of portal hypertension.

•She was diagnosed with hepatic encephalopathy, ascites and hepatorenal syndrome type II.

SURVEILLANCE (III)

- Lactulose, rifaximin, albumin and terlipressin were started.
- Screening of infections was negative.
- Diuretics were kept to a minimum
- Renal function improved but ascites persisted requiring paracentesis every 5-7 days.
- In January 2014 the patient had a traumatic hemoperitoneum secondary to a paracentesis.
- She was discharged without major complications 7 days after.

QUESTION 4

With the diagnosis of refractory (untreatable) ascites please choose one of the following options:

1) Continue with paracentesis whenever needed.
2) TIPS placement
3) Evaluation for Liver Transplantation
4) Measure HPVG
5) Other alternatives for refractory ascites: Denver Shunt or ALFApump

HEPATIC HAEMODYNAMIC ASSESSMENT

HPVG 8 mmHg

QUESTION 5

With the diagnosis of refractory (untreatable) ascites and HPVG of 8 mmHg, please choose one of the following options:

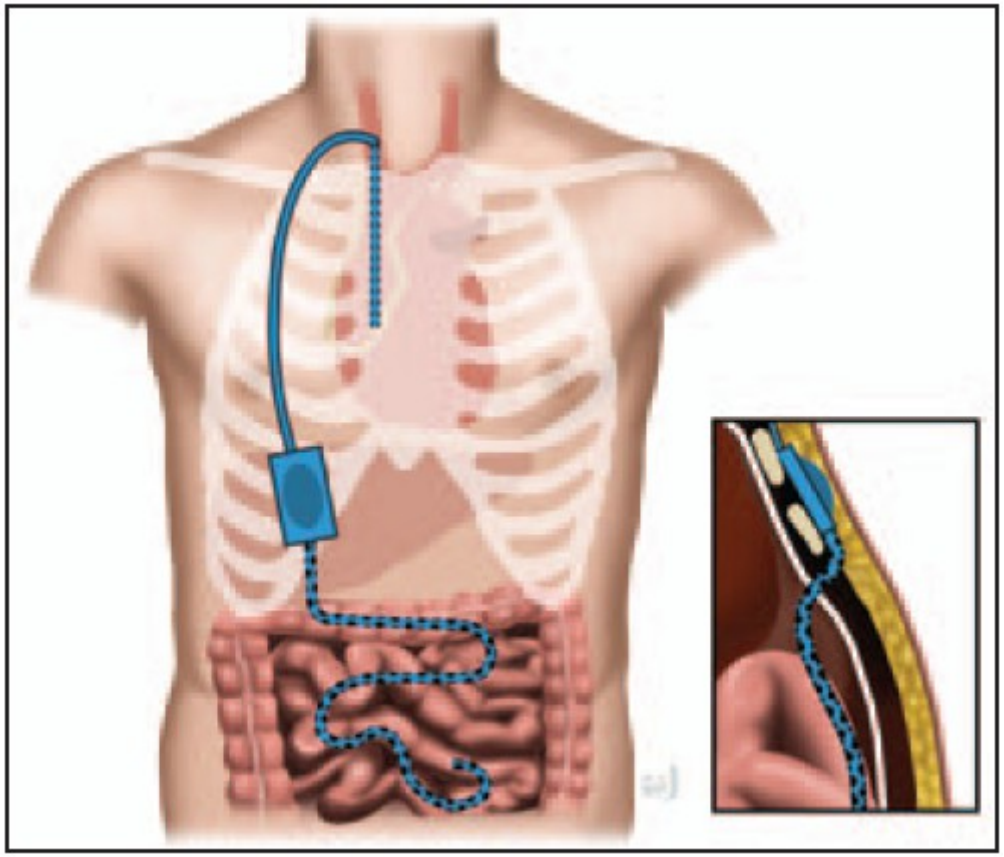
1) Continue with paracentesis whenever needed.

2) Indicate TIPS

3) Evaluation for Liver Transplantation

4) Other alternatives for refractory ascites: Denver Shunt or ALFApump

DENVER SHUNT



Martin et al. AJR (2012)

DENVER SHUNT VS TIPS

TIPS Versus Peritoneovenous Shunt in the Treatment of Medically Intractable Ascites

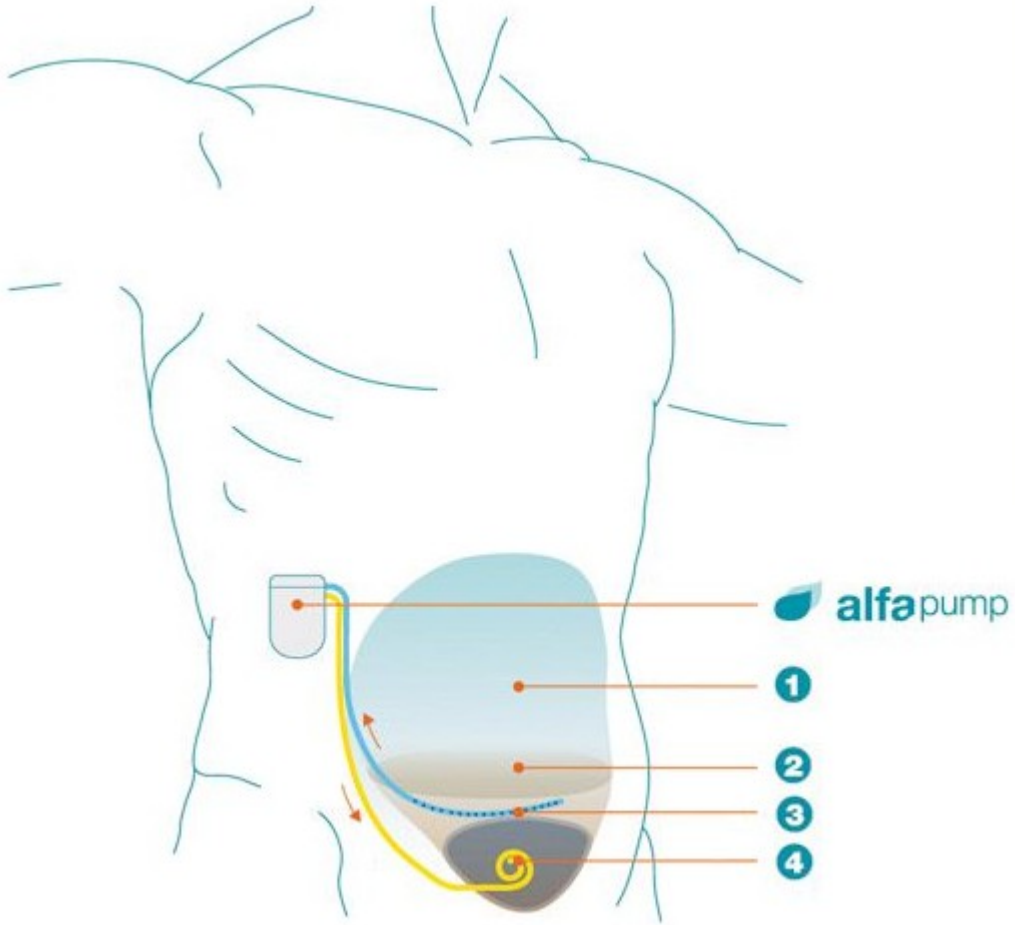
A Prospective Randomized Trial

"There was an insignificant trend in earlier relief of ascites with the Denver shunt"

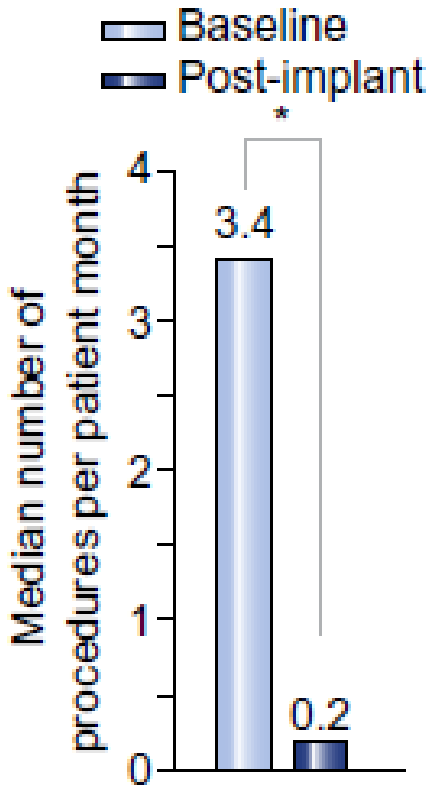
"All patients with Denver shunt failed shunting at 5 years"

Rosemurgy et al. Ann Surg (2004)

ALFAPUMP



ALFAPUMP



Serious adverse events	Number of patients		*p* value
	Cohort I (n = 21)	Cohort II (n = 19)	
Cirrhosis related			
Total	17	13	n.s.
Hepatic encephalopathy	8	9	n.s.
Renal dysfunction	9	4	n.s.
Infections			
Total			
No. of occurences = 0	5 (24%)	11 (58%)	
No. of occurences = 1	12 (57%)	7 (37%)	
No. of occurences >1	4 (19%)	1 (5%)	0.09
Spontaneous bacterial peritonitis			
No. of occurences = 0	13 (62%)	15 (79%)	
No. of occurences = 1	6 (29%)	4 (21%)	
No. of occurences >1	2 (10%)	0 (0%)	0.48

Bellot et al. J Hepatol (2013)

FOLLOW-UP

- In April 2014 the Denver shunt was placed
- No complications occurred
- There was no need of further paracentesis
- In June 2015 the shunt is still patent and the patient is asymptomatic.

TAKE HOME MESSAGES

- In chronic hepatitis B, not invasive fibrosis tests allow to identify patients with advanced fibrosis and cirrhosis.
- Transient elastography (TE) should be preferred to other non-invasive methods.
- TE is useful to select candidates for antiviral therapy, and adds to prognosis.
- In a patient with decompensated liver cirrhosis, TE has limited utility.
- Liver biopsy may still be indicated in doubtful cases, and collagen proportional area is a promising tool for sub-classifying cirrhosis.

RECOMMENDED READING

Clinical Practice Guidelines

EASL JOURNAL OF HEPATOLOGY

EASL Clinical Practice Guidelines: Management of chronic hepatitis B virus infection

European Association for the Study of the Liver*

Introduction

Our understanding of the natural history of hepatitis B virus (HBV) infection and the potential for therapy of the resultant disease is continuously improving. New data have become available since the previous EASL Clinical Practice Guidelines (CPGs) prepared in 2008 and published in early 2009 [1]. The objective of this manuscript is to update the recommendations for the optimal management of chronic HBV infection. The CPGs do not fully address prevention including vaccination. In addition, despite the increasing knowledge, areas of uncertainty still exist and therefore clinicians, patients, and public health authorities must continue to make choices on the basis of the evolving evidence.

Context

Epidemiology and public health burden

Approximately one third of the world's population has serological evidence of past or present infection with HBV and 350–400 million people are chronic HBV surface antigen (HBsAg) carriers. The spectrum of disease and natural history of chronic HBV infection are diverse and variable, ranging from an inactive carrier state to progressive chronic hepatitis B (CHB), which may evolve to cirrhosis and hepatocellular carcinoma (HCC) [2–4]. HBV-related end stage liver disease or HCC are responsible for over 0.5–1 million deaths per year and currently represent 5–10% of cases of liver transplantation [5–8]. Host and viral factors, as well as coinfection with other viruses, in particular hepatitis C virus (HCV), hepatitis D virus (HDV), or human immunodeficiency virus (HIV) together with other co-morbidities including alcohol abuse and obesity, can affect the natural course of HBV infection as well as efficacy of antiviral strategies [2–8]. CHB may present either as hepatitis B e antigen (HBeAg)-positive or HBeAg-negative CHB. The prevalence of the HBeAg-negative form of the disease has been increasing over the last decade as a result of aging of the HBV-infected population and predominance of specific HBV genotypes and represents the majority of cases in many areas, including Europe [4,9,10]. Morbidity and mortality in CHB are linked to persistence of viral replication and evolution to cirrhosis and/or hepatocellular carcinoma (HCC). Longitudinal studies of untreated patients with CHB indicate that, after diagnosis, the 5-year cumulative incidence of developing cirrhosis ranges from 8% to 20%. The 5-year cumulative incidence of hepatic decompensation is approximately 20% for untreated patients with compensated cirrhosis [2–4,11–13]. Untreated patients with decompensated cirrhosis have a poor prognosis with a 14–35% probability of survival at 5 years [2–4,12]. The worldwide incidence of HCC has increased, mostly due to persistent HBV and/or HCV infections; presently it constitutes the fifth most common cancer, representing around 5% of all cancers. The annual incidence of HBV-related HCC in patients with CHB is high, ranging from 2% to 5% when cirrhosis is established [13]. However, the incidence of HBV related HCC appears to vary geographically and correlates with the underlying stage of liver disease and possibly exposure to environmental carcinogens such as aflatoxin. Population movements and migration are currently changing the prevalence and incidence of the disease in several low endemic countries in Europe and elsewhere. Substantial healthcare resources will be required for control of the worldwide burden of disease.

Natural History

Chronic HBV infection is a dynamic process. The natural history of chronic HBV infection can be schematically divided into five phases, which are not necessarily sequential.

(1) The "immune tolerant" phase is characterised by HBeAg positivity, high levels of HBV replication (reflected by high levels of serum HBV DNA), normal or low levels of aminotransferases, mild or no liver necroinflammation and no or slow progression of fibrosis [2,3,6,8]. During this phase, the rate of spontaneous HBeAg loss is very low. This first phase is more frequent and more prolonged in subjects infected perinatally or in the first years of life. Because of high levels of viremia, these patients are highly contagious.
(2) The "immune reactive HBeAg-positive phase" is characterised by HBeAg positivity, relatively lower level of replication compared to the immune tolerant phase (as reflected by lower serum HBV DNA levels), increased or

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Clinical Practice Guidelines

EASL JOURNAL OF HEPATOLOGY

EASL-ALEH Clinical Practice Guidelines: Non-invasive tests for evaluation of liver disease severity and prognosis

European Association for the Study of the Liver*,
Asociación Latinoamericana para el Estudio del Hígado

Introduction

Liver fibrosis is part of the structural and functional alterations in most chronic liver diseases. It is one of the main prognostic factors as the amount of fibrosis is correlated with the risk of developing cirrhosis and liver-related complications in viral and non-viral chronic liver diseases [1,2]. Liver biopsy has traditionally been considered the reference method for evaluation of tissue damage such as hepatic fibrosis in patients with chronic liver disease. Pathologists have proposed robust scoring system for staging liver fibrosis such as the semi-quantitative METAVIR score [3,4]. In addition computer-aided morphometric measurement of collagen proportional area, a partly automated technique, provides an accurate and linear evaluation of the amount of fibrosis [5]. Liver biopsy gives a snapshot and not an insight into the dynamic changes during the process of fibrogenesis (progression, static or regression). However, immunohistochemical evaluation of cellular markers such as smooth muscle actin expression for hepatic stellate cell activation, cytokeratin 7 for labeling ductular proliferation or CD34 for visualization of sinusoidal endothelial capillarization or the use of two-photon and second harmonic generation fluorescence microscopy techniques for spatial assessment of fibrillar collagen, can provide additional "functional" information [6,7]. All these approaches are valid provided that the biopsy is of sufficient size to represent the whole liver [4,8]. Indeed, liver biopsy provides only a very small part of the whole organ and there is a risk that this part might not be representative for the amount of hepatic fibrosis in the whole liver due to heterogeneity in its distribution [9]. Extensive literature has shown that increasing the length of liver biopsy decreases the risk of sampling error. Except for cirrhosis, for which micro-fragments may be sufficient, a 25 mm long biopsy is considered an optimal specimen for accurate evaluation, though 15 mm is considered sufficient in most studies [10]. Not only the length but also the caliber of the biopsy needle is important in order to obtain a piece of liver of adequate size for histological evaluation, with a 16 gauge needle being considered as the most appropriate [11] to use for percutaneous liver biopsy. Interobserver variation is another potential limitation of liver biopsy which is related to the discordance between pathologists in biopsy interpretation, although it seems to be less pronounced when biopsy assessment is done by specialized liver pathologists [12]. Beside technical problems, liver biopsy remains a costly and invasive procedure that requires physicians and pathologists to be sufficiently trained in order to obtain adequate and representative results – this again limits the use of liver biopsy for mass screening. Last but not least, liver biopsy is an invasive procedure, carrying a risk of rare but potentially life-threatening complications [13,14]. These limitations have led to the development of non-invasive methods for assessment of liver fibrosis. Although some of these methods are now commonly used in patients for first line assessment, biopsy remains within the armamentarium of hepatologists when assessing the etiology of complex diseases or when there are discordances between clinical symptoms and the extent of fibrosis assessed by non-invasive approaches.

Methodological considerations when using non-invasive tests

The performance of a non-invasive diagnostic method is evaluated by calculation of the area under the receiver operator characteristic curve (AUROC), taking liver biopsy as the reference standard. However, biopsy analysis is an imperfect reference standard: taking into account a range of accuracies of the biopsy, even in the best possible scenario, an AUROC >0.90 cannot be achieved for a perfect marker of liver disease [15]. The AUROC can vary based on the prevalence of each stage of fibrosis, described as spectrum bias [16]. Spectrum bias has important implications for the study of non-invasive methods, particularly in comparison of methods across different study populations. If extreme stages of fibrosis (F0 and F4) are over-represented in a population, the sensitivity and specificity of a diagnostic method will be higher than in a population of patients that has predominantly middle stages of fibrosis (F1 and F2). Several ways of preventing the "spectrum bias" have been proposed including the adjustment of AUROC using the DANA method (standardization according to the prevalence of fibrosis stages that define advanced (F2–F4) and non-advanced (F0–F1) fibrosis) [17,18] or the Obuchowski measure (designed for ordinal gold standards) [19]. What really matters in clinical practice is the number of patients correctly classified by non-invasive methods for a defined endpoint according to the reference standard (i.e. true positive and true negative).

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Thank you