Received: 26 July 2016 | Accepted: 30 December 2016

DOI: 10.1111/liv.13357

VIRAL HEPATITIS

WILEY Liver INTERNATIONAL

A meta-analysis of single HCV-untreated arm of studies evaluating outcomes after curative treatments of HCV-related hepatocellular carcinoma

Giuseppe Cabibbo[1] | Salvatore Petta[1] | Marco Barbàra[1] | Gabriele Missale[2] | Roberto Virdone[3] | Eugenio Caturelli[4] | Fabio Piscaglia[5] | Filomena Morisco[6] | Antonio Colecchia[7] | Fabio Farinati[8] | Edoardo Giannini[9] | Franco Trevisani[10] | Antonio Craxì[1] | Massimo Colombo[11] | Calogero Cammà[1] | on behalf of the ITA.LI.CA study group[a]

[1]Section of Gastroenterology, DI.BI.M.I.S., Palermo, Italy

[2]Unit of Infectious Diseases and Hepatology , Teaching Hospital-University of Parma, Italy

[3]Division of Internal Medicine 2, Ospedali Riuniti Villa Sofia-Cervello, Palermo, Italy

[4]Division of Gastroenterology, Belcolle Hospital, Viterbo, Italy

[5]Division of Internal Medicine, Department of Medical and Surgical Sciences, Alma Mater Studiorum, Bologna, Italy

[6]Division of Gastroenterology, Department of Medicine and Surgery, University of Naples Federico II, Italy

[7]Department of Medical and Surgical Sciences, University of Bologna, Italy

[8]Department of Surgery, Oncology and Gastroenterology, University of Padua, Italy

[9]Gastroenterology Unit, Department of Internal Medicine, IRCCS Azienda Ospedaliera Universitaria San Martino IST, Genoa, Italy

[10]Division of Semeiotics, Department of Medical and Surgical Sciences, Alma Mater Studiorum, Bologna, Italy

[11]Humanitas Clinical and Research Center, Rozzano, Italy

Correspondence

Calogero Cammà, Sezione di Gastroenterologia, DIBIMIS, University of Palermo, Palermo, Italy.
Email: calogero.camma@unipa.it

Handling Editor: Morris Sherman

Abstract

Background & Aims: Determining risk for recurrence or survival after curative resection or ablation in patients with hepatitis C virus (HCV)-related hepatocellular carcinoma (HCC) is important for stratifying patients according to expected outcomes in future studies of adjuvant therapy in the era of direct-acting antivirals (DAAs). The aims of this meta-analysis were to estimate the recurrence and survival probabilities of HCV-related early HCC following complete response after potentially curative treatment and to identify predictors of recurrence and survival.

Methods: Studies reporting time-dependent outcomes (HCC recurrence or death) after potentially curative treatment of HCV-related early HCC were identified in MEDLINE through May 2016. Data on patient populations and outcomes were extracted from each study by three independent observers and combined using a distribution-free summary survival curve. Primary outcomes were actuarial probabilities of recurrence and survival.

Abbreviations: BCLC, Barcelona Clinic Liver Cancer; DAA, direct acting antiviral; HCC, hepatocellular carcinoma; RCT, randomized controlled trial.

[a]Additional members of the ITA.LI.CA groups are listed in the Appendix.

Results: Eleven studies met the inclusion criteria. Pooled estimates of actuarial recurrence rates were 7.4% at 6 months and 47.0% at 2 years. Pooled estimates of actuarial survival rates were 79.8% at 3 years and 58.6% at 5 years. Heterogeneity among studies was highly significant for all outcomes. By univariate meta-regression analyses, lower serum albumin, randomized controlled trial study design and follow-up were independently associated with higher recurrence risk, whereas tumour size and alpha-foetoprotein levels were associated with higher mortality.

Conclusions: This meta-analysis showed that recurrence risk and survival are extremely variable in patients with successfully treated HCV-related HCC, providing a useful benchmark for indirect comparisons of the benefits of DAAs and for a correct design of randomized controlled trials in the adjuvant setting.

KEYWORDS

hepatocellular carcinoma, prognosis, recurrences, survival

1 | INTRODUCTION

Hepatocellular carcinoma (HCC) is the second-most-prevalent global cause of cancer-related death and the leading cause of mortality in patients with cirrhosis.[1,2] Cirrhosis is the strongest risk factor for HCC, with hepatitis C virus (HCV) being a major risk factor in the Western world and Japan. In patients diagnosed at an early disease stage (Barcelona Clinic Liver Cancer Stage 0 or A - BCLC 0/A), surgical resection and local ablation are potentially curative treatments, with 5-year overall survival (OS) rates of 60-80% (surgical resection) and 40-70% (loco-regional ablation).[3-5] Unfortunately, tumour recurrence and decompensation of underlying cirrhosis contribute to long-term mortality after curative treatment of early HCC.[3-6] In particular, tumour recurrence remains high and heterogeneous when assessed in studies performed in the adjuvant setting.

To date, there is no standard-of-care for adjuvant therapy because no treatment has a clearly proven benefit in terms of HCC recurrence and survival after potentially curative treatment of HCC.[4,5] The STORM trial[7] of sorafenib as an adjuvant treatment after potentially curative therapy for early HCC showed no significant treatment effect for HCC recurrence or OS. Several previous meta-analyses,[8-11] performed on published data from small trials in heterogeneous populations of patients with HCV-related, successfully treated, early HCC, showed conflicting results of the benefits of interferon-(IFN) based regimens on HCC recurrence and OS. However, the lack of availability of individual data limited the possibility of analysing recurrence and survival as time-dependent variables.

Although patient outlook may improve with the increased availability of safe and effective direct-acting antivirals (DAAs), the current standard-of-care for HCV,[12] the long-term effect of viral eradication on HCC outcomes has yet to be established. No randomized controlled trials (RCTs) testing DAAs have been conducted in patients with HCV-related early HCC. Because of the clinical breakthrough because of the effect of DAAs, it will not be feasible to design future RCTs with an untreated control arm in this population.

Key points

- Cirrhosis is the strongest risk factor for hepatocellular carcinoma (HCC), with HCV being the major risk factor in the Western world and Japan.
- HCC recurrence contributes to long-term mortality after curative resection or ablation.
- An unexpectedly high rate of HCC recurrence after treatment with direct-acting antivirals (DAAs) was recently reported; however, no appropriate comparison of HCV-untreated controls exists.
- Pooled probabilities of recurrence and survival after potentially curative treatment of HCC in HCV-untreated patients provide a useful benchmark for comparisons of the benefits of HCV eradication by DAAs and correct design of trials of new adjuvant drugs.

Recently, Reig et al.[13] reported a single-centre small study evaluating the benefits of DAA-based therapy on HCV-related early HCC that achieved a complete response. They observed an unexpected and surprisingly high rate of early recurrence of HCC, raising concern about the benefits of DAA-based therapy in the adjuvant setting.

There is a great deal of heterogeneity among patients with HCV-related early HCC who achieve complete response after curative treatment and do not receive adjuvant treatment. Therefore, accurate estimates of recurrence and OS rates are essential for evaluating natural history, assessing treatment effect size, calculating sample size and interpreting results of clinical trials. Knowledge of factors that influence outcomes among patients not receiving adjuvant treatment may be important for designing future clinical trials.

To resolve uncertainty by increasing statistical power, we performed a meta-analysis of single HCV-untreated arm studies of patients with HCV-related early HCC who did not receive any adjuvant

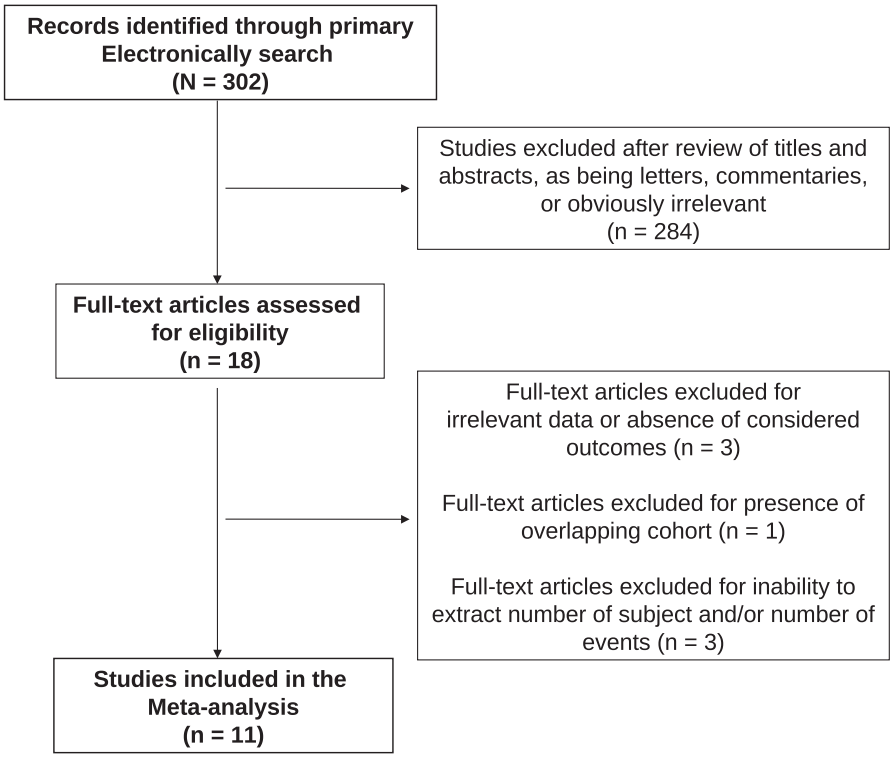

FIGURE 1 Study flow-chart

treatment after successful treatment. Our aims were as follows: (i) to estimate the pooled actuarial probabilities of recurrence and OS among patients who did not receive any adjuvant treatment; (ii) to analyse variabilities in recurrence risk and OS by considering the heterogeneity among studies; and (iii) to identify factors associated with the risk of recurrence and survival.

2 | MATERIALS AND METHODS

2.1 | Selection of trials

This meta-analysis was performed in accordance with the PRISMA statement (see Table S1).[14] The primary sources of the reviewed studies were MEDLINE, CANCERLIT, the Cochrane Controlled Trials Register and the Cochrane Library, with the following medical subject headings (MeSH): *hepatocellular carcinoma*; *liver cancer*, *primary liver carcinoma*; *therapy*; *treatment*; *adjuvant treatment*, *interferon*, *prevention*, *hepatitis C*, *recurrence*, *survival*; *randomized* or *non-randomized trial*, *clinical trial and cohort study*. The search included literature published through May 2016 with no lower date limit. To identify additional studies, the computer search was supplemented with manual searches of the reference lists of all retrieved review articles and primary studies. When the results of a single study were reported in more than one publication, only the most recent and complete data were included in the meta-analysis.

Studies were included in the analysis if: (i) they included patients with HCV-related early HCC who achieved complete response after surgical resection or ablation; (ii) they were RCTs or non-RCTs (nRCTs) comparing IFN-based therapy with no treatment; (iii) they were cohort studies including patients who did not receive any adjuvant treatment; (iv) they assessed HCC recurrence and OS as time-dependent outcomes; and (v) they were full-length papers.

Among the 302 studies reviewed, the inclusion criteria were met by 11 studies: four RCTs,[15-18] four nRCTs,[19-22] and three cohort studies[23-25] (Figure 1). Studies were excluded if they considered HCC patients with an etiology other than HCV or if they were published only in abstract form (because methodological quality could not be assessed).

2.2 | Review of studies

Several study- and patient-level variables were extracted from all eligible studies and entered into a structured database. Study-level variables included the last name of the first author, publication year, region where the study was conducted, study design, number of subjects, number of centres (single vs multiple), outcomes measured and study validity. Patient-level variables included mean age, sex, number of HCC nodules, mean size of the main tumour, percentage of patients who underwent surgical resection, serum levels of albumin and bilirubin, number of platelets, percentage of Child-Pugh class A patients and alpha-foetoprotein (AFP) levels. Each trial was evaluated and classified by three independent investigators (G.C., C.C., S.P.). Discrepancies among reviewers were not frequent (interobserver variation <10%) and were resolved by discussion.

2.3 | Assessment of study quality

All reports were assessed for study quality. Conflict between reviewers was resolved by consensus. Studies were graded using the following parameters: (i) study design: RCT, nRCT, or cohort study; (ii)

TABLE 1 Study- and patient level characteristic of the studies in the meta-analysis

First author (year)	Study type	Sample	Centres	Regions[a]	Median F-Up	Quality score	Mean age	Male gender (%)
Ikeda (2000)[15]	RCT	10	1	1	25	4	65	60
Suou (2001)[19]	nRCT	22	1	1	48	3	62	81
Shiratori (2003)[16]	RCT	25	1	1	85	6	63	68
Hung (2005)[20]	nRCT	40	1	1	NA	2	63	75
Nishiguchi (2005)[17]	RCT	15	1	1	54	4	60	100
Yamanaka (2005)[23]	cohort	26	1	1	NA	2	68	73
Mazzaferro (2006)[18]	RCT	38	4	2	45	7	67	68.4
Kudo (2007)[21]	nRCT	84	1	1	NA	3	66	71
Jeong (2007)[22]	nRCT	42	1	1	32	3	63	69
Kanogawa (2014)[24]	cohort	82	4	1	47	3	66	77
Petta (2016)[25]	cohort	328	24	2	33	4	69	66

RCT, randomized controlled trial; nRCT, non-randomized controlled trial; cohort, cohort study; F-Up, follow-up; HCC, hepatocellular carcinoma; C-P, Child-Pugh; AFP, alpha-foetoprotein.

[a]1 corresponds to Asia-Pacific studies; 2 to North American and European studies.

study type: prospective or retrospective; (iii) consecutive inclusion of patients: yes or no; (iv) percentage of dropout <10%; (v) adequately defined complete response after curative treatment; (vi) follow-up >3 years; and (vii) adequately defined protocol for recurrence surveillance. Each parameter was given a numerical score of 0 or 1. Studies with a score >5 were classified as good quality; otherwise, studies were rated as poor quality (Table S2).

2.4 | Statistical analyses

Crude rates of 6-month and 2-year recurrence and 3-year OS were extracted as outcome measures. Pooled estimates of 6-month and 2-year recurrence and 3-year OS rates were calculated using random-effects logistic regression analysis after applying sample weights according to the sample size. Heterogeneity among studies was assessed by the Pearson χ^2 test and the I^2 statistic.

Only univariate logistic meta-regression analyses were used to examine associations between patient- or study-level covariates and 2-year HCC recurrence and 3-year OS rates. We did not consider multivariate meta-regression analysis because we lacked complete data for identification of candidate variables that could explain heterogeneity. Begg's funnel plots were generated and Egger's regression asymmetry test was used to examine potential publication bias related to 2-year recurrence and 3-year OS rates respectively.

In clinical trials with a time-dependent outcome (death or disease recurrence), survival curves were used to describe the risk of the event over time. In meta-analyses of studies reporting a survival curve, the most informative finding was a summary survival curve. We used the nonparametric approach reported by Combescure et al.[26] to assess pooled survival probabilities from several single-arm studies. This approach is a version for aggregated data of the product-limit estimator of survival, and uses random effects to model between-study heterogeneity. The between-study covariance matrix was estimated using the multivariate extension of DerSimonian and Laird's method.[27,28]

Compared to meta-analyses of survival probabilities at a single time point,[29] this approach has several advantages. First, the estimation of the pooled survival probability at time *t* also involves all studies ending before *t* because these studies contribute to the estimated conditional survival probabilities for time intervals prior to *t*. Second, this approach does not require assumptions about the shape of survival curves. Finally, the pooled survival probabilities are guaranteed not to increase over time.

For all analyses, a p value <.05 was considered statistically significant. All analyses and graphics were completed with the R Statistical Computing Environment (R Foundation for Statistical Computing, Vienna, Austria).

3 | RESULTS

3.1 | Description of studies

Eleven studies,[15-25] all published since 2000 and including four RCTs,[15-18] four nRCTs,[19-23] and three cohort studies,[24-26] fulfilled the inclusion criteria and were selected for review. Nine studies[15-17,19-24] were Asian-Pacific, whereas two[18,25] were North American and European. All but one trial[20] reported recurrence curves. Nine reported curves on survival.[16-22,24,25]

Table 1 reports the distributions of the main study- and patient-level characteristics of the single arms of the 11 studies.[15-26] A total of 701 patients were included in the untreated arms of these 11 studies. The size of the single arm in each study ranged from 10[15] to 328[25] patients. The percentage of men ranged from 60%[15] to 100%.[17] Mean patient age was 64 years, ranging from 60 years[17] to 69 years.[25] Eight studies[15,16,18,20-23,25] reported the percentage of patients with a single

Solitary HCC (%)	Size of main Tumour (mm)	Surgical resection (%)	Albumin (gr/dL)	Bilirubin (mg/dL)	Platelet ($\times 10^9$/L)	C-P class A (%)	AFP (ng/mL)
90	20	80	3.2	0.8	109	NA	NA
NA	21	63	NA	NA	NA	NA	NA
64	23	0	3.4	0.8	97	NA	23
60	26	0	NA	NA	123	82	189
NA	26	100	3.6	NA	112	80	NA
50	NA	0	NA	NA	NA	58	NA
76.3	30	100	4	1	141	92	21
92	15	0	3.6	0.7	105	NA	12
85	15	22	3.9	NA	115	83	31
NA	NA	23	NA	NA	NA	100	11
86	2.2	27	NA	1	123	94	NA

HCC nodule, which ranged from 60%[20] to 90%.[15] All but two studies[23,24] reported data on the size of the main tumour, which ranged from 15 mm[21] to 30 mm.[18]

The percentage of patients treated with surgical resection differed greatly among trials, from 0%[17,21,23] to 100%.[17,18] Mean albumin and bilirubin levels were comparable among the studies, ranging from 3.2 g/dL[15] to 4 g/dL,[18] and from 0.7 mg/dL[21] to 1 mg/dL[18,25] respectively. Most studies[15-18,20-22,25] reported platelet counts, which ranged from 97×10^6/dL[16] to 141×10^6/dL.[18] Among the studies, the frequency of a Child-Pugh score of A ranged from 80%[17] to 100%.[24] Only six studies[16,18,20-22,25] provided information about mean AFP levels, which ranged from 11 ng/mL[24] to 189 ng/mL.[20] Methodological quality scores ranged from 3[19,21,22,24] to 7[18] on a scale of 0-7 (Table 2).

3.2 | HCC recurrence

Pooled estimates of 6-month, 1-year and 2-year recurrence actuarial probabilities were 7.4% (95% confidence interval [CI], 4.0-10.0%; range, 0-12.5%), 20% (95% CI: 12.7-27.4%; range, 4.9-62.5%) and 47% (95% CI: 39.5-54.4%; range, 31.8-100%) respectively (Table 3). There was statistically significant heterogeneity among studies for the 2-year recurrence probability (p=.0002) (Figure 2A). Univariate logistic meta-regression analysis was used to identify potential sources of heterogeneity among studies. Of the 15 variables assessed, only three variables were associated with an increase in the 2-year recurrence rate: study design (RCTs vs other studies; p<.001), albumin levels (p=.0245), and study duration (p=.0319) (Table S3).

Hepatocellular carcinoma recurrence curves extracted from studies and the summary HCC recurrence curve are shown in Figure 3A. The median time to recurrence (95% CI) was 25.4 months (21.3-29.5 months). Baseline albumin level was the only patient-level covariate to be significantly associated with HCC recurrence. Pooled actuarial HCC recurrence curves stratified by albumin level are shown in Figure S1. Figures S2 and S3 show Forest Plot of 2-year HCC recurrence rates and summary curves of the HCC recurrences stratified according to type of HCC treatments.

3.3 | Survival

Pooled estimates of 3-year and 5-year survival actuarial probabilities were 79.8% (95% CI: 74.2-85.8%; range, 65.3-95.1%), and 58.6% (95% CI: 51.1-67.1%; range, 46.7-78.4%) respectively (Table 3). There was statistically significant heterogeneity among studies at 3 years (p<.001) (Figure 2B). Univariate logistic regression analysis was used to identify potential sources of heterogeneity among the studies. Among the 15 variables assessed, only two patient-level covariates were associated with a decrease in the 3-year survival rate: size of the main tumour (p=.0074) and AFP level (p=.0374) (Table S3). OS curves extracted from the studies and summary survival curves are shown in Figure 3B. Median survival (95%CI) was 68.1 months (60-93 months). Pooled actuarial OS curves stratified by size of the main tumour are shown in Figure S4.

Figures S5 and S6 show Forest Plot of 3-year survival rates and summary curves of the survival stratified according to type of HCC treatments.

3.4 | Publication bias

The funnel publication bias plot for the 2-year recurrence rates (Figure S7) and the Egger test for publication bias showed that the risk of having missed or overlooked trials was not significant (p=.07). The funnel publication bias plot for 3-year survival rates (Figure S8) and the Egger test for publication bias showed that the risk of having missed or overlooked trials was not significant (p=.33).

TABLE 2 Assessment of study quality

First author (year)	Study design	Study type	Patients consecutively included	Percentage of drop-out	Complete radiological response adequately defined after curative treatment	Follow-up >3 years	Defined protocol for recurrence surveillance	Score
Ikeda (2000)[15]	1	1	0	1	0	0	1	4
Suou (2001)[19]	0	0	0	0	1	1	1	3
Shiratori (2003)[16]	1	1	1	0	1	1	1	6
Hung (2005)[20]	0	0	0	0	1	0	1	2
Nishiguchi (2005)[17]	1	1	1	0	0	1	0	4
Yamanaka (2005)[23]	0	0	0	0	1	0	1	2
Mazzaferro (2006)[18]	1	1	1	1	1	1	1	7
Kudo (2007)[21]	0	0	1	0	1	0	1	3
Jeong (2007)[22]	0	0	1	0	1	0	1	3
Kanogawa (2014)[24]	0	0	0	0	1	1	1	3
Petta (2016)[25]	0	1	1	0	1	0	1	4

4 | DISCUSSION

We performed a meta-analysis of individual and aggregated data of single HCV-untreated arm from 11 studies of HCV-related early HCC, in which patients achieved a complete response after curative resection or ablation. Our data revealed a pooled 2-year recurrence actuarial probability of 47% and a pooled 3-year survival actuarial probability of 79.8%. We observed high heterogeneity among studies in the recurrence and survival rates. Although the number of included patients in the available studies was large, suggesting robustness of the estimated rates, the 95% CIs of 2-year recurrence (39.5-54.4%) and 3-year survival (74.2-85.8%) distributions were wide. Clinical heterogeneity of recurrence and survival was a common feature of these studies, with 2-year recurrence and 3-year survival rates ranging from 31.8% to 95.5% and from 65% to 95.5% respectively.

To explain the wide variability in the natural course of patients with HCV-related early HCC after curative treatment, we stratified studies according to patient- and study-level variables identified by meta-regression analyses. Mean tumour size and AFP level were significant predictors of 3-year survival, confirming that cancer-related factors significantly impact survival in patients with compensated cirrhosis. When studies were separated by tumour size, the 3-year survival was much higher in studies including patients with a mean tumour size <22 mm than in those with a mean tumour size >22 mm.

Although mean baseline AFP levels significantly correlate with survival, heterogeneity of survival rates among studies may reflect variability in the molecular characteristics and biological behaviour of the tumour. At present, no clinical feature-based scoring system accounts for the molecular characteristics and pathobiology of the tumour (invasiveness, doubling time, angiogenesis and microvascular invasion).[30] The inclusion of molecular variables,[31] such as signature gene expression profiling,[32] in future staging systems could improve their predictive ability.

Our analyses were unable to explain fully the observed heterogeneity in HCC recurrence. No patient-level variable except baseline serum albumin level was significantly associated with 2-year recurrence. Furthermore, only follow-up duration and study design remained significantly associated with recurrence rate. We found that baseline serum albumin level was a robust predictor of HCC recurrence by meta-regression analysis. This finding is consistent with the results of two published prospective studies suggesting that serum albumin is an independent predictor of HCC recurrence in patients with HCV-[33] or HBV-related[34] HCC after attempted curative treatments.

Regarding study-level variables, study design (RCTs vs other studies) and follow-up duration were markers of better study quality and better outcome ascertainment. This finding was particularly true for time-to-event outcomes like recurrence, which are likely to be affected by the follow-up duration.

The availability of safe and effective IFN-free antiviral regimens for the cure of HCV infection opens the crucial question of whether virological eradication by DAAs improves outcomes (recurrence and/or survival). In this particular clinical setting, DAAs are used without

TABLE 3 Recurrence and survival actuarial probability of the studies in the meta-analysis

	Recurrence (%)			Survival (%)	
First author (year)	6-month	1-year	2-year	3-year	5-year
Ikeda (2000)[15]	12.5	62.5	100	NA	NA
Suou (2001)[19]	4.5	13.6	31.8	95	78.4
Shiratori (2003)[16]	8	24	68	84	48
Hung (2005)[20]	NA	NA	NA	65.3	NA
Nishiguchi (2005)[17]	0	20	47	80	46.7
Yamanaka (2005)[23]	8	23	60	NA	NA
Mazzaferro (2006)[18]	10.5	37.6	53.9	71.1	48.7
Kudo (2007)[21]	4.8	8.6	45	81.5	66.2
Jeong (2007)[22]	0	4.9	32.7	95.1	72.7
Kanogawa (2014)[24]	4.9	18.8	44.6	78.3	60.1
Petta (2016)[25]	9.5	21	40.6	79.3	64.8

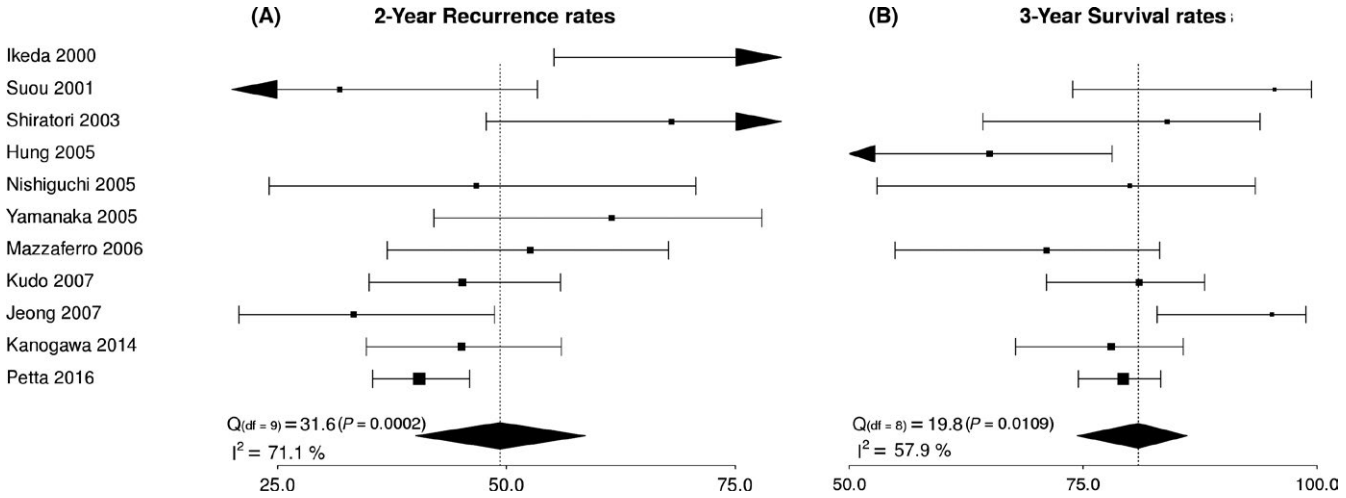

FIGURE 2 Forest plot of (A) 2-year HCC recurrence rates and (B) 3-year survival rates of studies included in the meta-analysis, obtained using a random-effects model. Studies are arranged by publication year

any evidence of long-term effectiveness, and trials comparing DAAs to placebo are unfeasible. The concern cannot be resolved by extrapolating data on IFN-based treatments to DAAs. Several meta-analyses[8-11] have suggested that IFN treatment may prevent HCC recurrence, with conflicting data on survival. Nevertheless, it may be that IFN exerts its effect on HCC recurrence not only indirectly by leading to virological eradication, but also directly via its intrinsic antifibrotic, antiproliferative, immunomodulatory, antiangiogenic and antitumoural activities.

This complex and uncertain picture has been further complicated by a recent uncontrolled study by Reig et al.[13] Despite serious methodological concerns, this study noted an unexpectedly high early HCC recurrence rate in a small cohort of patients with successfully treated HCC after viral eradication by DAAs.[13] The 6-month actuarial probability of early HCC recurrence assessed by the summary actuarial curve in our meta-analysis on HCV viraemic patients (7.4%) was higher than the probability re-analysed by Cammà et al. (5.2%),[35] which was based on individual data reported by Reig et al.[13] Therefore, our data provide indirect evidence that IFN-free regimens for HCV do not increase the probability of early HCC recurrence. According to this result, our meta-analysis can be considered a useful benchmark for obtaining indirect comparisons among different uncontrolled studies estimating benefit in the adjuvant setting, such as for DAA-based therapies and new adjuvant drugs.

Studies that were pooled in this meta-analysis predominantly included "healthier" patients enrolled in clinical trials of IFN-based adjuvant treatment. This choice limits the broad application of the results to the "sickest" patients who would potentially be treatable with IFN-free regimens for HCV. However, patients with early HCC who underwent resection or ablation had well-compensated cirrhosis. Therefore, we are confident that a selection bias, applying to IFN-based but not to IFN-free regimens, is unlikely in this setting of patients with well-compensated cirrhosis and potentially curable early HCC.

As with all meta-analyses, the methodology of the current study leads to a potential limitation of the generalizability of its results to new populations and settings. The included studies were performed in highly specialized centres, mostly from the Asian-Pacific area. Because

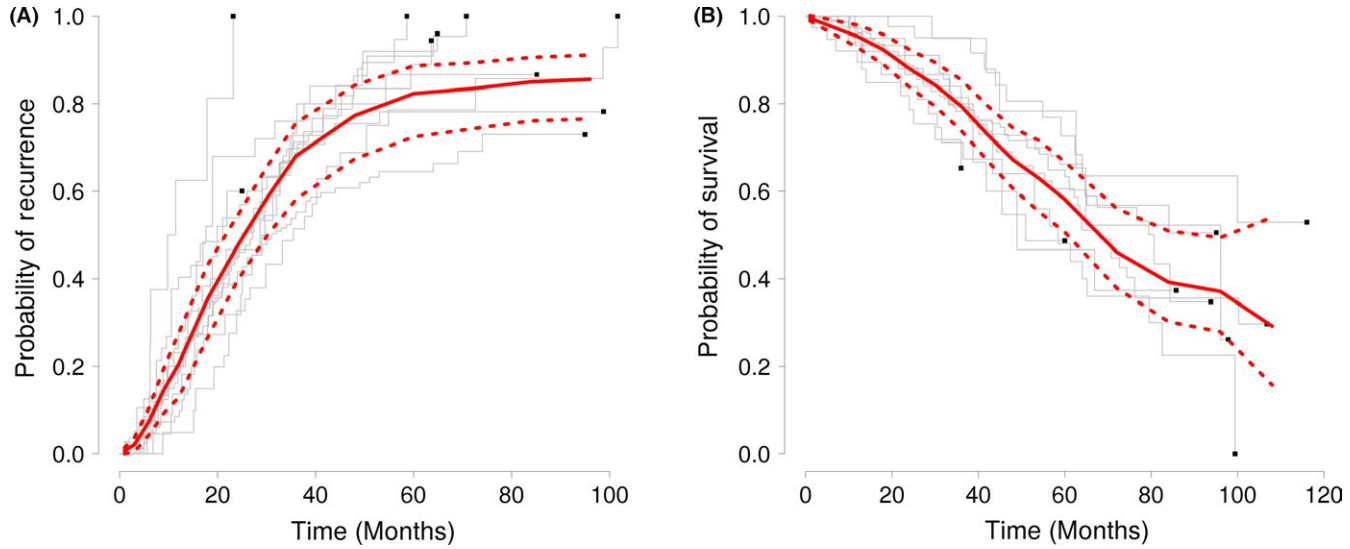

FIGURE 3 Curves of (A) HCC recurrence and (B) OS in studies included in the meta-analysis. Grey lines represent recurrences in each study. Black squares indicate the end of the follow-up. Thick lines represent the summarized recurrence curves with the 95% confidence bands (dashed lines) obtained using the approach of Combescure et al. with random effects [Colour figure can be viewed at wileyonlinelibrary.com]

of the HCV-related etiology of the underlying cirrhosis, the region of the study was not associated with recurrence or survival rates by meta-regression. Although the overall sample size of patients analysed exceeded 700, it was not large enough to allow definitive conclusions. Differences in baseline severity of illness (single vs multi-nodular HCC), treatment modalities (resection vs ablation), and length of follow-up in the study populations may limit the accuracy of this meta-analysis. We attempted to control these differences by including covariates that described patient- and study-level features. Unfortunately, our study is limited by the patient-level covariates reported in each of the studies, which were not consistent across trials. Therefore, these summary results describe only between-study, not between-patient, variation because they reflect group averages rather than individual data. Moreover, there were likely other potentially important confounders for which we did not control and that might have affected the results. Lack of data on microscopic vascular invasion, histological grading and gene profiling[32] could also affect the accuracy of the results.

Despite the lack of availability of individual data for all patients included in the meta-analysis, we were able to analyse recurrence and survival as time-dependent variables. As expected, we found that follow-up duration was strongly linked to the 2-year recurrence rate by meta-regression analysis. This result further emphasizes that the results of meta-analyses of time-to-event outcomes are likely to be affected by censoring and by the different follow-up durations of included studies.[36] These limitations are particularly important when the follow-up across trials is heterogeneous, as were the mean follow-up periods of the studies included in our meta-analysis (range: 25-85 months). Moreover, advantages in combining aggregate and individual patient data, including the ability to reduce biases, have been illustrated in several meta-analyses.[37] Another limitation of the present study is the lack of data about recurrence and survival rates in other clinical settings (HBV infection, non-alcoholic and alcoholic fatty liver disease, etc.), an issue worthy of investigation in future meta-analyses.

Concerning the methodological quality of the included studies, the following points should be emphasized. (i) Only two studies had a percentage of drop-out <10%, leading to ascertainment bias. (ii) Two studies did not adequately define complete response, leading to possible measurement bias. (iii) One study did not have a defined surveillance protocol, leading to ascertainment bias. Finally, (iv) only five studies had a follow-up >3 years. Results of meta-analyses of time-to-event outcomes are likely affected by censoring and follow-up duration of individual trials. However, the evaluation of methodological quality did not seem to influence the outcomes of the meta-regression.

The available evidence from this meta-analysis is sufficient to conclude that in patients with HCV-related early HCC who achieved a complete response: (i) the 2-year recurrence (47%) and 3-year survival (79.8%) pooled actuarial probability are extremely variable, and no single patient or study characteristic can fully explain this heterogeneity; and (ii) study design (RCTs), low albumin levels, and follow-up duration are associated with a higher likelihood of recurrence, whereas tumour size and AFP levels are associated with a lower survival. These pooled reported actuarial recurrence and survival probabilities provide a useful benchmark for indirect comparisons of the benefit of HCV eradication by DAAs and for a correct design of RCTs of new adjuvant drugs.

CONFLICT OF INTEREST

The authors do not have any disclosures to report.

REFERENCES

1. Torre LA, Bray F, Siegel RL, et al. Global cancer statistics, 2012. *CA Cancer J Clin*. 2015;65:87–108.
2. Hepatobiliary Cancers. NCCN Clinical Practice Guidelines in Oncology (NCCN Guidelines) 2015: 1–114.
3. European Association for the Study of the Liver; European Organisation for Research and Treatment of Cancer. EASL-EORTC

clinical practice guidelines: management of hepatocellular carcinoma. *J Hepatol*. 2012;56:908–943.
4. Bruix J, Reig M, Sherman M. Evidence-based diagnosis, staging, and treatment of patients with hepatocellular carcinoma. *Gastroenterology*. 2016;150:835–853.
5. Cabibbo G, Reig M, Gadaleta-Caldarola G, et al. The calm before the storm: a report from the International Liver Cancer Association Congress 2015 – part 1. *Future Oncol*. 2016;12:281–284.
6. Bruix J, Sherman M, American Association for the Study of Liver Diseases. Management of hepatocellular carcinoma: an update. *Hepatology*. 2011;53:1020–1022.
7. Bruix J, Takayama T, Mazzaferro V, et al. Adjuvant sorafenib for hepatocellular carcinoma after resection or ablation (STORM): a phase 3, randomised, double-blind, placebo-controlled trial. *Lancet Oncol*. 2015;16:1344–1354.
8. Singal AK, Freeman DH Jr, Anand BS. Meta-analysis: interferon improves outcomes following ablation or resection of hepatocellular carcinoma. *Aliment Pharmacol Ther*. 2010;32:851–858.
9. Miao RY, Zhao HT, Yang HY, et al. Postoperative adjuvant antiviral therapy for hepatitis B/C virus-related hepatocellular carcinoma: a meta-analysis. *World J Gastroenterol*. 2010;16:2931–2942.
10. Zhuang L, Zeng X, Yang Z, et al. Effect and safety of interferon for hepatocellular carcinoma: a systematic review and meta-analysis. *PLoS One*. 2013;8:e61361.
11. Zhang W, Song TQ, Zhang T, et al. Adjuvant interferon for early or late recurrence of hepatocellular carcinoma and mortality from hepatocellular carcinoma following curative treatment: a meta-analysis with comparison of different types of hepatitis. *Mol Clin Oncol*. 2014;2:1125–1134.
12. AASLD/IDSA HCV Guidance Panel. Hepatitis C guidance: AASLD-IDSA recommendations for testing, managing, and treating adults infected with hepatitis C virus. *Hepatology*. 2015;62:932–954.
13. Reig M, Mariño Z, Perelló C, et al. Unexpected early tumor recurrence in patients with hepatitis C virus -related hepatocellular carcinoma undergoing interferon-free therapy: a note of caution. *J Hepatol*. 2016;65:719–726.
14. Shamseer L, Moher D, Clarke M, et al. Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015: elaboration and explanation. *BMJ*. 2015;349:g7647.
15. Ikeda K, Arase Y, Saitoh S, et al. Interferon beta prevents recurrence of hepatocellular carcinoma after complete resection or ablation of the primary tumor-A prospective randomized study of hepatitis C virus-related liver cancer. *Hepatology*. 2000;32:228–232.
16. Shiratori Y, Shiina S, Teratani T, et al. Interferon therapy after tumor ablation improves prognosis in patients with hepatocellular carcinoma associated with hepatitis C virus. *Ann Intern Med*. 2003;138:299–306.
17. Nishiguchi S, Tamori A, Kubo S. Effect of long-term postoperative interferon therapy on intrahepatic recurrence and survival rate after resection of hepatitis C virus-related hepatocellular carcinoma. *Intervirology*. 2005;48:71–75.
18. Mazzaferro V, Romito R, Schiavo M, et al. Prevention of hepatocellular carcinoma recurrence with alpha-interferon after liver resection in HCV cirrhosis. *Hepatology*. 2006;44:1543–1554.
19. Suou T, Mitsuda A, Koda M, et al. Interferon alpha inhibits intrahepatic recurrence in hepatocellular carcinoma with chronic hepatitis C: a pilot study. *Hepatol Res*. 2001;20:301–311.
20. Hung CH, Lee CM, Wang JH, et al. Antiviral therapy after non-surgical tumor ablation in patients with hepatocellular carcinoma associated with hepatitis C virus. *J Gastroenterol Hepatol*. 2005;20:1553–1559.
21. Kudo M, Sakaguchi Y, Chung H, et al. Long-term interferon maintenance therapy improves survival in patients with HCV-related hepatocellular carcinoma after curative adiofrequency ablation. A matched case-control study. *Oncology*. 2007;72(Suppl. 1):132–138.
22. Jeong SC, Aikata H, Katamura Y, et al. Effects of a 24-week course of interferon-alpha therapy after curative treatment of hepatitis C virus-associated hepatocellular carcinoma. *World J Gastroenterol*. 2007;13:5343–5350.
23. Yamanaka Y, Shiraki K, Miyashita K, et al. Risk factors for the recurrence of hepatocellular carcinoma after radiofrequency ablation of hepatocellular carcinoma in patients with hepatitis C. *World J Gastroenterol*. 2005;11:2174–2178.
24. Kanogawa N, Ogasawara S, Chiba T, et al. Sustained virologic response achieved after curative treatment of hepatitis C virus-related hepatocellular carcinoma as an independent prognostic factor. *J Gastroenterol Hepatol*. 2015;30:1197–1204.
25. Petta S, Cabibbo G, Barbara M, et al. Hepatocellular carcinoma recurrence in patients with curative resection or ablation: impact of HCV eradication does not depend on the use of interferon. *Aliment Pharmacol Ther*. 2017;45:160–168.
26. Combescure C, Foucher Y, Jackson D. Meta-analysis of single-arm survival studies: a distribution-free approach for estimating summary survival curves with random effects. *Stat Med*. 2014;33:2521–2537.
27. Earle CC, Pham B, Wells GA. An assessment of methods to combine published survival curves. *Med Decis Making*. 2000;20:104–111.
28. DerSimonian R, Laird N. Meta-analysis in clinical trials. *Control Clin Trials*. 1986;7:177–188.
29. Cabibbo G, Enea M, Attanasio M, et al. A meta-analysis of survival rates of untreated patients in randomized clinical trials of hepatocellular carcinoma. *Hepatology*. 2010;51:1274–1283.
30. Maida M, Orlando E, Cammà C, et al. Staging systems of hepatocellular carcinoma: a review of literature. *World J Gastroenterol*. 2014;20:4141–4150.
31. Villa E, Colantoni A, Cammà C, et al. Estrogen receptor classification for hepatocellular carcinoma: comparison with clinical staging systems. *J Clin Oncol*. 2003;21:441–446.
32. Villa E, Critelli R, Lei B, et al. Neoangiogenesis-related genes are hallmarks of fast-growing hepatocellular carcinomas and worst survival. Results from a prospective study. *Gut*. 2016;65:861–869.
33. Iwadou S, Nouso K, Kuwaki K, et al. Time-dependent analysis of predisposing factors for the recurrence of hepatocellular carcinoma. *Liver Int*. 2010;30:1027–1032.
34. Kim SU, Jung KS, Lee S, et al. Histological subclassification of cirrhosis can predict recurrence after curative resection of hepatocellular carcinoma. *Liver Int*. 2014;34:1008–1017.
35. Cammà C, Cabibbo G, Craxì A. Direct antiviral agents and risk for hepatocellular carcinoma early recurrence: much ado about nothing. *J Hepatol*. 2016;65:861–862.
36. Vale CL, Tierney JF, Stewart LA. Effects of adjusting for censoring on meta-analyses of time-to-event outcomes. *Int J Epidemiol*. 2002;31:107–111.
37. Jeng GT, Scott JR, Burmeister LF. A comparison of meta-analytic results using literature vs individual patient data. Paternal cell immunization for recurrent miscarriage. *JAMA* 1995;274:830–836.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

How to cite this article: Cabibbo G, Petta S, Barbàra M, et al. A meta-analysis of single HCV-untreated arm of studies evaluating outcomes after curative treatments of HCV-related hepatocellular carcinoma. *Liver Int*. 2017;37:1157–1166. https://doi.org/10.1111/liv.13357

APPENDIX

Additional members of the ITA.LI.CA groups

Dipartimento di Scienze Mediche e Chirurgiche, Alma Mater Studiorum – Università di Bologna: Laura Bucci, Marco Zoli, Fabio Piscaglia, Francesca Garuti, Barbara Lenzi, Maurizio Biselli, Paolo Caraceni, Alessandro Cucchetti, Annagiulia Gramenzi, Alessandro Granito, Donatella Magalotti, Carla Serra, Giulia Negrini, Lucia Napoli, Veronica Salvatore, Francesca Benevento; *Dipartimento di Scienze Chirurgiche e Gastroenterologiche, Università di Padova*: Luisa Benvegnù, Alessia Gazzola, Francesca Murer, Caterina Pozzan, Veronica Vanin; *Dipartimento di Medicina Interna, Unità di Gastroenterologia, IRCCS-Azienda Ospedaliera Universitaria San Martino-IST, Università di Genova*: Alessandro Moscatelli, Gaia Pellegatta, Antonino Picciotto, Vincenzo Savarino; *Unità Operativa di Chirurgia, Policlinico S. Marco, Zingonia*: Francesca Ciccarese, Paolo Del Poggio, Stefano Olmi; *Unità di Medicina Interna e Gastroenterologia, Complesso Integrato Columbus, Università Cattolica di Roma, Roma*: Gian Lodovico Rapaccini Nicoletta de Matthaeis; *Unità Operativa di Medicina, Azienda Ospedaliera Bolognini, Seriate, Italia*: Mariella Di Marco Claudia Balsamo, Elena Vavassori; *Unità Operativa di Gastroenterologia, Ospedale Belcolle, Viterbo*: Paola Roselli; *Unità Operativa di Medicina Protetta, Ospedale Belcolle, Viterbo*: Serena Dell'Isola, Anna Maria Ialungo, Elena Rastrelli; *Dipartimento Biomedico di Medicina Interna e Specialistica, Unità di Gastroenterologia, Università di Palermo:* Calogero Cammà, Giuseppe Cabibbo, Simona Attardo, Margherita Rossi, Andrea Costantino; *Dipartimento Biomedico di Medicina Interna e Specialistica, Unità di Medicina Interna 2, Azienda Ospedaliera Ospedali Riuniti Villa Sofia-Cervello, Palermo*: Andrea Affronti, Marco Affronti, Marta Mascari; *Ospedale Regionale di Bolzano, Unità di Gastroenterologia, Bolzano*: Martina Felder, Andrea Mega; *Unità di Medicina Interna e Gastroenterologia, Policlinico Gemelli, Università Cattolica di Roma, Roma*: Antonio Gasbarrini, Maurizio Pompili, Emanuele Rinninella; *Unità Operativa Gastroenterologia e Malattie del Ricambio, Azienda Ospedaliero-Universitaria Pisana, Pisa*: Rodolfo Sacco, Valeria Mismas; *Dipartimento di Medicina Interna; Ospedale per gli Infermi di Faenza, Faenza*: Francesco Giuseppe Foschi, Anna Chiara Dall'Aglio, Valentina Feletti, Arianna Lanzi, Federica Mirici Cappa, Elga Neri, Giuseppe Francesco Stefanini, Stefano Tamberi; *Unità di Malattie Infettive ed Epatologia, Azienda Ospedaliero-Universitaria di Parma. Parma:* Andrea Olivani, Elisabetta Biasini; *Dipartimento di Medicina Clinica e Chirurgia, Unità di Gastroenterologia, Università di Napoli "Federico II", Napoli:* Gerardo Nardone, Maria Guarino; *Clinica di Gastroenterologia, Università Politecnica delle Marche, Ancona*: Gialuca Svegliati-Baroni, Alessio Ortolani; *Unità di Gastroenterologia, Ospedale Sacro Cuore Don Calabria, Negrar*: Alberto Masotto, Fabiana Marchetti, Matteo Valerio; *Medicina Interna ed Epatologia, Dipartimento di Medicina Sperimentale e Clinica – Università di Firenze, Firenze*: Fabio Marra, Sami Aburas, Andrea L. Inghilesi; *Dipartimento di Medicina Diagnostica e Prevenzione, Azienda ospedaliero-universitaria di Bologna, Unità Operativa di Radiologia*: Alberta Cappelli, Rita Golfieri, Cristina Mosconi, Matteo Renzulli; *Dipartimento di Medicina Clinica e Chirurgia, Unità di Epato-Gastroenterologia, Università di Napoli "Federico II", Napoli*: Piero Coccoli, Marco Sanduzzi Zamparelli.