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Predictors of hepatocellular carcinoma in HCV cirrhotic patients treated with direct acting antivirals

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ABSTRACT

Background: Despite the dramatic improvement in viral eradication rates that has been reached with direct antiviral agents (DAAs), the real benefit of viral eradication after DAAs on hepatocellular carcinoma (HCC) development is still controversial.
Aim: To prospectively assess the risk of HCC occurrence and early recurrence in a large cohort of DAA-treated HCV-cirrhotic patients and to identify potential predictors of HCC development.
Methods: We analyzed data prospectively collected from 1927 consecutive HCV-infected cirrhotic patients treated with DAA from January to December 2015 in 10 tertiary liver centers in Italy and followed-up for one year after therapy. 161 patients had a previous HCC.
Results: 38/161 subjects developed tumor recurrence during the follow-up (recurrence rate = 24.8 per 100-year), patients with SVR had a significantly lower rate of recurrence. Lack of SVR and alpha-fetoprotein (AFP) were independent predictors of HCC recurrence. 50/1766 patients without a previous HCC history developed HCC during follow-up (incidence rate = 2.4 per 100-year). Lack of SVR was the strongest predictor of HCC development. Furthermore, patients with SVR and no stigmata of portal hypertension have a lower incidence rate of HCC (1.0 per 100-year).
Conclusions: SVR is associated with a significant decrease of recurrent or de novo HCC. Baseline AFP and signs of portal hypertension can help to stratify the risk of HCC.
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1. Introduction

Patients with hepatitis C virus (HCV) related cirrhosis have an expected high rate of progression to liver decompensation, hepatocellular carcinoma (HCC), and eventually death [1]. Numerous studies, mainly including interferon (IFN) treated patients, have demonstrated that the achievement of sustained virological response (SVR), results in improved clinical outcomes, including lower risk of decompensation and HCC development [2,3].

The treatment of HCV infection has been revolutionized by direct acting antiviral (DAA) agents characterized by high SVR rates and optimal safety profile even in advance stages of disease. Nevertheless, the benefits of viral eradication after DAAs on disease progression, including HCC development, are still under investigation. It is unclear whether DAA induced SVR reduces the risk of HCC in patients with HCV infection. Retrospective observational studies suggested that treatment with DAA might increase the risk of HCC early recurrence [4,5], stimulating the scientific community to investigate the association. A number of reports and meta-analyses did not confirm the initial findings [6–8]. The increased HCC recurrence associated to DAA is still a matter of debate, since available data present several limitations. First, data on clinical outcomes after DAAs therapy refer to a rather short follow-up period; indeed, malignant transformation might occur years before clinical recognition [9]. Second, no randomized clinical trials have been performed and the available data are retrospective, and include heterogeneous groups of patients, tumor characteristics, and tumor treatments.

We herein present a prospective multicentric Italian cohort of almost 2000 HCV cirrhotic patients treated with DAA and followed for one year after SVR. We assessed the risk of HCC occurrence and early recurrence after DAA in a large prospective cohort of DAA-treated HCV-cirrhotic patients; further we looked for factors influencing the risk of HCC.

2. Methods

We herein present a retrospective analysis of a prospective multicenter cohort of HCV-infected cirrhotic patients that were consecutively treated with DAA from January to December 2015 in 10 tertiary liver centers in Italy.

2.1. Clinical assessment and HCV treatment

Patients were treated according to the rules defined by the Italian Medical Agency (AIFA). DAA treatment was decided by the clinicians according to viral genotype and severity of liver disease, following the European Association for the Study of the Liver (EASL) Guidelines and based on the standard of care available in Italy at the time of enrolment.

Diagnosis of cirrhosis was established by the presence of at least one of the following elements: previous liver biopsy with stage 4 fibrosis by METAVIR score (or equivalent Ishak), esophageal and/or gastric varices at endoscopy, liver stiffness measurement (LSM) higher than 12.5 kPa at Transient Elastometry (FibroScan®). Functional class of cirrhosis was attributed by Child–Turcotte–Pugh (CTP) and MELD score. Patients with Metavir F3 and Ishak 3–4, HBV or HIV co-infection, and/or active HCC were excluded. Further, we excluded from the study patients with significant alcohol intake. Importantly patients with active HCC were not allowed to receive DAA treatment according to AIFA rules; therefore, all patient included in the study had a rigorous definition of complete radiologic response (CRR) before treatment. Further, patients on waiting list for Liver Transplant were excluded from the study.

Demographics, clinical characteristics, history of HCC, and comorbidities at baseline were included in a dedicated dataset before starting treatment. Follow-up data of all patients included in the study were recorded starting at the end of DAA treatment and until 12 months after the assessment of SVR or until the date of one of the following evolutionary events: death, liver transplantation or loss to follow-up. Patientś status was established as alive or death at the end of follow up.

During antiviral treatment patients were followed-up monthly for clinical and laboratory evaluation. Virological response to therapy was assessed by quantitative HCV RNA detection and SVR was defined as undetectable serum HCV RNA by sensitive qualitative real-time polymerase chain reaction assay 12 weeks after cessation of therapy. Virological failures and early discontinuation of therapy were also registered.

The protocol was approved by the local Institutional Review Committees, each patient gave written informed consent before enrolment and the protocol was conducted in accordance with the Declaration of Helsinki.

2.2. HCC: radiology and pathology evaluation

Patients with previous HCC were included only after assessment of CRR to treatment. HCC was diagnosed by pathology or by non-invasive criteria according to international guidelines [10,11]. CRR was defined, according to EASL criteria [11], as absence of residual tumor or complete necrosis, assessed by Computed Tomography (CT) Scan or Magnetic Resonance (MR). Per protocol, every single patient with previous HCC had CRR assessed by CT Scan or MR 40 days after HCC treatment, repeated after 3 months, and every 6 months after that. Further, CT Scan or MR was repeated before starting DAA (maximum of 3 months). Nodules with undefined radiological pattern were studied with MRI every 3 months, and HCC excluded after 6 months of stability and lack of evolution (i.e. both dimension or variation of radiological pattern). Previous HCC treatment included ablation, resection or chemoembolization. Patients treated with systemic therapy or patients with extrahepatic lesions were excluded from the study.

Cirrhotic patients with no prior history of HCC underwent ultrasound (US) screening by a specialized dedicated specialist and alpha-fetoprotein (AFP) determination every 6 months, all patients had a recent assessment before stating DAA (maximum time lapse 3 months).

When de novo HCC diagnosis was established, treatment was determined using a multidisciplinary approach according to Barcelona Clinic Liver Cancer (BCLC) schedule and EASL guidelines [11].

2.3. Statistical analysis

Differences in frequencies of characteristics of patients with previous HCC and those without a previous history of HCC at initiation of DAAs were assessed with the Chi-square or the Fisher exact test for categorical variables and with the Student T test for continuous variables. The start point of the analysis in both cohorts was the beginning of DAA treatment.

The average annual recurrence and incidence rates of HCC were calculated dividing the number of events recorded during follow-up by the total number of person-years of observation accumulated during the same period.

The curves showing the cumulative recurrence and incidence of HCC during follow-up were drawn according to the Kaplan–Meier method, and difference between groups was assessed with the log rank test. The number of patients at risk and the proportion of patients who developed an event was reported at different

Table 1
HCC recurrence in 161 cirrhotic patients with a previous history of HCC, according to demographic and clinical characteristics. Median age of the cohort was 65.8 ± 10.0 years. Lack of SVR and AFP above 10 ng/dL are independent predictors of HCC recurrence at the multivariable analysis. Variables that are not reported in the table were not entered in the multivariable model due to collinearity with other variables (i.e. bleeding with varices, diuretics with ascites); class of CHILD and MELD were not entered in the multivariable model as single components were included (i.e. ascites, hepatic encephalopathy, bilirubin, INR, albumin, creatinine).

		Patients	HCC recurrence		Univariate analysis		Multivariable analysis	
		N	N	Rate/100-year	HR (95% CI)	p-Value	HR (95% CI)	p-Value
All patients		161	38	24.8				
Timing of previous HCC[a]	<12 months	58	10	18.0	1.00		1.00	
	≥12 months	100	28	29.8	**2.20 (1.00–4.83)**	**0.05**	2.09 (0.94–4.61)	0.07
Treatment for previous HCC	TACE	21	5	23.0	1.00		–	
	Curative[c]	122	31	26.9	1.32 (0.46–3.76)	0.61	–	
Age at initiation of DAA	<50 years	10	1	12.8	1.00		–	
	≥50 years	151	37	25.4	1.66 (0.23–12.1)	0.62	–	
Sex	Men	111	25	23.7	1.00		–	
	Women	50	13	27.1	1.09 (0.53–2.25)	0.81	–	
BMI	Normal	66	14	22.8	1.00		–	
	Overweight	60	13	20.7	0.93 (0.42–2.05)	0.86	–	
	Obese	14	4	31.9	1.41 (0.46–4.34)	0.55	–	
HCV genotype	HCV1a/1b	114	25	23.7	1.00		–	
	HCV 2	16	5	31.6	1.15 (0.43–3.09)	0.79	–	
	HCV 3	20	5	23.1	0.58 (0.19–1.77)	0.34	–	
	HCV 4	11	3	28.8	1.28 (0.38–4.26)	0.69	–	
Previous antiviral treatment	No	59	12	21.4	1.00		–	
	Yes	97	25	26.7	1.34 (0.66–2.74)	0.42	–	
Varices	None	86	17	18.7	1.00		–	
	F1	56	15	31.5	1.69 (0.81–3.54)	0.16	–	
	F2	14	5	43.0	2.20 (0.81–6.00)	0.12	–	
	F3	4	1	30.7	1.91 (0.25–14.4)	0.53	–	
Ascites	No	131	32	24.8	1.00		–	
	Yes	30	6	24.7	1.03 (0.43–2.47)	0.95	–	
Hepatic encephalopathy	No	154	38	25.4	1.00		–	
	Yes	6	–	–	–	–	–	
Previous bleeding	No	154	37	25.2	1.00		–	
	Yes	6	1	19.9	0.96 (0.13–7.07)	0.97	–	
Drugs	Beta-blocker	40	12	32.7	1.46 (0.72–2.98)	0.29	–	
	Albumin	2	–	–	–	–	–	
	Lattulose	8	1	15.8	0.76 (0.10–5.56)	0.79	–	
	Diuretics	39	6	15.5	0.55 (0.23–1.32)	0.18	–	
	Antibiotics	2	–	–	–	–	–	
Class of child	A	137	35	26.3	1.00		–	
	B/C	22	3	16.4	0.42 (0.10–1.75)	0.23	–	
Diabetes	No	119	26	22.3	1.00		–	
	Yes	38	10	29.4	1.51 (0.72–3.16)	0.27	–	
Stiffness	<25 kPa	45	7	15.2	1.00		–	
	≥25 kPa	53	19	37.3	2.21 (0.92–5.35)	0.08	–	
MELD	<10	110	22	20.6	1.00		–	
	≥10	48	15	34.0	0.84 (0.26–2.76)	0.78	–	
Ribavirin	No	37	8	24.7	1.00		–	
	Yes	122	30	25.2	0.87 (0.39–1.93)	0.72	–	
SVR	Yes	153	34	22.6	1.00		1.00	
	No	8	4	134.2	**8.43 (2.85–24.9)**	**0.0001**	**5.21 (1.75–15.5)**	**0.003**
Creatinine	<1.2 mg/dL	151	34	23.6	1.00		–	
	≥1.2 mg/dL	10	4	42.6	2.01 (0.71–5.72)	0.19	–	
INR	≤1.3	136	30	23.1	1.00		–	
	>1.3	24	8	36.4	1.79 (0.81–3.96)	0.15	–	
Albumin	≥3.6 g/dL	118	29	24.5	1.00		–	
	<3.6 g/dL	43	9	25.7	0.91 (0.40–2.10)	0.83	–	
Bilirubin	≤1.1 g/dL	103	26	26.8	1.00		–	
	>1.1 g/dL	56	12	22.7	0.78 (0.38–1.59)	0.49	–	
Platelets	≥110,000/μL	74	15	21.1	1.00		–	
	<110,000/μL	87	23	28.0	1.18 (0.60–2.29)	0.63	–	
Alpha-fetoprotein[b]	<10 ng/dL	51	3	5.4	1.00		–	
	≥10 ng/dL	57	20	40.2	**6.32 (1.82–21.9)**	**0.004**	**6.19 (1.81–21.2)**	**0.004**

[a] Timing of previous HCC refers to the time between diagnosis of previous HCC and DAA start.
[b] AFP cut-off was chosen based on previous reports and guidelines [9].
[c] Curative treatments include resection, ablation of nodules with diameter <2 cm, and liver transplantation.

time points. Univariate and multivariable Cox proportional hazards regression analysis was used to assess the prognostic significance of various patients and clinical characteristics on the recurrence or development of HCC during follow-up. Data analysis was performed using the SAS software (version 9.2, Cary NC, USA). All tests were two-sided and p-values <0.05 were considered statistically significant.

3. Results

3.1. Baseline features of patients, SVR rates and follow-up

1950 patients initiated DAA therapy between January and December 2015 and were enrolled in the study. Information on previous HCC was not available/recorded for 9 patients and infor-

Table 2
HCC incidence in 1766 cirrhotic patients with no previous history of HCC, according to demographic and clinical characteristics. Median age of the cohort was 61.7 ± 11.0 years. Variables that are not reported in the table were not entered in the multivariable model due to collinearity with other variables (i.e. bleeding with varices, diuretics with ascites).

		Patients	HCC incidence		Univariate analysis		Multivariable analysis	
		N	N	Rate/100-year	HR (95% CI)	p-Value	HR (95% CI)	p-Value
All patients		1766	50	2.4				
Age at initiation of DAA	<50 years	237	2	0.7	1.00		1.00	
	≥50 years	1529	48	2.7	4.00 (0.97–16.5)	0.06	**4.36 (1.04–18.3)**	**0.04**
Sex	Men	1094	31	2.4	1.00		–	
	Women	672	19	2.4	1.05 (0.59–1.86)	0.88	–	
BMI	Normal	723	21	2.5	1.00		–	
	Overweight	595	16	2.2	0.91 (0.47–1.73)	0.76	–	
	Obese	193	5	2.2	0.89 (0.34–2.37)	0.82	–	
HCV genotype	HCV1a/1b	1194	34	2.5	1.00		–	
	HCV 2	200	4	1.6	0.64 (0.23–1.80)	0.39	–	
	HCV 3	218	7	2.5	1.00 (0.44–2.27)	1.00	–	
	HCV 4	154	5	2.8	1.16 (0.45–2.98)	0.76	–	
Previous antiviral treatment	No	683	16	2.0	1.00		–	
	Yes	1046	32	2.6	1.23 (0.67–2.24)	0.51	–	
Varices	None	1049	15	1.2	1.00		1.00	
	F1	511	22	3.8	**3.01 (1.55–5.84)**	**0.001**	**2.25 (1.11–4.53)**	**0.02**
	F2	114	7	5.2	**4.35 (1.77–10.7)**	**0.001**	2.53 (0.96–6.68)	0.06
	F3	37	5	13.0	**11.0 (4.01–30.4)**	**<0.0001**	**4.97 (1.55–16.0)**	**0.007**
Ascites	No	1590	37	2.0	1.00		1.00	
	Yes	176	13	6.5	3.35 (1.74–6.45)	**0.0003**	1.47 (0.72–3.03)	0.29
Hepatic encephalopathy	No	1711	44	2.2	1.00		1.00	
	Yes	55	6	10.6	**4.84 (2.06–11.4)**	**0.0003**	1.64 (0.61–4.38)	0.33
Previous bleeding	No	1713	44	2.2	1.00			
	Yes	50	6	10.6	**4.93 (2.10–11.6)**	**0.0003**	[a]	
Drugs	Beta-blocker	240	16	5.9	**3.20 (1.76–5.81)**	**0.0001**	[a]	
	Albumin	25	5	18.1	**8.13 (3.22–20.5)**	**<0.0001**	[a]	
	Lattulose	63	4	6.0	2.62 (0.94–7.29)	0.06	[a]	
	Diuretics	238	16	5.9	**3.15 (1.74–5.73)**	**0.0002**	[a]	
	Antibiotics	40	1	2.3	0.94 (0.13–6.81)	0.95	–	
Class of child	A	1561	37	2.0	1.00		–	
	B/C	201	13	5.8	**2.99 (1.59–5.65)**	**0.0007**	[a]	
Diabetes	No	1384	36	2.2	1.00		–	
	Yes	348	13	3.3	1.52 (0.81–2.87)	0.20	–	
Stiffness	<25 kPa	990	23	2.0	1.00		–	
	≥25 kPa	500	15	2.5	1.34 (0.69–2.58)	0.39	–	
MELD	<10	1348	28	1.8	1.00		–	
	≥10	388	20	4.5	**2.88 (1.50–5.55)**	**0.002**	[a]	
Ribavirin	No	378	13	3.3	1.00		–	
	Yes	1377	37	2.2	0.66 (0.35–1.24)	0.19	–	
SVR	Yes	87	9	11.3	1.00		**1.00**	
	No	1679	41	2.1	**6.47 (3.14–13.4)**	**<0.0001**	**4.76 (2.19–10.3)**	**<0.0001**
Creatinine	<1.2 mg/dL	1692	48	2.4	1.00		–	
	≥1.2 mg/dL	67	1	1.3	0.57 (0.08–4.14)	0.58	–	
INR	≤1.3	1570	43	2.4	1.00		–	
	>1.3	179	7	3.3	1.46 (0.66–3.25)	0.36	–	
Albumin	≥3.6 g/dL	324	21	5.7	1.00		1.00	
	<3.6 g/dL	1429	29	1.1	**3.40 (1.93–5.98)**	**<0.0001**	1.39 (0.72–2.71)	0.33
Bilirubin	≤1.1 g/dL	1019	22	1.7	1.00		1.00	
	>1.1 g/dL	581	27	4.0	**2.28 (1.29–4.02)**	**0.005**	1.22 (0.65–2.27)	0.54
Platelets	≥110,000/μL	810	37	3.9	1.00		1.00	
	<110,000/μL	947	13	1.2	**3.14 (1.66–5.92)**	**0.004**	1.45 (0.70–3.00)	0.31
Alpha-fetoprotein	<10 ng/ml	617	12	1.6	1.00		1.00	
	≥10 ng/ml	501	21	3.5	**2.13 (1.05–4.34)**	**0.04**	1.86 (0.88–3.94)	0.11

[a] Class of CHILD and MELD were not entered in the multivariable model as single components were included (i.e. ascites, hepatic encephalopathy, bilirubin, INR, albumin, creatinine).

mation on SVR was not available for 14 patients (lost to follow-up). Thus, the current analysis is based on 1927 patients (98.8%) with complete information on both variables (previous HCC and response to DAA treatment). 161 (8,3%) patients had a previous history of HCC, whereas 1766 patients (91,7%) had no previous history of HCC. In order to avoid selection biases, the two cohorts have been clearly separated in the analysis and description.

Tables 1 and 2 show the baseline characteristics of the two cohorts of patients: cirrhotic patients with a previous history of HCC, and patients with no previous HCC respectively. The mean age was 61.7 ± 11.0 in patients with no previous HCC, and 65.8 ± 10.0 years in those with a previously treated HCC. Patients with a story of HCC had a more advanced disease, as suggested by presence of ascites, use of betablockers or diuretics, and LSM ≥ 25 kPa. HCC characteristics of the two cohorts are summarized in Table 3.

Of the whole cohort 1832 patients (95.1%) achieved SVR, whereas 95 patients (4.1%) experienced a virological failure. No difference in SVR rates was observed regardless the previous history of HCC [12]. The SVR rate was 95.0% (153/161) in patients with previous history of HCC, and 95.1% (1679/1766) in patients with no previous HCC. During the follow-up 20 patients died of liver related causes (only one HCC related), 16 patients died of non-liver-related causes.

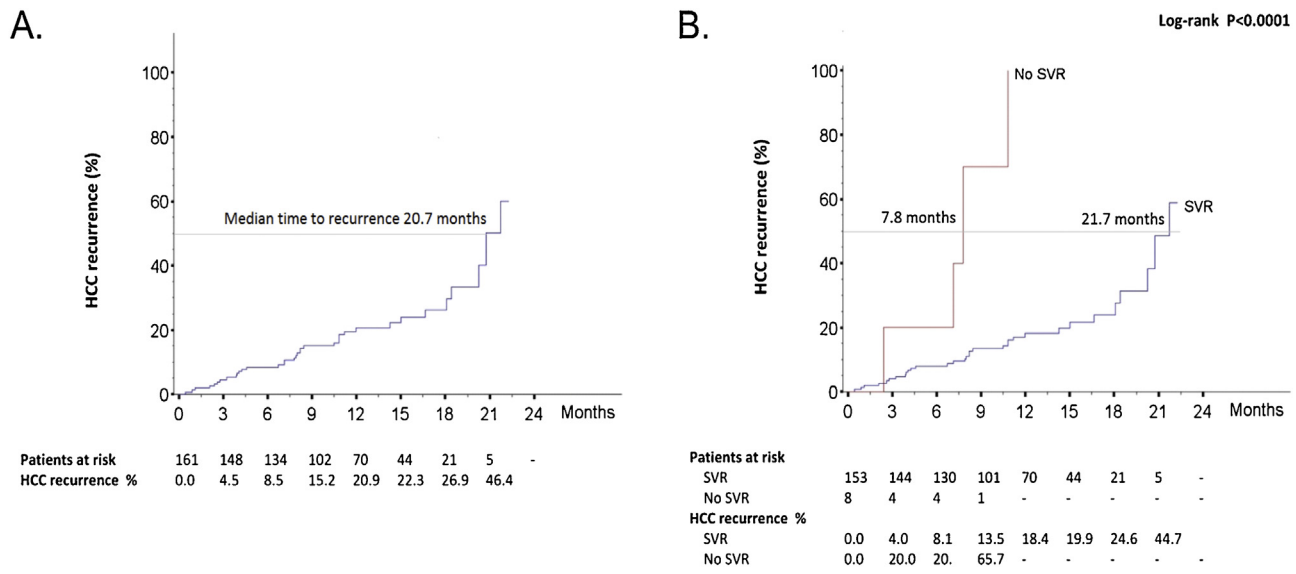

Fig. 1. HCC recurrence after DAA therapy in patients with HCV-related cirrhosis and previous history of HCC (n = 161). Recurrence rates are shown for all patients (A) and according to SVR status (B). Median time to HCC recurrence is indicated in the figure.

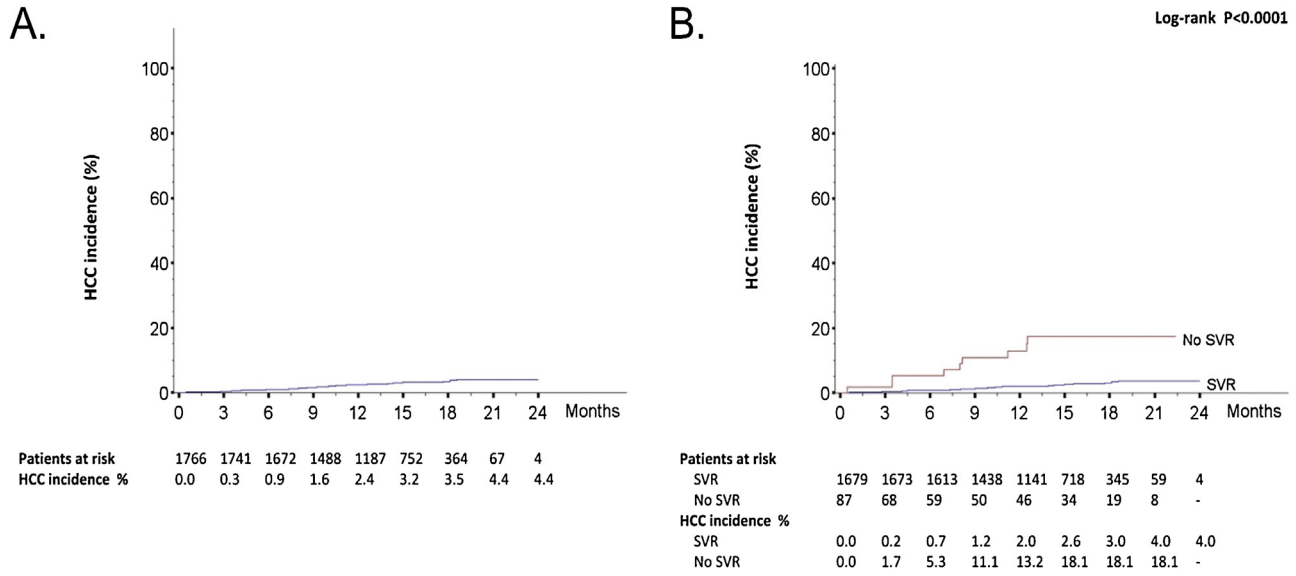

Fig. 2. HCC incidence after initiation of DAA therapy in cancer free patients with HCV-related cirrhosis (n = 1766). Occurence rates are shown for all patients (A) and according to SVR status (B). Median time to HCC recurrence is indicated in the figure.

3.2. HCC recurrence: incidence and predictors

38 of the 161 patients with previous history of HCC developed radiologic tumor recurrence during follow-up, with an average annual incidence rate of 24.8 per 100-year. Patients with SVR (n = 153,34 events) had an average annual incidence rate of 22.6 per 100-year; whereas patients with no virological response (n = 8,4 events) had an average annual incidence rate of 134.2 per 100-year. The 6-, 12-, and 18-month recurrence rates in patients with prior history of HCC were 8.5%, 20.9%, and 26.9% respectively (Fig. 1A). Patients who achieved SVR had a significantly lower rate of HCC recurrence than non SVR patients at all time points (log-rank $p < 0.0001$) (Fig. 1B). However, the number of patients with no SVR and HCC recurrence is low since only 8 patients were at risk.

The Cox regression analysis highlighted lack of SVR (HR 5.21; 95% CI: 1.75–15.5; $p = 0.003$) and AFP $\geq$ 10 ng/dL (HR 6.19; 95% CI: 1.81–21.2; $p = 0.004$) as independent predictors of HCC recurrence at the multivariable analysis (Table 1). We did investigate the influence of elements that could affect HCC recurrence (Table 1). In particular, we did not see significant association in our cohort of HCC recurrence with the timing of DAA therapy after prior HCC response nor the prior HCC therapy (curative versus TACE).

3.3. HCC incidence and predictors

1766 patients included in the study had no history of HCC before DAA treatment; 50 of them (2.8%) developed HCC during follow-up with an average annual incidence of 2.4 per 100-year. The 6-, 12-, and 18-month incidence rates in patients without prior history of HCC were 0.9%, 2.4%, and 3.5% respectively (Fig. 2.A). Similarly to patients with previous history of HCC, patients who achieved a SVR had a significantly lower rate of HCC occurrence than non-SVR patients at all time points ($p < 0.0001$) (Fig. 2B).

Lack of SVR was confirmed as the strongest predictor of HCC incidence from the univariate (HR 6.47; 95% CI: 3.14–13.4; $p < 0.0001$) and multivariable analysis (HR 4.76; 95% CI: 2.19–10.3; $p < 0.0001$)

Table 3
HCC characteristics of recurrent and "de novo" HCC in patients treated with DAA.

	HCC recurrence	HCC incidence
Total number of HCC	38/161	50/1766
Monofocal	25 (65.8%)	36 (72.0%)
Multifocal	13 (34.2%)	11 (22.0%)
Infiltrant	–	3 (6.0%)
Number of lesions		
1	25 (65.8%)	36 (72.0%)
2	5 (13.1%)	9 (18.0%)
3 or more	8 (21.1%)	5 (10.0%)
Dimension		
<20 mm	24 (63.1%)	26 (52.0%)
≥20 mm	14 (36.8%)	24 (48.0%)
Milano criteria		
Yes	25 (65.8%)	36 (72.0%)
No	13 (34.2%)	14 (28.0%)
BCLC		
A–0	25 (65.8%)	36 (72.0%)
B	8 (21.1%)	4 (8.0%)
C	5 (13.1%)	6 (12.0%)

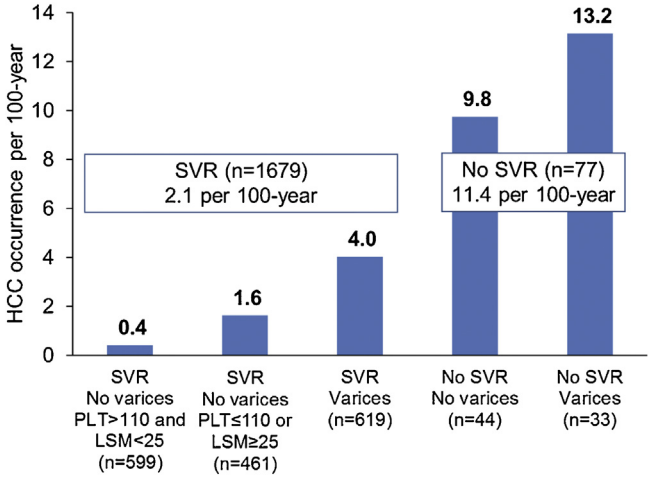

Fig. 3. Rate of HCC occurrence (per 100-year) according to SVR status and history of esophageal varices.

(Table 2). Patients older than 50 years of age and patients with presence of esophageal varices had a significantly higher risk of developing HCC at the multivariable analysis (Table 2).

The rate of HCC occurrence markedly increased combining the risk factors identified at the multivariable analysis (i.e. SVR status and history of esophageal varices) (Table 4A). Patients with SVR and no stigmatae of portal hypertension have an incidence rate of HCC of 1.0 per 100-year, whereas patients with no SVR and endoscopic varices have a 13 times higher incidence rate (Fig. 3). Furthermore, using the recently proposed expanded Baveno VI Criteria [13] we were able to identify a subgroup of 620 SVR patients (with both platelets >110,000/μL and LSM < 25 kPa) in whom the incident rate of HCC was extremely low (0.5 per 100-year) (Table 4B).

Post-treatment AFP > 10 ng/mL seems to be associated to a higher de novo HCC risk, however it does not reach significance in our cohort (Supplementary Table).

3.4. HCC characteristics

During follow-up, HCC recurred in 38/161 (23.6%) patients. At the time of HCC recurrence 19/38 (50%) patients had a single lesion, 24 (63.1%) were within Milano criteria, and 20 (52.6%) were in early stage according to BCLC. No patient developed extra-hepatic metastases.

Only 50/1766 subjects developed a new HCC during follow-up after DAA treatment; 34 (68.0%) of the diagnosed lesions were single, 42 (84.0%) patients developed an HCC within Milano criteria, and 32 (64.0%) of them were in early stage according to BCLC (Table 3).

4. Discussion

According to the World Health Organization, HCC is the fifth most common tumor worldwide and the third most common cause of cancer-related death [14]. The availability of effective and safe DAAs has finally allowed HCV eradication in patients once considered difficult to treat, including those with advance liver disease and prior HCC. However, the impact of HCV eradication by DAA therapies on the risk of HCC have been questioned, and contradictory data has emerged. We herein present a prospective multicentric Italian cohort of almost 2000 HCV cirrhotic patients treated with DAA, in which we confirm previous data showing a significant reduction in HCC incidence and recurrence in patients who achieved an SVR. Further, we identify baseline AFP and signs of portal hypertension as independent predictors that can help to stratify the risk of HCC and therefore guide patient management.

Since the initial availability of DAA, it seemed reasonable to expect that HCV eradication would reduce HCC risk; advanced cirrhotic patients, including those with previously treated HCC, were therefore considered ideal candidates for DAA therapy. The enthusiasm for DAAs was clouded by reports suggesting that treatment with DAAs could increase the risk of early HCC recurrence [4,5,15] and de novo HCC [16]. A retrospective Spanish study reported an unexpectedly high rate of HCC recurrence after antiviral treatment with DAAs in a small number of patients with previously treated HCC [4]. An Italian study confirmed the Spanish report and described an early HCC recurrence in 17 out of 59 (28.8%) patients after DAA [5]. Several larger studies from different geographical regions did not support the finding [6,17–20]. Two large studies at Veterans Affairs described a substantial decline of both all-cause mortality and HCC rates in a cohort of more than 50,000 patients with advanced HCV and relevant co-morbidities who were followed for two years after a successful response to DAA [21,22]. Finally, a large systematic review, meta-analyses, and meta-regression study comparing the rate of HCC occurrence in patients with HCV-related cirrhosis was recently published [7]. The authors included a total of 13,875 patients from 41 heterogeneous studies and aimed to compare the rate of HCC in patients who received HCV treatment, including both DAA versus IFN. However, the results might reflect the large heterogeneity of the population and the metaregression shows a weak I-squared. Despite the limits of the methodology itself and the extreme heterogeneity across analysed studies, the authors concluded that there was no evidence for differential HCC occurrence nor recurrence risk following SVR from DAA and IFN-based therapy [7,23].

Although the initial concerns of a correlation between HCC development and DAAs seem to have waned out, to date there are no prospective randomized studies that could prove with no doubt that DAA therapy does not increase the HCC risk [24]. We herein present a prospective collection of data from almost 2000 cirrhotic patients treated with DAA in a real-life setting, with all patients undergoing HCC surveillance through liver ultrasound, and demonstrate that lack of SVR and AFP are independent predictors of HCC recurrence, and that lack of SVR and portal hypertension are the strongest predictors of HCC incidence after DAA. Although this is a confirmatory study, the key novelty of our study lays on the risk stratification based on both DAA response and clinically significant

Table 4
Rate of HCC incidence (per 100-year) according to risk factors in patients without previous HCC treated with DAA.

A.

	Patients	Person-years	HCC	Rate per 100-year
Treated patients (total)	1766	2057	50	2.4
Patients with SVR	1679	1978	41	2.1
SVR and no varices	1060	1260	12	1.0
SVR with varices	619	718	29	4.0
Patients with no SVR	87	79	9	11.3
No SVR and no varices	44	41	4	9.8
No SVR with varices	33	38	5	13.2
Patients with SVR				
Normal albumin and platelets	934	1091	11	1.0
Either low albumin or low platelets (<110,000)	615	728	22	3.0
Both low albumin and low platelets (<110,000)	225	269	13	4.8
Patients with no SVR				
Normal albumin and platelets	31	25	2	8.0
Either low albumin or low platelets (<110,000)	33	30	4	13.3
Both low albumin and low platelets (<110,000)	29	30	3	10.0

B.

	Patients	Person-years	HCC	Rate per 100-year
Treated patients (total)	1766	2057	50	2.4
Platelets >110 and LSM < 25 kPa	637	742	5	0.7
Platelets $\leq$110 OR LSM $\geq$ 25 kPa	844	1002	33	3.3
Patients with SVR				
Platelets >110 and LSM < 25 kPa	620	729	4	0.5
Patients with no SVR				
Platelets >110 and LSM < 25 kPa	17	13.7	1	7.3

portal hypertension. Patients within the "Extended Baveno Criteria" (i.e. Platelets >110,000/μL and LSM < 25 kPa), had a very low probability of developing HCC and could be therefore candidate to a different surveillance program, whereas patients with oesophageal varices and lack of SVR might be candidate to a stricter surveillance.

Our results confirm that an SVR achieved with DAA based regimens is strongly associated to a reduction in the risk of developing both de novo or recurrent HCC, as demonstrated in the Veterans cohort [25], and another large Italian cohort [26]. These findings mimic what has been reported by several studies conducted in the IFN era [27,28], although those studies included only patients that were deemed fit to receive IFN and thus had well compensated cirrhosis with moderate portal hypertension. Our current study expands on those findings as it included also CTP B and C patients (11.5%) as well as those with previous episodes of decompensation (10.6%) or with significant portal hypertension defined as medium-large varices or primary prophylaxis with beta-blockers (14.5%). It was indeed recently demonstrated, that hepatic decompensation is the major driver of death in HCV cirrhotic patients with successfully treated early HCC [29].

Importantly, the absolute risk of HCC remains high in patients with cirrhosis and thus they should be enrolled in a long-term HCC surveillance program, according to International Guidelines [10,11]. Currently, international guidelines suggest surveillance using US, with or without AFP, every 6 months since there is not enough evidence to determine which type of surveillance test—US alone or the combination of US plus AFP—leads to a greater improvement in survival [10]. Importantly, our study demonstrates that baseline AFP values are a strong independent predictor of HCC recurrence in DAA treated patients, but not for de novo HCCs; these different risk profiles might be influenced by the low number of incident tumors in our cohort. However, the main reason for this finding is still largely unknown and further studies are required.

Lastly the presence of any grade of esophageal varices detected at baseline by esophagogastroduodenoscopy (EGDS) is associated with a greater risk of HCC development. This finding, in theory, would call for universal EGDS screening of all cirrhotic patients, a statement that clashes with the current recommendation to stratify patients on the basis of liver stiffness values and platelet count to identify those who should receive EGDS. This recommendation stems from the fact that cirrhotic patients with LSM < 25 KPa and platelet count >100,000, very rarely have varices that are at risk of bleeding (F2–F3). For this reason, we analyzed patients in our cohort following the recently proposed "Extended Baveno Criteria" (i.e. both platelets >110,000/μL and LSM < 25 kPa) and found that the HCC risk in those patients who would not undergo EGDS is extremely low, thus identifying a promising tool for cirrhotic SVR patient stratification [13].

While some authors have reported a more aggressive patter of HCC in patients treated with DAA [4,30], our results do not seem to confirm that suggestion. However, recurrent tumors in our cohort seem to be more aggressive than "de novo" HCC. Indeed, 63.1% of the recurrent tumors were within Milano criteria compared to 84% of patients with a newly developed HCC; further 52.6% of them were in early stage according to BCLC. No patient developed extrahepatic metastases.

Our study has, however, some limitations. First, even though only tertiary referral centers were included in the study and only CT/MR were used, there was no centralized assessment of HCC diagnosis nor response to treatment. Second, tumor characteristics and tumor treatments were not homogeneous and the time frames between tumor cure and DAA therapy was not standardized. Finally, our study has not a parallel control group of untreated patients. However, the rates of HCC occurrence herein reported are not superior to those observed in the past in the same geographical area in untreated patients with compensated cirrhosis [31].

In conclusion, based on our data, we strongly recommend antiviral treatment in cirrhotic patients with or without previously eradicated HCC. We believe that studies are needed in order to define the timing of DAA treatment after HCC eradication. We further strengthen the use of AFP in association to US surveillance. Finally, we believe our data might help to stratify HCC surveillance program in cirrhotic patients; non-responders to DAA deserve a stricter surveillance, whereas patients within the "Extended Baveno Criteria" could be candidate to a less frequent follow-up.

Conflict of interest

Prof. Marcello Persico has served as a speaker, a consultant and an advisory board member for Abbvie, BMS, Gilead, Merck/MSD and has received research funding from Abbvie and Gilead. Prof. Savino Bruno has served as a speaker, a consultant and an advisory board member for Abbvie, BMS, Gilead, Merck/MSD. Prof. Pietro Lampertico has served as a speaker, a consultant and an advisory board member for Bristol-Myers Squibb, Roche, Gilead, GlaxoSmithKline, MSD, Abbvie, Janssen, Arrowhead, Arbutus. Prof. Erica Villa has served as a speaker, a consultant and an advisory board member for Abbvie, MSD, Gilead and Roche and has received research funding from BMS, Abbvie and MSD. Prof. Vito Di Marco has served as a speaker, a consultant and an advisory board member for Abbvie, BMS, Gilead, Merck/MSD and has received research funding from Abbvie, BMS, Gilead, Merck. Prof. Pietro Andreone has served as a speaker, a consultant and an advisory board member for Abbvie, BMS, Gilead, INTERCEPT and has received research funding from MSD, Abbvie, Gilead Sciences, BMS. Prof. Alessio Aghemo has served as a speaker, a consultant and an advisory board member for Abbvie, BMS, Gilead, MSD, alfasigma and has received research funding from Abbvie, Gilead. Prof. Alfredo de Leo has served as speaker, consultant and an advisory board member for Abbvie, Gilead, BMS, MSD. Prof. Massimo Zuin has served as speaker and an advisory board member for Abbvie, BMS, Gilead. Dr. Vincenza Calvaruso has received research funding from Merck/MSD; invited speaker for Abbvie, BMS, Intercept, and participated in advisory boards for Abbvie, Gilead. Dr. Alessia Giorgini has served as speaker and an advisory board member for Abbvie, BMS, Gilead. Dr. Ana Lleo has served as a speaker for Abbvie, BMS, Gilead, and Intercept. Dr Maria Rendina has served as speaker, consultant and an advisory board member for Abbvie, Gilead, BMS, MSD. Dr. Vincenzo Boccaccio has served as a speaker for Abbvie, BMS, and Gilead. Prof. Patrick Maisonneuve, Dr. Andrea Aglitti, Dr. Barbara Coco, Dr. Giulia Troshina, Dr. Fabio Conti, Dr. Luca Marzi and Dr. Simona Bollani have no disclosures to declare.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.dld.2018.10.014.

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