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Hepatocellular carcinoma locoregional therapies for patients in the waiting list. Impact on transplantability and recurrence rate

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Summary

The practice of treating candidates for liver transplantation (LT) for hepatocellular carcinoma (HCC), with locoregional therapies, is common in most transplant centers. However, for T1 tumors and expected waiting times to LT <6 months, there is no evidence that these treatments are beneficial. For T2 tumors and for longer waiting times, neo-adjuvant treatments are usually performed with transarterial chemoembolization (TACE), ablation techniques and liver resection in selected cases. The treatment choice should be based on the BCLC staging system. At present, there is no evidence of the superiority of ablation/resection *vs.* TACE, but some studies showed better results of the former in achieving a complete response. The response to neo-adjuvant treatments should be evaluated through mRECIST criteria, but few studies adopted these criteria and properly analyzed factors affecting response. The simultaneous evaluation of the impact of neo-adjuvant therapies on dropout rate, post-LT HCC recurrence and patient survival is rarely reported. Tumor stage and volume, alpha-fetoprotein levels, response to treatments and liver function affect pre-LT outcomes. These same factors, together with vascular invasion and poor tumor differentiation, are major determinants of poor post-LT outcomes. Due to the low number of prospective studies with well-defined entry criteria and the variability of results, the role of downstaging is still to be defined. Novel molecular markers seem promising for the estimation of prognosis and/or response to treatments. With a persistent scarcity of organ donors, neo-adjuvant treatments can help identify patients with different probabilities of cancer progression, and consequently balance the priority of HCC and non-HCC-candidates through revised additional scores for HCC.

Introduction

Hepatocellular carcinoma (HCC) is one of the 5 most common malignancies worldwide, and its incidence is increasing in Western countries [1,2]. For patients with HCC and cirrhosis, liver transplantation (LT) represents the treatment of choice and provides excellent oncological results and a cure for cirrhosis. However, not all patients with HCC and cirrhosis can undergo transplantation because of the scarcity of liver donors.

HCC patients on the waiting list (WL) for transplantation can experience tumor growth beyond the accepted criteria for LT; the practice of treating HCC patients with hepatic resection or locoregional therapies before they are placed on the WL or while they are awaiting has thus gained favor and is now the standard of care in most transplant centers [3–6].

Radiofrequency ablation (RFA), percutaneous ethanol injection (PEI), and transarterial chemoembolization (TACE) have considerably improved in the last decade, and they can have a positive impact on tumor growth control [3–7]. Similarly, the great improvements in diagnostic techniques and surveillance schedules have led to earlier diagnoses and better accuracy, and this has resulted in the increased curability of liver tumors.

Locoregional treatments can be used as neo-adjuvant therapies with two intents in the setting of LT. The first one is to prevent the dropout from the WL in patients meeting accepted criteria of transplantability; in this case, locoregional treatments are defined as bridging procedures. The second one is to treat patients initially outside criteria for LT in order to reach T2 stage HCC, to fulfill Milan criteria (MC) [8], University of California San Francisco criteria (UCSFC) [9], or other criteria, which allows entry to the WL for LT after an adequate period of follow-up, to verify the effectiveness of neo-adjuvant treatment. In this case, locoregional therapies are used as downstaging procedures.

Keywords: Hepatocellular carcinoma; Liver transplantation; Locoregional treatments.

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Abbreviations: HCC, hepatocellular carcinoma; LT, liver transplantation; WL, waiting list; RFA, radiofrequency ablation; PEI, percutaneous ethanol injection; TACE, transarterial chemoembolization; MC, Milan criteria; UCSFC, University of California San Francisco criteria; mRECIST, modified Response Evaluation Criteria in Solid Tumors; CT, computed tomography; MRI, magnetic resonance imaging; AFP, alpha-fetoprotein; BCLC, Barcelona Clinic Liver Cancer; TACE-DEB, TACE with Drug-Eluting Beads; TARE, Trans-arterial radioembolization; MELD, Model for End-stage Liver Disease; HCV, hepatitis C virus; deMELD, dropout equivalent MELD.

Key Points

- Locoregional treatments are widely used in cirrhotic patients with hepatocellular carcinoma (HCC) listed for liver transplantation (LT) in order to prevent tumor progression, even though there is no strong evidence that neo-adjuvant treatments should be applied if the expected waiting time for LT is shorter than 6 months
- Neo-adjuvant treatments include transarterial chemoembolization (TACE), radiofrequency ablation, ethanol injection and liver resection, which should be selected according to the Barcelona Clinic Liver Cancer (BCLC) scoring system
- Other procedures, such as TACE with drug-eluting beads, transarterial radioembolization, radiotherapy, microwave ablation, cryoablation and irreversible electroporation, though promising, are still under investigation
- The efficacy of neo-adjuvant treatments should be evaluated by the rate of dropout from the WL and, methodologically, with a 3-month interval reassessment of modified Response Evaluation Criteria in Solid Tumors (mRECIST) and serum alpha-fetoprotein sampling
- Neo-adjuvant treatments have the 3 main purposes of controlling HCC progression for expected long waiting times, identifying patients with different probabilities of cancer progression and helping in balancing the priority of HCC and non-HCC candidates for LT

In order to assess the actual evidence of the impact of neo-adjuvant treatment in the management of potential candidates for LT, the following items will be discussed in the present paper:

(1) Are neo-adjuvant locoregional treatments indicated in patients considered for LT?
(2) How should response to locoregional treatments be evaluated, and what timing should be adopted for patient monitoring on the WL?
(3) Which types of locoregional treatment are available for patients considered for LT?
(4) Which is the best neo-adjuvant treatment in this setting?
(5) Which are the patient or tumor characteristics related to an unsuccessful neo-adjuvant therapy, a higher dropout rate, and a worse post-LT outcome?
(6) Can the effect of neo-adjuvant treatments be used to balance priority of HCC and non-HCC candidates?
(7) Are there new molecular markers for a better estimation of tumor biological behavior and/or response to treatment?

Are neo-adjuvant locoregional treatments indicated in patients listed for LT?

An international consensus conference was held in 2010 with the aim of reviewing current practice regarding LT in patients with HCC and to develop internationally accepted statements and guidelines [10]. Thirty-seven statements covering all issues of LT for HCC were produced; among these, 5 statements were focused on the management of patients on the WL. No recommendation could be made on bridging therapy in patients with United Network for Organ Sharing (UNOS) T1 HCC due to the absence of scientific evidence. In patients with UNOS T2 HCC and a likely waiting time longer than 6 months, locoregional therapy may be appropriate, but the low level of evidence for prognosis led to a weak recommendation. In fact, a cost-effective analysis based on Markov model and the review of cohort studies, indicate a benefit for bridging therapies if the waiting time is expected to be longer than 6 months [11–14]. However, in the clinical practice and given the often unpredictable waiting time for LT, there is a widespread attitude to treat most patients in the WL. In the following sections of this review, we will focus on the possible benefits derived from the routine adoption of neo-adjuvant treatments.

How should response to locoregional treatments be evaluated?

Whatever the type of locoregional therapy chosen, the response to neo-adjuvant treatments should be evaluated with the modified Response Evaluation Criteria in Solid Tumors (mRECIST) [15,16]. The RECIST criteria were amended as mRECIST in 2008, based on the concept that the evaluation of the treatment response should take into account the induction of intratumoral necrotic areas in estimating the decrease in tumor load, and not just a reduction in overall tumor size [17].

Patients can be followed with either contrast-enhanced spiral computed tomography (CT), preferably with use of multislice scanners, or contrast-enhanced dynamic magnetic resonance imaging (MRI). The administration of intravenous contrast is recommended for CT and MRI, if not medically contraindicated. In contrast-enhanced studies, it is mandatory to obtain a dual-phase imaging of the liver [15,16].

According to mRECIST criteria, the following definitions should be applied for tumor response to treatment: (A) complete response: the disappearance of any intratumoral arterial enhancement in all target lesions; (B) partial response: at least a 30% decrease in the sum of diameters of viable (contrast enhancement in the arterial phase) target lesions, taking as the reference the baseline sum of the diameters of target lesions; (C) progressive disease: an increase of at least 20% in the sum of the diameters of viable (enhancing) target lesions, taking as the reference the smallest sum of the diameters of viable (enhancing) target lesions recorded since the treatment started; (D) stable disease: any cases that do not qualify for either partial response or progressive disease [15,16].

Overall response is a result of the combined assessment of target lesions, non-target lesions, and new lesions.

Unfortunately, studies focusing on neo-adjuvant treatments before LT do not report the response to this therapy with uniform and/or well-defined parameters, and mRECIST, in particular, have rarely been used so far. Relationships between response to therapy and dropout from WL should represent the main aim of any dedicated study on this issue, and the capability of any proposed neo-adjuvant treatment should be assessed in the view of dropout due to tumor progression rather than response by itself.

Conversely, there is general agreement that monitoring of patients on the WL should be performed with the above reported

imaging techniques and the evaluation of serum alpha-fetoprotein (AFP) with a 3-month interval after listing [10].

Which types of locoregional treatment are available for patients considered for LT?

Currently, TACE and percutaneous ablation are the treatments most frequently used in patients listed for LT or included in a downstaging program. This is also derived from the revised Barcelona Clinic Liver Cancer (BCLC) staging system, according to which local ablation is considered the first-line treatment option for patients at early stages, not suitable for surgical therapies (BCLC 0-A), while TACE is the first-line therapy for patients at intermediate stages of the disease (unresectable, multifocal disease confined to the liver in the absence of portal vein thrombosis and in asymptomatic patients; BCLC B) [18].

Regarding local ablation, RFA is recommended in most instances as the main ablative therapy in tumors less than 5 cm due to a significantly better control of the disease, while PEI is recommended in cases where RFA is not technically feasible (around 10–15%). In selected cases, laparoscopic ablation can be performed. Other ablative therapies, such as microwave ablation, cryoablation, and irreversible electroporation are still under investigation [18].

TACE extends the survival of patients with BCLC stage B from a median of 16 months (untreated cases) to a median of up to 19–20 months, according to randomized controlled trials and a meta-analysis of pooled data [19]. Although not supported by studies with high levels of evidence, doxorubicin or cisplatin are the best chemotherapeutical agents to be used with TACE, while the procedure should be applied 3–4 times per year, since more intense regimes [i.e., TACE every 2 months] can induce liver failure in an unacceptable proportion of patients. Superselective TACE is recommended to minimize the ischemic insult to non-tumoral liver and to increase the rate of tumor necrosis [7,18].

Alternatives to TACE can be represented by TACE with Drug-Eluting Beads (TACE-DEB), radioembolization or external radiation. TACE-DEB has shown similar response rates to standard TACE, with less liver-related toxicity and systemic adverse events [20,21].

Transarterial radioembolization (TARE) with ^{131}I or ^{90}Y glass beads has shown a good anti-tumoral effect with an acceptable safety profile, but at present, it cannot be recommended as standard therapy, based on the absence of studies with high levels of evidence comparing TARE with TACE [22–25].

External three-dimensional conformal radiotherapy is under investigation [26], and at present there is no evidence to support this therapeutic approach in the management of HCC.

Which is the best neo-adjuvant treatment in patients considered for LT?

According to the previously mentioned international consensus conference held in 2010, no recommendation can be made for preferring any type of locoregional therapy to others, in patients listed for LT or in those entering a downstaging protocol [10]. This statement is based on the lack of randomized trials on this topic. Indeed, the potential benefits advocated for local ablation or TACE are derived from observational studies and cost-effectiveness analyses [10].

TACE is the treatment most frequently adopted, alone or in combination with ablation/resection, in patients listed for LT (bridging treatments) [3,27–42], or included in a program of downstaging [3,29,34,37,40–48]. Results of neo-adjuvant locoregional treatments are reported as related to the use of TACE alone [3,27,29,31,33,34,37,43,44,48], ablation alone [4,49,50], or multimodal treatments [28,30,32,35,36,38–42,45–47].

In general, most published series are single-center and with a low number of patients. Furthermore, there is a great heterogeneity among studies as regards the types of treatments considered, the center-specific criteria for LT for HCC, and the results reported in terms of: achievement of tumor necrosis, radiological response to treatment, dropout rate, waiting time for LT, proportion of patients transplanted, post-transplant survival, intention-to-treat survival and post-LT recurrence rate [3,4,27–51].

The highest accordance between different studies is in the demonstration that RFA achieves higher rates of complete necrosis (46–74%) [4,49,50] compared with TACE (22–29%) [27,35,48,51]. Cucchetti and colleagues showed that patients treated with potentially curative bridge treatments, namely resection and percutaneous ablation, had higher probabilities of achieving a complete tumor response compared to those treated with TACE [41], whereas Huo and colleagues reported a lower incidence of dropout among patients treated with RFA *vs.* other ablation procedures [36].

However, other outcome measures between different treatments are highly variable, and a clear superiority of ablation *vs.* TACE has not emerged so far. Considering all the above studies reporting results of either bridge or downstaging strategies, the WL dropout rate ranges between 0% and 35%, the tumor progression rate ranges between 0% and 20%, the average waiting time to LT is between 4 and 12 months, the proportion of patients transplanted is between 54% and 100%, the post-LT patient survival is between 76% at 3 years and 94% at 5 years, and the intention-to-treat survival is between 57% and 94% [3,4,28–42,49,50].

These figures have to be compared with initial studies reporting an actuarial probability of dropout of 15–30% at 1 year [9,52], and with an analysis based on Organ Procurement and Transplant Network data for WL registrations for HCC between 2005 and 2008, showing that the use of any type of ablation had a minimal effect on WL dropout, which decreased from 10.1% for no ablation to 8.0% for ablation at 180 days after listing [53].

The effect of neo-adjuvant treatments on survival after LT is even more difficult to assess. Many studies indicate survival rates similar to those of untreated individuals [27,35,51,54,55], but data from the U.S. activity of liver transplant for HCC from 1997 to 2006 showed a higher 3-year post-LT survival in patients who received pre-LT ablative procedures compared to those who did not, without any apparent difference among different types of pre-LT treatments [56].

In general, studies reporting no differences between treated and untreated patients tend to have shorter waiting times (<6 months) for LT [35,51], as clearly exemplified by a national survey of LT for HCC from 1998 to 2006 by Pellettier *et al.*, where the median waiting time to LT was 2 months [54].

This explains once again the low level of recommendation pertaining to neo-adjuvant treatments given during the Zurich conference, which is mostly based on a Markov-based cost-effectiveness analysis outlining the benefits for neo-adjuvant treatments when waiting times exceed 6 months [10,11].

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Which are the patient or tumor characteristics related to an unsuccessful neo-adjuvant therapy, a higher dropout rate, and a worse post-LT outcome?

Table 1 depicts the numerically most relevant single-center series reporting both neo-adjuvant treatments used and factors affecting at least one of the main outcome measures before or after LT (response to neo-adjuvant treatment, dropout rate from the waiting list, post-transplant survival, HCC recurrence, and intention-to-treat survival). Different types of therapies were used, and untreated patients were considered in some studies [28,30,35,38–42]. Also, in some papers, a proportion of patients were treated with an intent of downstaging to eligibility criteria for LT, but most authors did not state which further criteria for downstaging were used at the initiation of the study, with only two exceptions [40,41].

As reported above, these studies showed highly variable methods and results, which are extensively described in previous reviews [13,57–59]. However, under the present need to clearly define the priority to be assigned to different stages of the disease for better balancing with other indications for LT, the most important objective is to evaluate the impact of neo-adjuvant treatments among variables affecting 5 specific outcomes: response to neo-adjuvant treatments, dropout rates from the WL, patient survival after LT, HCC recurrence after LT, and intention-to-treat survival after listing. Very few studies addressed all these issues, while most of them focused on only one or 2 outcome measures. The assessment of response to neo-adjuvant treatments was not performed with uniform criteria, which variably included radiological complete response *vs.* partial or absent response, radiological complete or partial response *vs.* no response, or pathological response (necrosis) alone. In addition, most authors did not perform univariate and multivariate analysis of all possible predictive factors. Indeed, only 2 recent studies consistently adopted this methodology [40,41].

In general, tumor dimensional and biological characteristics (more advanced tumor stage, HCC outside MC or subjected to downstaging, and elevated serum AFP) had a negative impact on the response to neo-adjuvant treatments [4,37,39–41,49,50]. These factors, together with no response to neo-adjuvant treatments, absence of neo-adjuvant treatments, and poor liver function (expressed with Model for End-stage Liver Disease [MELD] score, ascites or serum bilirubin), conditioned the probability of dropout [3,30,32,36,39–41]. Interestingly, downstaging negatively affected these outcomes in more than one reported study, and it was also a negative predictor of post-LT survival, HCC recurrence and intention-to-treat survival according to some authors [3,29].

Finally, tumor recurrence and patient survival rates were partly determined by the same factors conditioning pre-LT outcomes (including response to neo-adjuvant treatments), and partly affected by the aggressive tumor behavior, expressed by vascular invasion and poor differentiation [3,28,29,34,35,38,40–42]. Hepatitis C virus (HCV) recurrence was an independent predictor of lower patient survival in one study [41] (Table 1).

Results of studies specifically addressing downstaging

Several authors specifically focused on the results of downstaging protocols [29,34,37,43–48]. Even in these studies, a high variability of methods and results is evident. Successful downstaging was defined as fulfilling of MC [37,44,47,48], UCSF criteria [45], MC with simultaneous drop of serum AFP below 400 ng/ml [46], or 30–50% decrease in size of HCC nodule(s) [29,34].

Treatments used for downstaging included a higher prevalence of TACE alone [29,34,43,44,48], with a few studies reporting multimodal procedures [45–47] or transarterial chemoinfusion [37]. Only 2 prospective studies established *a priori* upper numerical and dimensional limits of HCC, before entering the downstaging protocol [45,46].

The average number of neo-adjuvant treatments ranges between 1 and 5, the successful downstaging rate ranges between 24% and 75%, the proportion of patients transplanted ranges between 10% and 67%, the average waiting time to LT ranges between 2 and 10.9 months, the post-LT patient survival rate ranges between 41% at 4 years and 93.8% at 5 years, the recurrence-free survival rate ranges between 40% at 5 years and 92% at 2 years (with 4 studies having a $\leqslant$3 year survival estimate), and intention-to-treat survival ranges between 19% and 84% at 3 years (with 3 studies not reporting intention-to-treat survival) [29,34,37,43–48,57–59].

During the Zurich conference, these initial and varying results of downstaging formed the basis upon which LT was strongly indicated if the expected 5-year survival is comparable to that of HCC patients who meet the criteria for LT without requiring downstaging, while no recommendation could be made for preferring a specific locoregional therapy for downstaging [10].

Can the effect of neo-adjuvant treatments be used to balance priority of HCC and non-HCC candidates?

One important aspect related to locoregional treatments in potential candidates to LT is the definition of how the response to bridge therapies might impact on WL priority, especially *vs.* non-HCC patients, in an era of persistent scarcity of organ donors.

Allocation rules for patients with HCC, waiting for LT under the MELD-based policy, are still a difficult issue in continuous evolution. Since March 2005, the UNOS policy states that patients with a T1-HCC do not receive extra MELD points whereas a MELD score of 22 is given to patients with a T2-HCC [60,61].

The UNOS priority for HCC patients was based on the replacement of the native MELD score, assessing the risk of death due to liver failure, by an estimation of the risk of tumoral progression. However, HCC patients appear to be advantaged in the current system, the role of bridge therapies in modifying both dropout rates due to tumor progression and priority policy still needs to be clarified, and the question is raised as to whether added priority is necessary [62].

In fact, the current HCC policy does not seem to adhere to the general principles for liver allocation adopted with the introduction of the MELD score: HCC patients still have better access to the liver transplant donor pool compared with non-HCC patients and local WL HCC treatments are not taken into account [62,63]. For these reasons, the development of a more dynamic score has been highly endorsed, including the role of bridge therapies [63].

Three studies reported in Table 1 appear to address these issues [39–41]. A first report from De Giorgio and colleagues in 2010 suggested that response to bridge therapies can predict tumor progression beyond the MC, and should be taken into account in models designed to prioritize organ allocation. The authors applied a center-specific allocation policy for 206 HCC

Table 1. Numerically most relevant single-center series reporting both neo-adjuvant treatments used and factors affecting at least one of the main outcome measures before or after LT (response to neo-adjuvant treatment, dropout rate from the waiting list, post-transplant survival, HCC recurrence, and intention-to-treat survival).

Author [Ref.]	NAT	Bridging (No. of patients) Downstaging (No. of patients)	Indicators of a worse outcome				
			No response to NAT	Dropout from WL	Survival after LT	HCC recurrence after LT	ITT survival
Herrero, 2001 [28]	TACE, RFA, PEI, none	Bridging (47)	n.a.	n.a.	Stage IV HCC	Stage IV HCC	n.a.
Graziadei, 2003 [29]	TACE	Bridging (48) Downstaging* (15)	n.a.	n.a.	Downstaging	n.a.	Downstaging
Yao, 2003 [30]	TACE, RFA, PEI, resection, none	Bridging (70)	n.a.	≥3 nodules, 1 nodule >3 cm No NAT	n.a.	n.a.	n.a.
Hayashi, 2004 [31]	TACE	Bridging (20)	n.a.	None	None	None	n.a.
Fisher, 2004 [32]	TACE, TACI, RFA, PEI	Bridging (33)	n.a.	AFP >400 ng/ml T3 tumor stage Bilobar HCC	n.a.	n.a.	n.a.
Maddala, 2004 [33]	TACE	Bridging (54)	Multiple nodules	None	n.a.	None	n.a.
Mazzaferro, 2004 [4]	RFA	Bridging (50)	Tumor size >3 cm	n.a.	n.a.	n.a.	n.a.
Lu, 2005 [49]	RFA	Bridging (24)	Tumor size >2.5 cm Perivascular location	n.a.	n.a.	n.a.	n.a.
Pompili, 2006 [50]	RFA, PEI	Bridging (40)	Tumor size >3 cm	n.a.	n.a.	n.a.	n.a.
Otto, 2006 [34]	TACE	Bridging (34) Downstaging* (62)	n.a.	n.a.	n.a.	No response to NAT Number of nodules Poor differentiation	n.a.
Porrett, 2006 [35]	TACE, RFA, PEI, TARE, none	Bridging (64)	None	n.a.	No response to NAT	None	n.a.
Millonig, 2007 [3]	TACE	Bridging (101) Downstaging* (15)	n.a.	No response to NAT Downstaging	No response to NAT Downstaging AFP** >11 ng/ml	Out MC Downstaging	No response to NAT Downstaging AFP** >11 ng/ml
Huo, 2008 [36]	TACE, RFA, PAI, PEI	Bridging (390)	n.a.	NAT not including RFA Tumor stage	n.a.	n.a.	n.a.
De Luna, 2009 [37]	TACI	Bridging (95) Downstaging* (27)	Out MC	Older age	None	None	None
Vibert, 2010^ [38]	TACE, RFA, cryoablation, resection, none	Bridging (153) Downstaging* (not specified)	n.a.	n.a.	≥3 nodules AFP progression >15 ng/ml/month	Age >60 yr ≥3 nodules Out MC AFP progression >15 ng/ml/month	n.a.
De Giorgio, 2010§ [39]	TACE, RFA, PEI, resection, none	Bridging (170)	Tumor stage	Ascites Total tumor diameter No response to NAT	n.a.	n.a.	n.a.
Vitale, 2010§§ [40]	TACE, RFA, PEI, resection, none	Bridging (110) Downstaging* (37)	No HCV infection Elevated AFP Tumor size Tumor stage No NAT	No response to NAT Elevated bilirubin	None	No response to NAT Tumor size Tumor number Out MC MVI Poor differentiation	No response to NAT
Cucchetti, 2011 [41]	TACE, RFA, PEI, resection, none	Bridging (262) Downstaging (53)	TACE *vs.* others Multiple nodules AFP** >400 ng/ml Downstaging	MELD score Tumor stage No NAT No response to NAT	HCV infection MVI	No response to NAT MVI	No response to NAT
Ciccarelli, 2012 [42]	TACE, RFA, PEI, resection, none	Bridging (120) Downstaging* (17)	n.a.	n.a.	n.a.	Tumor number >3 AFP** ≥400 ng/ml No NAT MVI ALA for rejection	n.a.

HCC, hepatocellular carcinoma; NAT, neo-adjuvant treatments; WL, waiting list; LT, liver transplantation; ITT, intention-to-treat; TACE, transarterial chemoembolization; RFA, radiofrequency ablation; PEI, percutaneous ethanol injection; n.a., not assessed; TACI, transarterial chemoinfusion; AFP, alpha-fetoprotein; TARE, transarterial radioembolization; MC, Milan criteria; PAI, percutaneous acetic acid injection; HCV, hepatitis C virus; MVI, microvascular invasion; ALA, antilymphocytic antibodies.
*In these studies, the inclusion criterion for downstaging was simply defined as HCC initially outside MC or University of San Francisco Criteria, without further specification, except the study by Vitale *et al.* [40], in which poor tumor differentiation detected by pre-LT biopsy was an additional exclusion criterion.
**In these studies, AFP at the moment of diagnosis of HCC was considered.
^In this study, only AFP-secreting HCCs were considered, patients dropped out were excluded, and factors related to recurrence-free survival instead of HCC recurrence were investigated.
§In this study, the time to progression beyond the MC or death was considered instead of the dropout rate.
§§In this study, dropout criteria included poorly differentiated HCC at pre-LT biopsy.

candidates (T1: 31 patients, T2: 175 patients) and compared their observed progression rate beyond MC with the expected rate applying the UNOS risk estimation. In the De Giorgio experience, percutaneous ablation represented the most frequent strategy adopted (62.8%), followed by hepatic resection (20.7%) and TACE (14.6%). Total tumor diameter and recurrence/persistence of tumor activity at the 6-week control after bridge therapy were the tumor factors that significantly correlated with dropout from the WL; unfortunately, no criteria for definition of recurrence/persistence were provided, nor were characteristics of patients in relationship to response to treatment. However, the authors reported an interesting analysis showing that the current UNOS allocation policy largely overestimates the risk of tumor progression. They also suggested a modified allocation policy in which T2-HCC patients received their natural MELD scores at the time of diagnosis with 3-point increment in the MELD score at 3-month intervals and receiving 22-point MELD exception only at the time of recurrence/persistence after bridge therapies. Even if no data were provided about the ability of different bridge approaches in obtaining tumor response, taken together, these results support a potential role of bridge therapy in reducing the dropout risk and in determining allocation priority. Finally, it should be noted that no data were provided about the post-transplant course in relationship to response to bridge therapy [39].

Some answers raised by the De Giorgio study can be found in another report published by Vitale and colleagues in the same year [40]. In a population of 147 HCC candidates (38% outside Milan criteria), a complete or partial response to bridge therapies was observed in 57.8% (using RECIST criteria) of cases. Multimodal treatment was adopted in 24% of cases, and no significant difference was observed between percutaneous ablation and TACE modalities in relationship with tumor response. In particular, RFA was adopted in 64.7% of cases with complete/partial response and in 61.3% of cases with stable/progressive disease; TACE was adopted in 22% of cases with complete/partial response and in 11% of cases with stable/progressive disease ($p = 0.068$). Thus, no suggestions can be obtained about the optimal bridge therapy to be adopted, but the authors showed, in a competing-risk analysis, that the dropout probability of patients achieving complete or partial response was significantly lower than that of their counterpart. The authors suggested, and applied, a priority score to patients with stable/progressive disease. However, prioritizing patients not responding to bridge therapies may select lesions at a higher risk for recurrence after transplantation [64]. Even if Vitale and colleagues showed that patient survival was substantially unaffected, with a 3-year post-LT survival of 83% in patients with complete/partial response and 82% in patients with stable/progressive disease, they obtained a 6-fold increase in tumor recurrence after transplantation in the latter group (from 2% to 13%; $p = 0.04$). Bridge therapy could reduce the dropout risk of HCC candidates, but if a prioritization of non-responder patients is accepted, a worsening in the post-operative outcome could be expected [40].

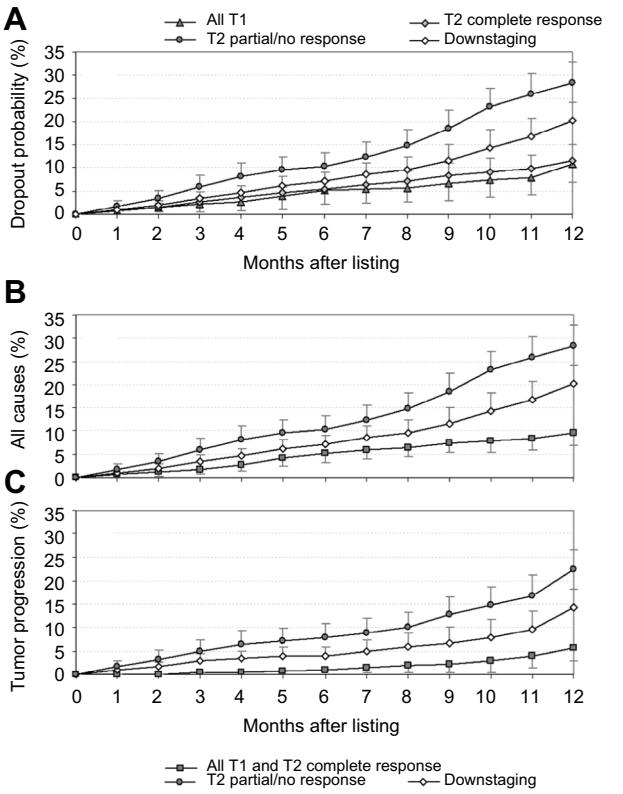
The findings of the Vitale report were made more robust by a retrospective study by Cucchetti and colleagues in 2011 [41], designed to assess the effectiveness of bridge therapy in preventing removal from the WL for tumor progression beyond the MC and in determining post-transplant outcomes. The study was conducted on data from 315 candidates, and competing-risk analysis was applied to control for transplant event.

At the 3-month control after the first bridge procedure, a complete response (assessed with mRECIST criteria) was observed in 49.1% of candidates. These patients showed a significant reduction in the dropout probability at 3, 6, and 12 months after treatment. The dropout probability was significantly affected by the MELD score, the tumor stage at diagnosis, and the response to bridge therapy.

The dropout risk for patients with T1 tumors was very similar to the risk observed for patients with T2 tumors and a complete response to bridge therapy ($p = 0.964$). Conversely, the dropout risk for patients with T2 tumors and a partial response or no response to bridge therapy was significantly higher than the risk for both T1 patients ($p = 0.001$) and T2 patients with a complete response ($p = 0.001$). The dropout risk for patients who were subjected to a downstaging procedure (T3–T4a patients) was significantly higher than the risk for all T1 patients and T2 patients with a complete response ($p = 0.024$) and was lower than the risk for T2 patients with a partial response or no response to bridge therapy ($p = 0.037$). The dropout risk of HCC patients with respect to the TNM stage, the response to bridge therapy, and the downstaging procedure is depicted in Fig. 1.

On the basis of these observations, the authors suggested that the priority of T2-HCC candidates can be reduced after successful bridge therapy, supporting the findings of De Giorgio and colleagues. Excluding hepatic resection that was related to a 4-fold increased probability of complete tumor response, TACE was the most frequent therapeutic strategy adopted (53.9%); percutaneous ablation (RFA or PEI), in combination or not with TACE, represented 26.0% of treatments. TACE was adopted in 49.3% of cases with complete response and in 57.9% of cases with partial or no response ($p = 0.177$); percutaneous ablation was adopted in 29.2% of cases with complete response and in 23.4% of cases with partial or no response ($p = 0.032$). Thus, differently from what was reported by Vitale *et al.* [40], patients treated with potentially curative bridge treatments, namely resection and percutaneous ablation, showed higher probabilities of achieving a complete tumor response. Other factors determining a lower probability of obtaining a complete response after treatment were AFP >400 ng/ml at diagnosis, multiple nodules of HCC, and inclusion in a downstaging protocol.

Focusing the analysis on transplanted patients, the authors found that patients with partial or no response to bridge therapies experienced a significantly higher recurrence-rate after



Patient at risk (No.)	At entry	3-month	6-month	12-month
All T1	40	33	28	19
T2 complete response	105	93	81	51
T2 partial/no response	117	100	85	45
Downstaging	53	43	39	16

Fig. 1. Competing risk analysis of dropout from the waiting list depending on TNM stage, downstaging procedure, and response to neo-adjuvant therapy. (A) T1 patients and T2 patients with a complete response to neo-adjuvant treatments had similar dropout risks, which were lower than the risk observed for T2 patients with a partial response or no response. (B) T1 patients and T2 patients with a complete response had a dropout risk lower than that observed for patients subjected to downstaging, whose risk was in turn lower than that observed for T2 patients with a partial response or no response. (C) The dropout risk due to tumor progression is shown according to tumor stages and responses to treatments ($p = 0.001$). (Cucchetti *et al.* [41] reprinted with permission).

transplant (5-year recurrence rate = 19.4% *vs.* 5.5% of the counterpart). The second factor affecting a lower recurrence-free survival was the presence of tumor microvascular invasion.

Patients with partial or no response to neo-adjuvant treatments had a non-significantly lower patient survival ($p = 0.098$), which was mainly influenced by HCV positivity and presence of microvascular invasion at histology. Conversely to what was concluded by Vitale in 2010, response to treatment was not suggested as a tool to give further priority to HCC patients, but to reduce it in selected T2-HCC patients. Concerning HCV–HCC patients, one consequence of this policy is that when a complete viral and tumor response is obtained after treatment, their probability of being transplanted becomes so low that they could be maintained inactive in the WL, with LT as an option in the case of future recurrent disease.

In general, and while waiting for more accurate molecular markers, the response to pre-LT treatments could represent a surrogate but extremely practical marker of tumor biology and could be helpful in the selection of candidates for LT. Especially in the case of patients with aggressive HCC (i.e., those at the upper limit of MC or UCSF criteria, or those with high or increasing levels of AFP), the test of time (3 months of observation after a locoregional treatment and subsequent re-staging) would be helpful in selecting HCC candidates at acceptable risk of HCC recurrence after LT. Importantly, this "ablate and wait" strategy [65] appears to be a valid approach also for balancing the priority to HCC or non-HCC patients on the waiting list.

Depending on the waiting time and the characteristics of the population on the list, each institution must calculate the most equitable policy for its patients. Periodical reassessment of the policy is also recommendable to avoid imbalance among candidates.

A continuous score incorporating MELD, AFP and tumor size may help prioritize HCC patients to better equate dropout rates with non-HCC patients and equalize access according to the above mentioned study by Washburn *et al.* [53,62].

In a very recent study based on the Scientific Registry of Transplant Recipients and including almost 50,000 adult candidates for LT, Toso *et al.* [66] demonstrated that the risk of dropout of HCC patients was independently predicted by MELD score, HCC size, HCC number, and AFP. They applied a proportional hazard competitive risk model, and by combining patient age and diagnosis, these factors allowed for the extrapolation of the risk of dropout. Since this model and MELD did not share compatible scales, a correlation between both models was computed according to the predicted risk of dropout, and dropout equivalent MELD (deMELD) points were calculated. The new and very complex formula is as follows:

$$\begin{aligned}\text{deMELD} = &-25 + 0.1 * \text{Age} + 1.6 * \text{MELD} \\ &+ 1.6 * \text{TumorSize} + 1.3 * \text{LogAFP} \\ &+ 6.0 \text{ if Nb Tumors} \geqslant 2 \\ &+ 0 \text{ if Diag} = \text{HCV} \\ &- 1 \text{ if Diag} = \text{HBC} \\ &+ 3 \text{ if Diag} = \text{Alcohol} \\ &+ 3 \text{ if Diag} = \text{NASH} \\ &+ 1 \text{ if Diag} = \text{Hemoc} \\ &+ 1 \text{ if Diag} = \text{Other}.\end{aligned}$$

The use of continuous variables, the evaluation of HCC and non-HCC patients with uniform criteria, and the "dynamic" concept of the revised MELD are the major advancements related to this equation. In particular, deMELD can be recalculated at any time and has the ability to capture changes of the risk of dropout from the WL, for example as a consequence of neo-adjuvant treatments. In this perspective, lesions with complete radiological response should not be counted as nodules, while those with partial or absent response should be considered with their maximum baseline diameter.

By applying this formula, the authors demonstrated that the current 22 points allocated to T2 patients in the US represent too many exception points, so that 95.2% of T2 HCC patients are unfairly advantaged as compared with the deMELD.

Are there new molecular markers for a better estimation of tumor biological behavior and/or response to treatment?

Serum markers such as AFP or protein induced by vitamin K absence or antagonism factor II increase predictive accuracy if

used in addition to morphological features of HCC [46,67–69], probably because they are reliable indicators of tumor biology, including microvascular invasion and tumor grade.

New specific molecular signatures or markers in HCC or in the adjacent hepatic tissue have been shown to correlate with outcome and to predict the risk of tumor recurrence after treatment [70]. Among these factors, epithelial cell adhesion molecule (EpCAM) [71,72], the g3-proliferation subclass [73], the expression status of the miR-26 miRNA precursor [74], two gene prognostic signature in non-tumor hepatic tissue [75,76], and fractional allelic imbalance have been described [77]. These molecular markers could be assessed in patients undergoing locoregional treatments in order to predict the risk of dropout and tumor recurrence after LT [18]. However, they require external validation before they can be used in the clinical practice.

Conclusions

According to the present evidence, the following concepts should be applied for the use of neo-adjuvant treatments in candidates for LT for HCC:

(1) A recent consensus conference established that neo-adjuvant treatments are indicated in patients considered for LT if the waiting time for LT is expected to be longer than 6 months. However, the widespread policy of treating most patients in the WL, especially those at risk of tumor progression, seems justified given the often unpredictable waiting time for LT and the opportunity to use the response to therapy as a prioritization parameter (see also conclusion 4).

(2) The response to treatments should be evaluated with mRECIST criteria and through a 3-month periodical reassessment, including radiological imaging and serum AFP sampling.

(3) The type of locoregional treatment should include TACE and standard ablation techniques (RFA, PEI), to be chosen according to the BCLC scoring system; ablation techniques can be applied laparoscopically, if permitted by the degree of liver function; liver resection can be used as bridging or downstaging procedure in BCLC 0-A stages in experienced centers; other procedures (TACE-DEB, TARE, radiotherapy, microwave ablation, cryoablation, and irreversible electroporation), though promising, should still be considered under investigation.

(4) Neo-adjuvant treatments, especially if applied as multimodal approach, have the 3 main purposes of (A) effectively controlling HCC for expected long waiting times; (B) identifying patients with high or low probability of cancer progression; (C) based on these risk categories, helping in balancing the priority of HCC and non-HCC-candidates through revised additional scores for HCC.

(5) Due to the low number of prospective studies with well-defined entry criteria and the high variability of results, the role of downstaging is still to be defined with large, multicenter trials.

(6) Although new molecular markers for a better estimation of tumor biological behavior and/or response to treatment have been investigated with promising results, at present, they cannot be translated into a routine use in clinical practice.

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Conflict of interest

The authors declared that they do not have anything to disclose regarding funding or conflict of interest with respect to this manuscript.

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