Research Article

JOURNAL OF HEPATOLOGY

Delisting of liver transplant candidates with chronic hepatitis C after viral eradication: A European study

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Background & Aims: All oral direct acting antivirals (DAA) have been shown to improve the liver function of patients with decompensated cirrhosis but it is presently unknown whether this clinical improvement may lead to the delisting of some patients. The aim of this study was to assess if and which patients can be first inactivated due to clinically improvement and subsequently delisted in a real life setting.

Methods: 103 consecutive listed patients without hepatocellular carcinoma were treated with different DAA combinations in 11 European centres between February 2014 and February 2015.

Results: The cumulative incidence of inactivated and delisted patients by competing risk analysis was 15.5% and 0% at 24 weeks, 27.6% and 10.3% at 48 weeks, 33.3% and 19.2% at 60 weeks. The 34 patients who were inactivated showed a median improvement of 3.4 points for MELD (delta MELD, p <0.0001) and 2 points for Child-Pugh (CP) (delta-CP, p <0.0001). Three variables emerged from the most parsimonious multivariate competing risk model as predictors of inactivation for clinical improvement, namely, baseline MELD classes (MELD 16–20: HR = 0.120; p = 0.0005, MELD >20:HR = 0.042; p <0.0001), delta MELD (HR = 1.349; p <0.0001) and delta albumin (HR = 0.307; p = 0.0069) both assessed after 12 weeks of DAA therapy.

Conclusions: This study showed that all oral DAAs were able to reverse liver dysfunction and favoured the inactivation and delisting of about one patient out-of-three and one patient out-of-five in 60 weeks, respectively. Patients with lower MELD scores had higher chances to be delisted. The longer term benefits of therapy need to be ascertained.

Lay summary: The excellent efficacy and safety profile of the new drugs against Hepatitis C virus, "direct acting antivirals" or DAAs, have made antiviral therapy possible also for patients with advanced liver disease and for those on the waiting list for liver transplantation (LT). This study shows for the first time that the DAAs may lead to a remarkable clinical improvement allowing the delisting of one patient out of 5.

Keywords: Direct acting antivirals; Liver transplantation; Delisting; HCV; Cirrhosis.

Received 16 November 2015; received in revised form 30 April 2016; accepted 4 May 2016; available online 17 May 2016

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Abbreviations: DAAs, direct acting antivirals; HCV, hepatitis C virus; LT, liver transplantation; HCC, hepatocellular carcinoma; MELD, model for end-stage liver disease; ELITA, European Liver and Intestine Transplant Association; HIV, human immunodeficiency virus; HBV, hepatitis B virus; HE, hepatic encephalopathy; HPS, hepato-pulmonary syndrome; HCV-RNA, hepatitis C virus-ribonucleic acid; SOF, sofosbuvir; RBV, ribavirin; DCV, daclatasvir; LDV, ledipasvir; SMV, simeprevir; INR, international normalized ratio; SD, standard deviation; IQR, interquartile range; CP, Child-Pugh; WL, waiting list; RVR, rapid virological response; EVR, early virological response; SVR, sustained virological response; EOT, end of treatment; EMA, European Medicines Agency; FDA, Food and Drug Administration.

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Introduction

The availability of new direct acting antivirals (DAAs) has radically changed the approach to the treatment of hepatitis C virus (HCV) infection and also the prognosis of patients with HCV-related liver disease. The excellent efficacy and safety profile of these drugs and the potential to use all interferon-free regimes, have made antiviral therapy possible also for patients with advanced liver disease and for those on the waiting list for liver transplantation (LT).

Interim and preliminary data from on-going clinical trials indicate that new DAAs given to patients with decompensated cirrhosis are highly effective in eradicating HCV infection and may lead, in some cases, to a significant clinical improvement [1–6] with reversal of de-compensation. These data are prompting the liver transplant community to explore whether the same favourable results can be obtained in liver transplant candidates but, more importantly, whether they may eventually allow the inactivation/delisting of some patients due to clinical improvement [7]. Several transplant centres across Europe have started using these drugs, but clinical trials or reports of field experience are lacking.

To verify the validity of this new scenario, we initially conducted a survey focused on HCV positive liver transplant candidates with decompensated cirrhosis without hepatocellular carcinoma (HCC) and who had been treated with the new DAAs at different European liver transplant centres. The preliminary results of this survey were discussed at an "ad hoc" European Liver and Intestine Transplant Association (ELITA) monothematic conference in Milan and were the basis for the development of an extended database recording patients with HCV-related decompensated cirrhosis and no HCC, listed for transplantation and treated with second generation DAAs while on the waiting list. The objective of this multicentre European study was to understand the impact of DAAs on inactivation/delisting due to clinical improvement in a real life setting.

Patients and methods

A monothematic conference organized by ELITA regarding the use of second generation DAAs both before and after LT was held in Milan on 6 March 2015. This event allowed experts from several liver transplant centres across Europe to share their experience on the day to day use of these novel treatments which became available about 1 year earlier.

At the conference, it was decided to retrospectively collect data from patients listed for decompensated cirrhosis and consecutively treated with 2nd generation DAAs during the waiting period between February 2014 and February 2015 and were followed until 31 December 2015. Eleven European centres participated to this study: Bergamo, Bologna, Milan Niguarda, Milan Policlinico, Montpellier, Paris Mondor, Villejuif Paris Paul Brousse, Palermo, Turin, Valencia and Vienna.

Inclusion criteria

Consecutive liver transplant candidates with decompensated HCV cirrhosis without HCC treated with second generation DAAs while listed for LT.

Criteria for listing

Basically, patients were listed if they had a MELD score >15 or a MELD score <15 with MELD exceptions such as refractory ascites not treatable with transjugular intrahepatic portosystemic shunt (TIPS) (8 cases), chronic hepatic encephalopathy (13 cases), hepato-pulmonary syndrome (2 cases) and refractory bleeding (2 cases). In addition, 11 patients were judged worth listing despite a MELD score <15 and no clear MELD exception. Overall 35 patients were listed with MELD <15. The distribution of patients with MELD <15 was similar across centres.

Exclusion criteria

HIV or HBV co-infected recipients were excluded from this study as well as patients who had started DAA treatment before listing.

Definitions and patient stratifications

In case of clinical improvement due to DAAs therapy the following definitions were used:

Inactivation: patient is placed "on hold" due to clinical improvement based on clinical judgement of local investigator. For such a patient the clinician judges that, based on liver function and/or clinical improvements, LT is presently no longer indicated, but the patient is not removed from the list until a long-term clinical improvement has been verified. Defining "clinical improvement leading to inactivation" was one the aims of the study.

Delisting: patient is off the list because a durable clinical improvement has been verified based on clinical judgement of local investigator.

End points

The primary end points were the probability to be inactivated due to clinical improvement.

Secondary end points included virological efficacy, DAA-related improvement of liver function, description of the objective criteria taken into account by investigators for considering inactivation and eventually delisting.

To achieve these goals, the following parameters were retrospectively collected:

Baseline: demographics, indication for LT, genotype, previous antiviral therapy, HCV-RNA levels, DAA regimen used (sofosbuvir/ribavirin-SOF/RBV or sofosbuvir/daclatasvir-SOF/DCV or sofosbuvir/ledipasvir-SOF/LDV or sofosbuvir/simeprevir-SOF/SMV) and duration, MELD score, CP scores and individual components of MELD and CP scores (bilirubin, INR, creatinine, albumin, ascites and encephalopathy). Cofactors for liver decompensation, such as alcohol consumption, bacterial infections, haemorrhagic events and portal vein thrombosis were also registered.

During therapy and follow-up: HCV-RNA levels at 4, 8, 12, 16 and 24 weeks. MELD, CP scores and individual components of MELD and CP scores (bilirubin, INR, creatinine, albumin, ascites and encephalopathy) at 12 and 24 weeks. For those receiving ribavirin, median ribavirin dose was registered.

Outcome. Seven clinical outcomes were identified and registered: 1) liver transplantation, 2) patient still waiting for a liver transplant, 3) death while waiting for transplant, 4) patient inactive in the transplant list due to clinical improvement, 5) patient delisted due to clinical improvement, 6) patient dropout due to other causes (e.g., clinical worsening, refused liver transplant) and 7) death after inactivation or delisting.

Type and duration of antiviral treatment

Planned duration of treatment was up to 48 weeks or until transplant for patients receiving SOF/RBV and up to 24 weeks or until transplant for those receiving SOF/DCV or SOF/LDV or SOF/SMV with or without RBV. DAAs combinations were used depending on genotype and drug availability.

Ethical approval was not sought as the study utilised data provided in the course of normal patient care and no patient-identifiable data were collected.

Statistical analysis

Descriptive statistical analysis was performed where data are expressed as median (interquartile range (IQR) or range). Categorical variables were compared with the Chi-square test or 2-sided Fisher's exact test, continues variables were analysed by the student's *t* test or by Wilcoxon's rank-sum test as appropriate. McNemar's test or Bowker's test were used to compare categorical variables before and after treatment while paired student's *t* test or Mann-Whitney *U* test was used for continues variables as appropriate.

The impact of DAA on liver functions (MELD, CP score, bilirubin, creatinine, etc.) was assessed over time at 12 and 24 weeks after start of therapy. The same parameters were analysed comparing inactivated vs non-inactivated patients. For

patients with LT or death before the 24 weeks, we considered the last available value. The MELD and CP score changes were also assessed stratifying patients in different MELD (<16, 16–20 and >20) and CP classes (B and C) at baseline.

Cumulative incidence curves for inactivation from waiting list were constructed considering time to inactivation and time to LT and time to death as competing risks [8]. Further, cumulative incidence curves for delisting were constructed considering time to delisting and time to LT and time to death as competing risks. Time to inactivation, time to delisting, time to LT and time to death were measured from the date of treatment start.

Clinical and demographic characteristics at baseline were tested as prognostic factors for inactivation. The prognostic significance of clinical and demographic characteristics was determined by univariate Cox regression model for competing risks [9]. Further, changes of MELD, CP scores and their individual components after 12 weeks of treatment were tested as possible dynamic predictors for inactivation using the same method. Changes of MELD and CP sores were tested as continuous and as two level-categorical data. The level of MELD and CP improvement (Delta MELD and Delta CP at 12 weeks) were identified using an outcome-oriented approach, as proposed by Contal and O'Quigley [10]. Variables that were lower than p <0.1 on univariate analysis were tested into multivariate Cox regression models for competing risks, and the most parsimonious model was selected. Based on the selected multivariate model, the inactivation cumulative incidence was estimated for hypothetical patients to show the effect of the variables included. The inactivation cumulative incidence of simulated patients was estimated by the function $\widehat{\boldsymbol{F}}(\boldsymbol{t},\boldsymbol{z_0}) = \mathbf{1} - \mathbf{exp}\{-\widehat{\Lambda}_1(\boldsymbol{t},\boldsymbol{z_0})\}\widehat{\mathbf{F}}(\mathbf{t},\mathbf{z_0}) = \mathbf{1} - \mathbf{exp}\{-\widehat{\Lambda}_1(\mathbf{t},\mathbf{z_0})\}$ where $\widehat{\Lambda}(\boldsymbol{t},\boldsymbol{z_0})\widehat{\Lambda}_1(\mathbf{t},\mathbf{z_0})$ is the empirical cumulative distribution hazard of inactivation and z_0 are the individual covariates [9]. All analyses were performed using SAS software (version 9.4; SAS Institute Inc., Cary, NC).

Results

Between February 2014 and February 2015, 134 patients were listed for decompensated cirrhosis without HCC; of these, 103 were treated with DAA and 31 were not. Reasons for not treating were the following: 21 patients were considered too close to LT (13 had a MELD score >25 and 8 a MELD score between 18 and 24); 6 patients were included in a study protocol of pre-emptive post LT DAA therapy; 2 patients declined the opportunity to be treated before LT and for 2 patients the compassionate drug did not arrive in time.

Baseline features of the 103 treated patients are reported in Table 1. The median (IQR) follow-up was 51.9 (32.9–67.4) weeks. Median MELD and CP scores at baseline were 16 and 10, respectively. Of note, 50.5% of the patients had been treated with SOF and RBV, 34% with SOF + DCV, 8.7% with SOF + LDV and 6.8% with SOF + SMV. The treatment duration for each type of DAA treatment is reported in Supplementary Table 1.

Virological outcomes

Virological clearance, defined as HCV-RNA <LLOQ with a detection threshold of 15 IU, was observed in 66 patients after 4 weeks of therapy (rapid virological response-RVR, 66/103, 64%) and in 32 additional patients after 12 weeks (early virological response-EVR, 98/100 = 98%), Fig. 1. Three patients could not be evaluated for EVR as two had received a LT and one had died before week 12. Of the 2 patients who remained viremic after 12 weeks, 1 underwent LT and the second had viral re-activation after he temporarily stopped SOF/RBV for 10 days due to acute pancreatitis.

Considering the 52 patients receiving SOF/RBV (which was planned for up to 48 weeks or until LT), 28 completed the entire course of therapy and 4 had a relapse. These 4 patients are presently on retreatment with a dual DAAs regimen. Twenty-two patients underwent LT while on therapy and the remaining 2 patients died of sepsis while on therapy. These 2 patients had a baseline MELD score of 17 and 22. Only one of the 22 patients who had received a LT, had a viral relapse after the operation and is under dual DAA therapy.

Fifty-one patients were treated with SOF/DCV (n = 35) or SOF/LDV (n = 9) or SOF/SMV (n = 7). They all completed their course of treatment and none relapsed while listed. One patient treated with SOF/DCV had a relapse after LT and he is presently under retreatment with IFN/SOF/DCV and RBV.

Table 1. Baseline characteristics of treated patients (N = 103).

Variables		Values
Age	Median (range)	54 (37-71)
Male	N (%)	70 (68.0)
HCV-RNA IU/ml	Median (range)	237,000 (1334-100,000,000)
Genotype, N (%)	1a	20 (19.4)
	1b	40 (38.8)
	2	3 (2.9)
	3	20 (19.4)
	4	20 (19.4)
MELD	Median (range)	16 (6-31)
MELD classes, N (%)	<16	51 (49.5)
	16-20	38 (36.9)
	>20	14 (13.6)
Child-Pugh	Median (range)	10 (7-13)
Child-Pugh classes, N (%)	A 5-6	0
	B 7-9	46 (44.6)
	C 10-14	57 (55.3)
Ascites, N (%)	None	15 (14.6)
	Medically controlled	61 (59.2)
	Medically uncontrolled	27 (26.2)
Encephalopathy, N (%)	None	54 (52.4)
	Medically controlled	48 (46.6)
	Medically uncontrolled	1 (1.0)
DAAs treatment, N (%)	SOF/RBV	52 (50.5)
	SOF/LDV ± RBV	9 (8.7)
	SOF/DCV ± RBV	35 (34.0)
	SOF/SMV ± RBV	7 (6.8)

MELD, Model for End-stage Liver Disease; DAAs, direct acting antivirals; SOF, sofosbuvir; RBV, ribavirin; LDV, ledipasvir; DCV, daclatasvir; SMV, simeprevir.

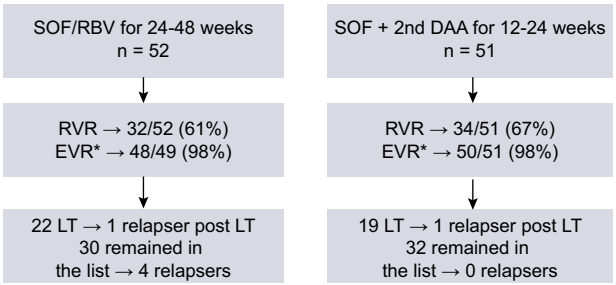

Fig. 1. Virological outcome of 103 liver listed patients with decompensated cirrhosis and treated with DAAs based therapy while listed. DAAs, direct acting antivirals; RVR, rapid virological response; EVR, early virological response; SOF, sofosbuvir; RBV, ribavirin. *Three patients could not be evaluated for EVR (see text).

Clinical outcome

Of the entire cohort of 103 patients, 4 (3.9%) died while on active list (2 of sepsis, 1 of massive bleeding and 1 of heart failure after TIPS placement), 41 (39.8%) underwent LT, 22 (21.3%) are waiting for a transplant and 34 (33%) were inactivated from the transplant list due to clinical improvement. Of these 34, 21 were delisted and one died from massive bowel infarction 4 months after the end of DAA treatment. The 2 remaining patients were delisted for reasons not related to clinical improvement (one for cardiac problems and the other declined the transplant).

Impact of DAA on liver function

The impact of DAA on liver function was assessed on 102 of the 103 patients because one patient died of sepsis before 12 weeks of treatment.

Impact on MELD score

The evolution of MELD score from start of DAA therapy to 24 weeks afterwards is shown in Supplementary Table 2. Overall, the median MELD score improved from 15.5 to 14.0 ($p = 0.0008$).

Impact on CP score

Median CP score improved from 10.0 before DAA to 8.0 on week 24 after start of treatment ($p < 0.001$) (Supplementary Table 2).

Impact of DAA on individual variables reflecting liver function

Details of the impact of DAA on bilirubin, prothrombin time, creatinine, albumin, ascites and encephalopathy are shown in Supplementary Tables 3–5. The more relevant biochemical changes after 24 weeks from start of therapy were a median increase of albumin by 0.3 g/dl ($p < 0.0001$) and a median reduction of bilirubin by 0.4 mg/dl ($p = 0.004$). In addition, the percentage of patients with refractory ascites halved from 26% at baseline to 13% after 24 weeks ($p < 0.0001$) and stage 2 HE regressed in almost 2 thirds of affected patients ($p < 0.0001$) (Supplementary Tables 4 and 5).

The improvement of MELD score, CP score and individual variables was already detectable at 12 weeks as shown in Supplementary Table 6.

Impact of DAA on inactivation and delisting

Characteristics of delisted patient groups

Overall, 34 patients were considered to improve clinically enough to be inactivated on the waiting list. Of these, 21 patients (62%) were de-listed after a more prolonged period of clinical improvement. During follow-up the cumulative incidence of inactivated patients at 24, 48 and 60 weeks after the start of DAAs therapy, as assessed by competing risk analysis was 15.5%, 27.6% and 33.3% (Fig. 2A) while, the cumulative incidence of delisted patients at the same time points was 0%,10.3% and 19,2% respectively (Fig. 2B).

The median (IQR) time from the start of treatment to the inactivation and delisting was 25.6 (16.9–38.0) and 52.4 (37.1–59.3) week respectively. The minimum time interval between start of therapy and inactivation or delisting was 12 and 27 weeks respectively.

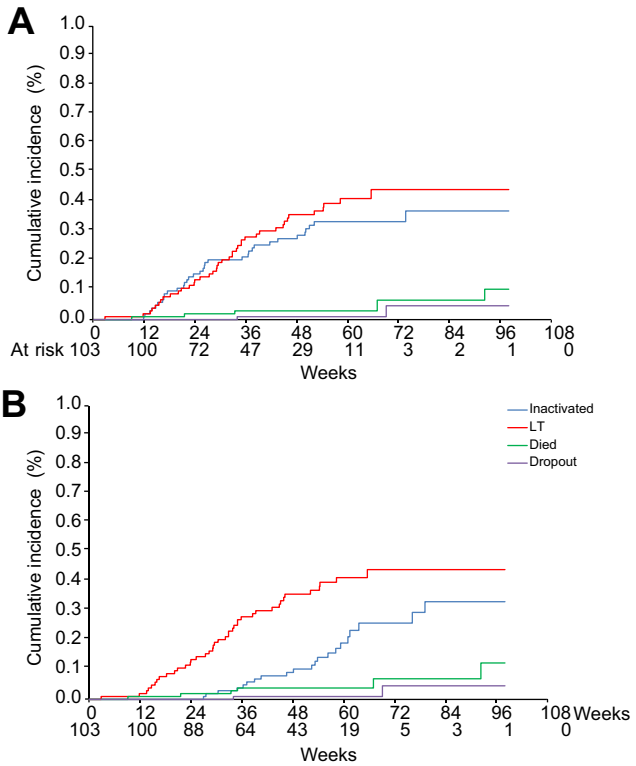

Fig. 2. Competing risk cumulative incidence of patients. (A) Patients that were inactivated, dropped out, underwent LT or died. (B) Of those patients that were delisted, dropped out, underwent LT or died.

Clinical features of inactivated and delisted patients

The clinical features of patients who could be inactivated for clinical improvement, were compared with those observed in patients who were non-inactivated (Table 2). Patients who were inactivated had a baseline median MELD and CP score of 14 and 9, respectively. The median improvement after 24 weeks from start of therapy was minus 3.4 points for MELD (delta MELD, $p < 0.0001$) and minus 2 points for CP (Delta CP, $p < 0.0001$). The greatest improvement of the MELD score was observed within the first 12 weeks from start of therapy, from 14 to 12 during the first 12 weeks and from 12 to 10.5 from week 12 to week 24 (Fig. 3A). In contrast CP steadily improved until week 24 (Fig. 3B).

The more relevant biochemical changes after 24 weeks from start of therapy in patients who could be inactivated for clinical improvement, were a median increase of albumin by 0.5 g/dl ($p = 0.0002$), a median reduction of bilirubin by 0.9 mg/dl ($p = 0.0036$) and median reduction in INR by 0.13 points ($p = 0.002$) (Table 2). In contrast, creatinine, which was in the normal range in most cases at starting of DAA, did not change significantly over the study period. Additionally, in inactivated patients, ascites improved dramatically (all cases of refractory ascites at baseline became treatable with diuretics after 24 weeks) and stage 2 HE regressed in all patients but 1 (Supplementary Tables 7 and 8).

The improvement of MELD score, CP score and individual variables in inactivated patients was already detectable at 12 weeks as shown in Supplementary Table 9.

Table 2. Change of biochemical parameters, MELD and Child-Pugh after 24 weeks of follow-up comparing non-inactivated *vs.* inactivated patients (N = 102).[#]

	Non-inactivated (68 patients)		Inactivated (34 patients)		
Variables	N	Median (IQR)	N	Median (IQR)	*p* value*
Albumin at start of therapy	64	2.9 (2.7-3.25)	31	3.1 (2.8-3.6)	0.141
Albumin at 24 wk	59	3.1 (2.8-3.5)	33	3.5 (3.3-4.3)	<0.0001
Delta albumin (24 wk-start)	58^	0.14 (-0.1-0.4)	31^^	0.5 (0.2-0.9)	0.0002
Bilirubin at start of therapy	68	3.35 (2.36-5.1)	34	2.455 (1.63-3.05)	0.0046
Bilirubin at 24 wk	64	2.8 (1.95-4.515)	34	1.38 (0.99-2)	<0.0001
Delta bilirubin (24 wk-start)	64[§]	-0.045 (-1.035-0.555)	34	-0.925 (-1.62- -0.13)	0.0036
INR at start of therapy	68	1.52 (1.305-1.765)	34	1.385 (1.27-1.53)	0.0145
INR at 24 wk	63	1.49 (1.39-1.68)	34	1.25 (1.2-1.39)	<0.0001
Delta INR (24 wk-start)	63[†]	0.02 (-0.13-0.11)	34	-0.13 (-0.26-0.01)	0.002
Creatinine at start of therapy	68	0.9 (0.745-1)	34	0.905 (0.71-1.11)	0.9462
Creatinine at 24 wk	63	0.9 (0.7-1.03)	34	0.875 (0.71-1)	0.8707
Delta creatinine (24 wk-start)	63[†]	0.02 (-0.09-0.11)	34	0.0075 (-0.09-0.11)	0.7053
MELD at start of therapy	68	16 (14-19)	34	14 (12-16)	0.0019
MELD at 24 wk	68	15.65 (13-19)	34	10.5 (9-13)	<0.0001
Delta MELD (24 wk-start)	68	0 (-2-2)	34	-3.4 (-5- -1)	<0.0001
CPT at start of therapy	68	10 (9-11)	34	9 (8-10)	0.0009
CPT at 24 wk	66	9 (8-11)	34	6.5 (6-7)	<0.0001
Delta CPT (24 wk-start)	66[‡]	0 (-1-1)	34	-2 (-4- -1)	<0.0001

[#]1 patient who died before week 12 is not considered in the analysis; *Mann-Whitney *U* test; ^4 data not used because patients supplemented with albumin, 2 data missing because of LT before week 12 and 4 missing data at week 24; ^^2 data not used because patients supplemented with albumin; [§]2 data missing at week 24 because of LT before week 12; [†]2 data missing at week 24 because of LT before 12 weeks and 3 missing data at week 24; [‡]2 missing data at week 24 because of LT before week 12.

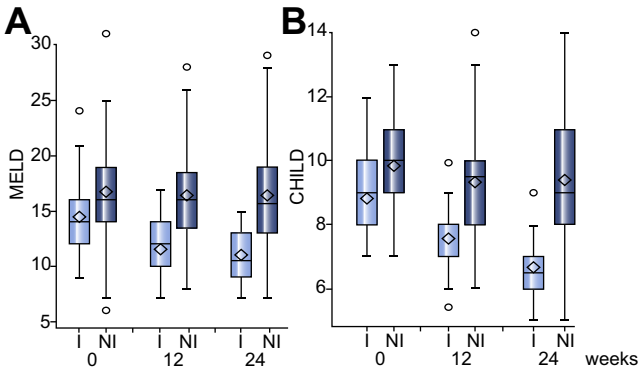

Fig. 3. MELD and Child-Pugh score changes over time in inactivated (N = 34) and non-inactivated patients (N = 68). (A) MELD score changes at 12 and 24 weeks from start of therapy. (B) Child-Pugh score changes at 12 and 24 weeks from start of therapy. I, inactivated; NI, non-inactivated. 1 patient who died before week 12 is not considered in the analysis.

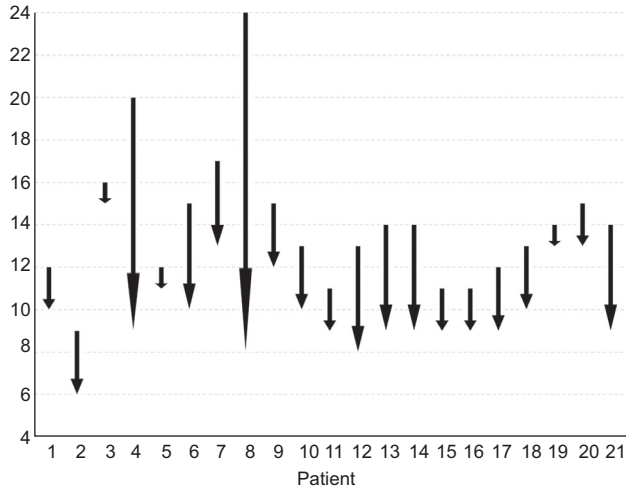

Fig. 4. Delisted patients: individual MELD score at baseline and at delisting.

The twenty-one patients that were delisted, had the following MELD score at start of therapy: 24 (1 patients), 20 (1 patient), 17 (1 patient), 16 (1 patient), 15 (3 patients), 14 (4 patients), 13 (3 patients), 12 (3 patients), 11 (3 patients) and 9 (1 patient). Sixteen of them (77%) showed a complete regression of signs of liver decompensation. These patients no longer had ascites or any kind of fluid retention at delisting and were off any diuretic therapy. When chronic HE was present, it also completely regressed and patients were off any medical therapy. These patients reported they felt well and became very active. Their median MELD score at delisting was 10 (8–12). The remaining 5 patients (23%) still presented some fluid retention at delisting requiring low doses of diuretics. Two of these 5 patients had a refractory ascites at start of therapy. The MELD score at delisting of these 5 patients was 6, 8, 9 and 13 (2 patients) (Fig. 4).

Predictors of inactivation for clinical improvement

We considered both static factors (demographics and clinical characteristics at baseline) and various dynamic parameters (improvement of CP, MELD and of their individual components after 12 weeks of DAAs therapy) in the univariate Cox proportional analysis for competing risks (Supplementary Table 10). Three static factors, namely, baseline MELD score, baseline CP score and baseline INR were significantly associated with inactivation. Among the dynamic parameters, 5 emerged as significant predictors (p <0.05) of inactivation, namely, 12-week delta MELD

Table 3. Competing risk analyses of inactivation from list.

Variable	Category	HR (95% CI)	*p* value
Delta MELD at 12 wk	c.v.	1.349 (1.2-1.516)	<0.0001
MELD at baseline	<16	1	Ref.
	16-20	0.12 (0.036-0.396)	0.0005
	>20	0.042 (0.013-0.138)	<0.0001
Delta albumin at 12 wk	c.v.	0.307 (0.13-0.724)	0.0069

Multivariable model. c.v., continuous variable.

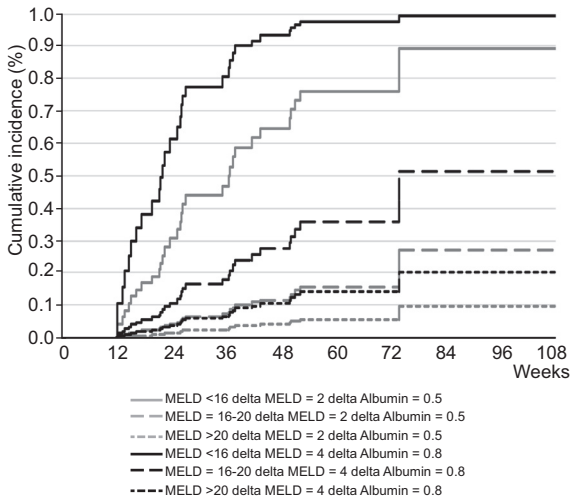

Fig. 5. Estimated cumulative incidence of inactivation based on the 3 variables that emerged from the multivariable model. Baseline MELD, 12-week delta MELD and 12-week delta albumin. Six hypothetical patients are presented.

and 12-week Delta CP (as continuous and two level categorical variables), delta albumin, Delta bilirubin and Delta INR.

All variables with a $p < 0.05$ at univariate analysis were then tested in a multivariable Cox regression models for competing risks analysis and the most parsimonious model with the following three variables was selected: baseline MELD categories (MELD 16–20: HR = 0.120, $p = 0.0005$; MELD >20: HR = 0.042. $p < 0.0001$), continuous 12-week delta MELD (HR = 1.349; $p < 0.0001$) and continuous 12-week delta albumin (HR = 0.307; $p = 0.0069$) (Table 3). These results indicate that MELD and albumin improvement after 12 weeks of treatment are associated with a higher probability of the patient being inactivated; while a higher MELD score at baseline decreases this probability. Therefore, for the same improvement of MELD and albumin, a patient starting with a MELD score lower than 16 has a higher probability of being inactivated compared to a patient starting from a higher MELD score.

To show the combined effects of these variables, we estimated the cumulative incidence of inactivation of 6 hypothetical patients as reported in Fig. 5.

Baseline MELD class stratification, delta MELD and inactivation for clinical improvement

Patients were stratified at baseline into 3 different MELD classes (<16, 16–20 and >20) and the resulting rates of inactivation were reported taking into account the MELD changes after 12 weeks of therapy (Fig. 5). Among patients with MELD score <16, the probability of inactivation was 27.3, 85.7 and 100% in patients with 12-week delta MELD <2, 2 to 4 and >4, respectively. In contrast, in patients with a baseline MELD score ranging from 16 to 20, the probability of inactivation was 10.5% in patients with 12-week delta MELD <2, but 16.7 and 42.9% in patients with 12-week delta MELD of 2–4 or 4. Eventually, among patients with baseline MELD score >20, inactivation was observed only in 2 patients with a 12-week delta MELD >4 (Fig. 6). These 2 patients had a rapid deterioration of liver function 4 and 6 weeks before the start of DAA therapy (acute-on chronic liver failure (ACLF) associated with sepsis).

Relisting of inactivated/delisted patients

One inactivated patient had to be relisted for the occurrence of a small HCC. No patient so far had to be relisted for liver decompensation.

Discussion

All oral DAAs have revolutionized the management of patients with chronic HCV infection and SVR rates higher than 90% can be obtained in patients with compensated cirrhosis [1–6]. However, in patients with decompensated cirrhosis efficacy data of DAAs are still scarce and it is presently unknown whether viral clearance may result in a lasting improvement of signs of liver decompensation. Since preliminary data from phase 3 studies

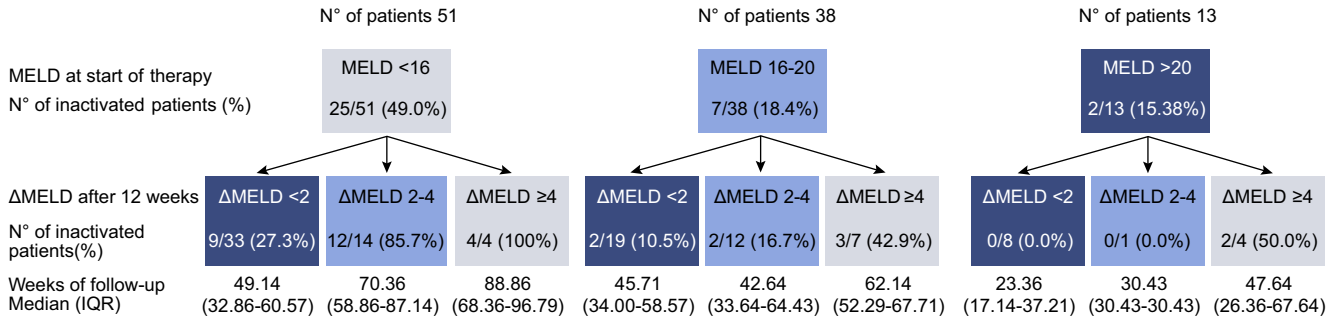

Fig. 6. Inactivated patients stratified by MELD at baseline and Delta MELD after 12 weeks (N = 102). One patient who died before week 12 is not considered in the analysis.

[1,2] show that a meaningful biochemical and clinical improvement can be obtained in some patients with HCV-related decompensated cirrhosis on DAA therapy it is possible that some patients listed for LT due to decompensated cirrhosis can improve their liver function to an extent that some of them can be inactivated from the waiting list and eventually delisted. This would be similar to what was observed in the past [11] when HBV-liver transplant candidates with decompensated cirrhosis were treated with nucleos/tide (NUC). Almost one third of these HBV patients were eventually delisted while on NUC therapy and their clinical improvement could be maintained for up to 5 years. Whether DAA-related clinical improvement may lead to the delisting of some HCV patients is therefore a major and still unanswered question that may have important consequences both at the patient level and also in terms of organ-sparing. Indeed, we are currently facing a "cohort effect" with many listed patients who could not be treated in the past and that are now eligible to be treated with the new DAAs. In Europe the percentage of patients listed for LT due HCV liver diseases, ranges from 25% to 60% and half of them have decompensated cirrhosis [12]. Therefore, pre-LT DAA treatment and associated delisting could significantly reduce the need for LT and save organs.

The aims of the present study were to investigate the magnitude of liver function improvement due to DAA, to determine the percentage of patients who can be inactivated/delisted after DAA treatment and to find possible predictors for inactivation/delisting.

Several important results emerge from this multicentre retrospective European study:

- all oral DAA therapies are highly effective in patients with decompensated cirrhosis listed for LT as more than 90% of treated patients maintained a lasting viral suppression before transplantation, irrespective of the DAA regimen.
- despite the very high virological response obtained in a population which was previously considered as "difficult to treat", improvement of liver function leading to inactivation on the waiting list and eventual delisting, is frequent but not constant. Taking into account the competing risks of LT, dropout due to other causes and death before LT, the 24, 48 and 60 week-probabilities of inactivation from the waiting list were 15.5, 27.6 and 33.3% while the probabilities of delisting were 0%, 10.3% and 19.2% at the same time points. Inactivated patients showed a significantly more pronounced improvement than patients who were not inactivated with a median decrease of MELD score of 3.4 points, which was achieved during the first 12 weeks of therapy. In contrast, the MELD score of patients who were not inactivated remained grossly unchanged (Table 2). The effect of DAA treatment on CP score in inactivated patients was equally relevant with a decrease of 2 points from baseline to week 24, but occurred later than the MELD improvement. This indicates that although DAA effect on biochemical parameters of liver function is rapid, the effect on the two pivotal symptoms of decompensated cirrhosis, namely ascites and encephalopathy, is slower and merits a further period of observation to make the clinician confident enough to propose delisting. Delisting usually followed inactivation by at least 3 months since the usual tendency of the treating clinician was to inactivate patients who improved significantly but not to remove them from the list until a long-term clinical improvement was verified.
- It is worth noting that around one fourth of the patients in this multi-centre cohort had a MELD score at listing lower than 15, which is below the generally accepted threshold of benefit. However, among the 35 patients with MELD <15, 24 had been listed for MELD exceptions and 11 (8.2% of the entire cohort) without a clear MELD exception. It is highly likely that clinicians felt more confident offering DAA therapy to patients with lower MELD scores because these patients would not be too close to LT and would better tolerate the drugs. This is confirmed by the finding that the vast majority of untreated patients during the study period had a much higher MELD score.

Based on multivariate competing risks analysis and simulations, we propose the following strategy taking into account MELD at listing and evolution of MELD and albumin over the first 12 weeks of DAA treatment:

- Patients with a MELD score <16. These patients have a low probability of dying on the waiting list (overall mortality of 2% in this series) or alternatively they may be served by MELD exceptions for their access to transplant. These patients also have a very high probability of achieving an SVR (>80%) and an overall chance to improve clinically and being inactivated in 50% of the cases (Fig. 5). Therefore, DAA treatment can be recommended with the aim of achieving either viral elimination and delisting because of liver function improvement, or transplantation with no risk of HCV recurrence. We propose to inactivate these patients over the first 12 weeks of treatment in order to ensure at least a one month of full viral suppression to prevent HCV recurrence (6) and to evaluate delta MELD and delta albumin on week 12. In case of a relevant MELD and albumin improvement (delta MELD >2 and delta albumin >0.5 g/dl), these patients may subsequently be inactivated for another 12 week period due to a high probability of improvement of ascites and encephalopathy on week 24 (Fig. 3) and eventual delisting. Patients who do not improve under DAAs on week 12, should be re-activated and considered for LT under the MELD exception rule if acceptable.
- Patients with a MELD score between 16 and 20. The chances of being inactivated are around 20% and therefore the decision to treat pre or post LT should be considered only on the individual basis considering the expected waiting time and clinical conditions (slowly progressive *vs.* rapidly progressive disease i.e., acute on chronic liver failure, ACLF) as well as the competing risk of LT. In stable patients, the same initial policy as in patients with baseline MELD <16 can be advised, with subsequent MELD and albumin reassessment on week 12. In patients with MELD improvement >4 and albumin increase >0.5, a further 12-week period of inactivation should be considered because of a reasonable chance of further improvement of ascites and encephalopathy (Fig. 5). Hopefully the early assessment of biochemical changes at 12 weeks should prevent futile transplantation in patients likely to be delisted.
- Patients with a MELD score >20. The only 2 patients that were delisted had developed an ACLF induced by sepsis early before starting DAAs. In addition, for these candidates, there is a significant competing risk of early LT or death both before and after LT. The awareness of these competing risks and cost-effectiveness considerations suggest that DAA treatment should be considered after LT rather than before LT. This final

consideration refers to the possibility of a MELD purgatory effect, which may hamper the access to LT in patients with minimal to mild MELD decrease without a clinically relevant improvement, here again pleading in favour of DAA treatment post-transplant.

There are some limitations to the current study. Firstly, the retrospective design and the lack of clear criteria for delisting at the conception of the study; indeed, the identification of delisting criteria was a secondary end-point of the study. Secondly, some centres did not have the entire spectrum of DAA available. Therefore, the impact of DAA on delisting might even be higher when optimal DAA therapies are used. Thirdly, the follow-up is too short to assess the long-term risk of death, of further deterioration and of development of HCC.

In conclusion, the present study shows that in decompensated cirrhotics, second generation DAAs are very effective and often lead to a remarkable clinical improvement allowing inactivation and eventually delisting in a substantial number of patients. In addition to the individual benefit to the patients, the strategy of inactivating patients with significant MELD and albumin improvement on week 12 may result in saving around 30% of organs dedicated to transplantation of HCV patients, with an important effect on organ shortage, particularly in programs with a high prevalence of HCV liver diseases. A word of caution is to be mentioned regarding how long the clinical improvement will last. We suggest designing long-term multinational observational studies on patients who have been listed for decompensated HCV cirrhosis and subsequently delisted because of clinical improvement. It will be critical to assess the long-term risks of death, further re-deterioration and development of HCC more specifically. These factors still need to be verified.

Conflict of interest

LSB: Received grant/research support from Gilead, AbbVie and BMS as well as contributing to the Gilead consulting/advisory board; MS: Received grant/research support from Gilead and Bayer; PAC: Received a research grant from Gilead; GPP: Has acted as a speaker/advisor for BMS, Gilead, MSD, Janssen, Astellas and Novartis; AC: Received grant/research support from Astellas, Novartis, Gilead, AbbVie, BMS, MSD and Janssen SF: Speaker/Advisor: Gilead, BMS, MSD, AbbVie and Janssen; CD: Received grant/research support from Astellas, Novartis and Chiesi; MB: Received grant/research support from Gilead and acts as a speaker/advisor for MSD, Gilead, BMS, Astellas, Novartis and Janssen.

Author contribution

LSB and CD: conception, design, interpretation of the data and drafting of the article; MB and MS: critical revision for important intellectual content; PAC and RF: analysis and interpretation of data, statistical analysis; SR, SM, CM, FD, RV, JPP, AC, SF, GP, FF, CV: acquisition of data and critical revision of the manuscript; GB, WP and PM: critical revision of the manuscript.

Acknowledgments

Medical writing support was provided by Kerry Flanagan and Clinical Research Assistance by Frederique Franco (13).

Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.jhep.2016.05.010.

References

[1] Manns M, Samuel D, Gane E, Mutimer D, Mc Caughan G, Buti M, et al. Ledipasvir and sofosbuvir plus ribavirin is patients with genotype 1 or 4 hepatitis C virus infection and advanced liver disease: a multicentre, open-label, randomised, phase 2 trial. Lancet Infect Dis 2016 (Epub ahead of print).
[2] Afdhal N, Everson GT, Calleja JL, McCaughan G, Bosch J, Denning J, et al. Effect of Long-Term Viral Suppression With Sofosbuvir + Ribavirin on Hepatic Venous Pressure Gradient in HCV-Infected Patients With Cirrhosis and Portal Hypertension. 2015 International Liver Congress: 50th Annual Meeting of the European Association of the Study of the Liver (EASL) 2015; Abstract LP13.
[3] Foster GR, Irving W, Cheung MC, Walker AJ, Hudson BE, Verma S, et al. Impact of direct antiviral therapy in patients with chronic hepatitis C and decompensated cirrhosis. J Hepatol 2016;64:1224–1231.
[4] Poordad F, Schiff ER, Vierling JM, Landis C, Fontana RJ, Yang R, et al. Daclatasvir, sofosbuvir, and ribavirin combination for HCV patients with advanced cirrhosis or post-transplant recurrence: ALLY-1 study. J Hepatol 2015;62:S261–S262.
[5] Jacobson IM, Poordad F, Firpi-Morell R, Everson GT, Verna EC, Bhanja S, et al. Efficay and safety of grazoprevir and elbasivir in hepatitis C genotype 1 infected patients with CP class B cirrhosis . (C-SALT PART A). 2015 International Liver Congress: 50th Annual Meeting of the European Association of the Study of the Liver (EASL) 2015; Abstract O 08.
[6] Curry MP, Forns X, Chung RT, Terrault NA, Brown Jr R, Fenkel JM, et al. Sofosbuvir and ribavirin prevent recurrence of HCV infection after liver transplantation: an open-label study. Gastroenterology 2015;148:100–107.
[7] Ruiz I, Feray C, Pawlotsky JM, Hezode C. Patient with decompensated hepatitis C virus-related cirrhosis delisted for liver transplantation after successful sofosbuvir-based treatment. Liver Transplant 2015;21:408–409.
[8] Gray R. A class of k-sample tests for comparing the cumulative incidence of a competing risk. Ann Stat 1988;16:1141–1154.
[9] Fine JP, Gray RJ. A proportional hazards model for the subdistribution of a competing risk. J Am Stat Assoc 1999;94:496–509.
[10] Contal C, O'Quigley J. An application of change-point methods in studying the effect of age on survival in breast cancer. Comput stat Data Anal 1999;30:253–270.
[11] Jang JW, Choi JY, Kim YS, Woo HI, Choi SK, Lee CH, et al. Long-term effect of antiviral therapy on disease course after decompensation in patients with hepatitis B virus–related cirrhosis. Hepatology 2015;61:1809–1820.
[12] Burra P, Germani G, Adam R, Karam V, Marzano A, Lampertico P, et al. Liver transplantation for HBV-related cirrhosis in Europe: An ELTR study on evolution and outcomes J Hepatol 2013;58:287–296.