Randomised controlled trial

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Pembrolizumab in combination with LEnvatinib in participants with hepatocellular carcinoma before liver transplant as Neoadjuvant TherapY—PLENTY pilot study

Zicheng Lv, MD[a,b], Xuelin Xiang, Ph.D[c], June-kong Yong, MD[a], Yi Zhou, MS[c], Yichi Wu, MD[c], Linman Li, MS[c], Yuanhao Wang, Ph.D[c], Zijie Zhang, Ph.D[c], Qiang Xia, MD, Ph.D[a,c,*], Hao Feng, MD, Ph.D.[a,b,c,*]

Background: The high recurrent rate after liver transplantation (LT) remains a clinical challenge, especially for those exceeding the Milan criteria (MC) and with high RETREAT scores. Therefore, the authors aim to investigate whether neoadjuvant systemic therapy allows safely administered and effectively reduces post-LT recurrence for those patients.

Methods: In this prospective, randomized, open-label, pilot study, patients with HCC exceeding the MC were randomly assigned to PLENTY or control group before LT. The primary endpoint of the study was the recurrence-free survival after LT.

Results: Twenty-two patients were enrolled and randomly assigned: 11 to the PLENTY group and 11 to the control group. The 30-month tumor-specific RFS was 37.5% in the PLENTY group and 12.5% in the control group. The 12-month tumor-specific RFS after LT was significantly improved in the PLENTY group (87.5%) compared to the control group (37.5%) ($P = 0{\cdot}0022$). The objective response rate in the PLENTY group was 30 and 60% when determined by RECIST 1.1 and mRECIST, respectively. Six patients (60%) had significant tumor necrosis, including three (30%) who had complete tumor necrosis at histopathology. No acute allograft rejection after LT occurred in the PLENTY and Control group.

Conclusion: Neoadjuvant pembrolizumab plus lenvatinib before LT appears to be safe and feasible, associated with significantly better RFS for patients exceeding the MC. Despite the limitations of small sample size, this is the first RCT to evaluate neoadjuvant PD-1 blockade combined with tyrosine kinase inhibitors in LT recipients, the results of this study will inform future research.

Keywords: hepatocellular carcinoma, immunotherapy, liver transplantation, neoadjuvant therapy, recurrence-free survival

Introduction

Hepatocellular carcinoma (HCC) is a leading cause of cancer-related death worldwide[1]. Liver transplantation (LT) is indeed become the most effective treatment for HCC as it not only eliminates the tumors but also the diseased liver with the underlying hepatocarcinogenic factors. Even so, recurrence is still inevitable for some patients; the 24-month cumulative incidence of recurrence is as high as 24.2%[2]. The Milan criteria (MC) are the most common criteria for LT in HCC patients[3], but in recent years, significant efforts have been made to modify selection criteria with the goal of maximizing transplant benefits for patients with HCC. However, it also results in an increase in the risk of tumor recurrence[4]. Thus, many centers began to employ tumor down-staging (DS) or bridging strategies with locoregional therapy (LRT) for LT to reduce the viable tumor burden to meet acceptable LT criteria. LRT-DS patients had superior RFS (60 vs. 54%) compared with not being down-staged[5].

Recently, the treatment landscape of advanced HCC has been evolving rapidly, notably, there are no guidelines or consensus on

[a]Department of Liver Surgery, Renji Hospital (Punan Branch), School of Medicine, Shanghai Jiao Tong University, [b]Clinical Research Unit, Renji Hospital, School of Medicine, Shanghai Jiao Tong University and [c]Shanghai Engineering Research Centre of Transplantation and Immunology

Zicheng Lv, June-kong Yong, and Xuelin Xiang contributed equally to this work.

Sponsorships or competing interests that may be relevant to content are disclosed at the end of this article.

*Corresponding authors. Address: Department of Liver Surgery, Renji Hospital, School of Medicine, Shanghai Jiao Tong University, Shanghai 200127, People's Republic of China. E-mail: surgeonfeng@live.com (H. Feng); Department of Liver Surgery, Renji Hospital, School of Medicine, Shanghai Jiao Tong University, Shanghai 200127, People's Republic of China. Tel.: +86 21 68383990, fax: +86 21 58752345. E-mail: xiaqiang@medmail.com.cn) (Q. Xia).

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the application of immune checkpoint inhibitors (ICIs) with tyrosine kinase inhibitors (TKIs) before LT due to safety concerns of acute graft rejection[6–8]. Our previous research[9] preliminarily proved that the combination of PD-1 blockades with lenvatinib and subsequent LT were feasible and effective under close monitoring. We assume that ICIs combined with lenvatinib as neoadjuvant therapy prior to LT could reduce the risk of post-LT recurrence and improve the RFS as well as OS rate in patients with HCC. Therefore, we assigned the prospective, pilot study to investigate whether pembrolizumab plus lenvatinib as a neoadjuvant therapy before LT can be safely administered and effectively reduce post-LT recurrence for patients with HCC exceeding the MC. This trial is registered with ClinicalTrial.gov.

HIGHLIGHTS

- Up to now, only 25 cases, including seven patients in our center, underwent neoadjuvant immunotherapy before liver transplantation (LT) was published, and neither high-quality trial nor evidence of long-term prognosis has been published.
- Neoadjuvant therapy with pembrolizumab and lenvatinib yielded favorable objective response rates and significantly improved recurrence-free survival without increasing graft rejection after LT.
- This is the first randomized controlled trial to evaluate neoadjuvant PD-1 blockade combined with tyrosine kinase inhibitors in LT recipients with hepatocellular carcinoma presenting beyond the Milan criteria, provides more evidence of the efficacy and safety of the neoadjuvant systematic treatment, especially the superior recurrence-free survival.

Methods

Study design and participants

PLENTY study was a prospective, pilot study designed to investigate the efficacy and safety of pembrolizumab plus lenvatinib as pre-LT neoadjuvant therapy for patients with HCC beyond MC and without extrahepatic spread, conducted at the department of Liver Surgery. Patients aged 18–80 years with HCC exceeding MC and planned for LT were eligible for inclusion. Major inclusion criteria included Eastern Cooperative Oncology Group (ECOG) performance status 0-1, Child-Pugh A-B7 liver score (5 to 7 points), and adequate organ function. A full list of eligibility criteria was referred to in the Supplement protocol (Supplemental Digital Content 1, http://links.lww.com/JS9/C976). The study was reviewed and approved by the Ethical Committee of Hospital. Written informed consent on study aims, participation requirements, and the right to refuse was obtained from all participants. The trial was conducted in accordance with the principles of the Declaration of Helsinki and Good Clinical Practice. All procedures were in accordance with the STROBE guidelines. The work has been reported in line with Consolidated Standards of Reporting Trials (CONSORT) Guidelines[10,11].

Study procedures

Following written informed consent and completion of baseline assessments, patients deemed suitable candidates for the study were enrolled and subsequently randomized into the two groups with a ratio of 1:1. Randomization was performed at the Clinical Research Unit of Hospital using the computer-generated random number code. The randomization codes were concealed in sequentially numbered opaque envelopes by the clinical research nurse, who was not involved in data analysis. In addition, the clinical research coordinators were in charge of randomly assigning the patients to different groups based on allocation sequence. No masking was applied to clinical outcome assessors.

Interventions

Patients who assigned to PLENTY group received 200 mg of pembrolizumab intravenously every 3 week until ~6 weeks before LT according to pharmacokinetic concentration-time profiles of pembrolizumab[12], or unacceptable toxicity developed. According to the estimated LT waiting time of 2 months in our center [mostly, when the patient was ranked fourth (blood type O) to seventh (blood type AB) on the waiting list], the pembrolizumab was discontinued. Simultaneously, lenvatinib was given orally at 8 mg once a day until 1–2 weeks before LT. Patients assigned to the control group received LRT such as transarterial chemoembolization (TACE), etc. Both groups received the best supportive care at the discretion of the investigator. The tumor stage was evaluated with enhanced CT or MRI scan, and restaging scans were done at least every 6 weeks until LT for patients. Tumor necrosis on pathological examination was assessed by consensus of two dedicated hepatopathologists (F.H. and L.Z.B.) who visually estimated and agreed upon the percentage of necrosis seen within the resected tumor bed.

All the patients underwent donor after cardiac death whole graft orthotopic LT[13], including a tapered dose of methylprednisolone, a drug regimen of tacrolimus, and mycophenolate mofetil. Two more days longer for those patients using pre-LT pembrolizumab in the methylprednisolone tapering schedule. Gradual mycophenolate mofetil withdrawal was assessed on a case-by-case basis; the tapering-off time was generally controlled within 2 months after LT. Sirolimus was administrated 2 mg once a day from ~6–8 weeks after LT[14]. For patients who relapsed more than 6 months after LT, lenvatinib was reused until progression.

Follow-up protocol

Median follow-up was 37.1 months (range, 26.2–45.6). Enhanced CT or MRI scan was done every 6–8 weeks in the first year after LT and at least every 12 weeks after 1 year. Diagnosis of recurrence was assessed by two trained and experienced radiologists (Z.Z.G. and Q.L.J.) or by pathology consistent with HCC if suspected of recurrence but could not be confirmed by imaging. Patients with tumor recurrence received optimal treatment assigned by the multidisciplinary team in our center.

Study measures

The primary endpoint of the study was the recurrence-free survival (RFS) after LT between the two groups. RFS was defined as the time from LT until the first documentation of HCC recurrence (local, regional, or distant), which was assessed by two trained and experienced radiologists (Z.Z.G. and Q.L.J.) or by pathology consistent with HCC if suspected of recurrence but could not be confirmed by imaging, or death due to any cause (both cancerous

and noncancerous causes of death). Secondary endpoints included (1) overall survival (OS, from randomization to death, and after LT); (2) objective response rate (ORR, according to Response Evaluation Criteria in Solid Tumors [RECIST] version 1.1 and modified RECIST [mRECIST][15]), defined as the proportion of patients with a complete response or partial response (PR); (3) adverse event, defined as any unfavorable and unintended sign, symptom, or disease (new or worsening) associated with the therapy. Adverse events were graded according to the Common Terminology Criteria for Adverse Events (CTCAE, version 5.0). The exploratory endpoints were immunological/biomarker changes in the peripheral blood and significant tumor necrosis, defined as more than 70% necrosis of resected tumor under pathological examination[16].

Sample size calculation and statistical analysis

Survival analyses with the Kaplan–Meier method and log-rank test were done to assess various time-to-event endpoints. Proportions of patients with an overall response (according to RECIST 1.1 and mRECIST) were estimated along with 95% CIs. R version 4.2.0 was used for the statistical analysis. This study is registered with ClinicalTrials.gov.

Concerning the sample size of the pilot study, since no previous reference on RFS or OS after LT in patients who received ICIs before LT is available, the sample size for this pilot study was determined by ORR. In the previous retrospective study, the ORR who received ICIs before LT was 70%[9]. At 90% power and a 5% significance level, 20 patients (10 patients in each group) were recruited for this pilot study. Data analysis was performed when 20 patients completed the liver transplant procedure.

Results

Baseline and waitlist characteristics

A total of 22 patients with HCC beyond MC were enrolled between 3 February 2020 and 5 September 2021. All patients received protocol-assigned treatment except one patient in the PLENTY group failed to be followed up after four cycles of combined therapy when ready to undergo LT. He was found to recontact 15 months later and showed a PR, thus, he was excluded from the efficacy and safety analysis (Fig. 1). Randomization provided a reasonable balance of demographic and clinical factors in both groups (Table 1). Individual details are provided in the Supplement Tables (Supplemental Digital Content 2, http://links.lww.com/JS9/C977). One patient in the control group died from tumor progression while on the waiting list (Fig. 1). No LRT was received during the study period for the treatment arm patients. Each arm included four patients with a history of LRT prior to enrollment (more than 3 months before enrollment). Finally, 20 patients received LT, and the median time on the waiting list was longer in the PLENTY group compared to the control group ($P < 0.001$; Table 1) due to four cycles (range, 2–5) of combined therapy, and a washout period of 60.5 days (range, 25–193) for pembrolizumab.

Recurrence and survival

Until November 2023, tumor recurrence after LT was observed in eight patients (seven in the control group and one in the PLENTY group) during a median follow-up of 33.4 months (range, 23.1–45.0), and three patients in the control group died from tumor progression. The median RFS after LT was 12.4 months (95% CI: 5.63-NA).

RFS was significantly improved in patients who received neoadjuvant pembrolizumab plus lenvatinib treatment compared with those in the control group (log-rank $P = 0{\cdot}017$; Fig. 2A). The 12-month RFS rate was 70% (95% CI: 34.8–93.3) in PLENTY group and 30% (95% CI: 6.67–65.2) in the control group, respectively. Concerning the 12-month tumor-specific RFS, it was 87.5% in the PLENTY group and 37.5% in the control group ($P = 0{\cdot}0022$; Fig. 2B). The 30-month tumor-specific RFS was 37.5% in the PLENTY group and 12.5% in the control group.

During a median follow-up of 37.1 months after randomization, two patients in the PLENTY group and five patients in the control group died. One patient in the PLENTY group died from GVHD 43 days after LT. He had a 51-day washout period before LT, and GVHD was diagnosed 18 days after LT. The patient developed a mild decrease in white blood cells and hyponatremia on day 18 after LT. On postoperative day 23, the patient developed skin rash, accompanied by fever and dysphagia. Methylprednisolone, rucotinib, balliximab, and immunoglobulin are used to treat the disease. The liver function indexes such as transaminase and bilirubin did not appear abnormal during the course of treatment, and immune related hepatitis was excluded. The diagnosis of GvHD was confirmed by skin biopsy. The fatal adverse event was evaluated as unlikely to be associated with pembrolizumab according to the WHO-UMC system by the multidisciplinary team. Another patient in the PLENTY group died from intra-abdominal coagulopathic hemorrhage 6 days after LT. Neither of the deaths was considered to be related to the neoadjuvant treatment.

No statistical difference was observed in OS between the two groups (Fig. 2C, D). To gain further insight into the durability of response, swimming plots for each patient were generated, including the time points of combined therapy, LT, recurrence, and death (Fig. 2E, F).

Tumor responses

The average Risk Estimation of Tumor Recurrence After Transplant (RETREAT) score was 5 in the PLENTY group and 4.9 in the control group from recruitment, respectively. And the RETREAT score at LT was 4.8 in the PLENTY group and 4.9 in the control group, respectively. Radiological response before LT was evaluated in 10 patients in the PLENTY group. Thirty percent of patients had a PR and 70% had stable disease (SD) according to RECIST 1.1. While as for mRECIST, 10% of patients had a complete response, 50% had a PR, and 40% had SD. Thus, the ORR was 30% (95% CI: 6.67–65.2) and 60% (95% CI: 26.2–87.8) when determined by RECIST 1.1 and mRECIST, respectively. The median time to response was 63 days (range, 45–88) per mRECIST. Spider plots and waterfall plots of patients' target lesion changes from baseline were presented in Figure 3. Pathological response to the neoadjuvant therapy was also evaluated after LT in 10 patients in the PLENTY group; six patients had significant tumor necrosis (60%, 95% CI: 26.2–87.8), including three who had complete tumor necrosis (30%, 95% CI: 6.67–65.2) at histopathology. Decreasing serum alpha-fetoprotein (AFP) concentrations, AFP-L3 percentages, and des-gamma-carboxyprothrombin (DCP) concentrations

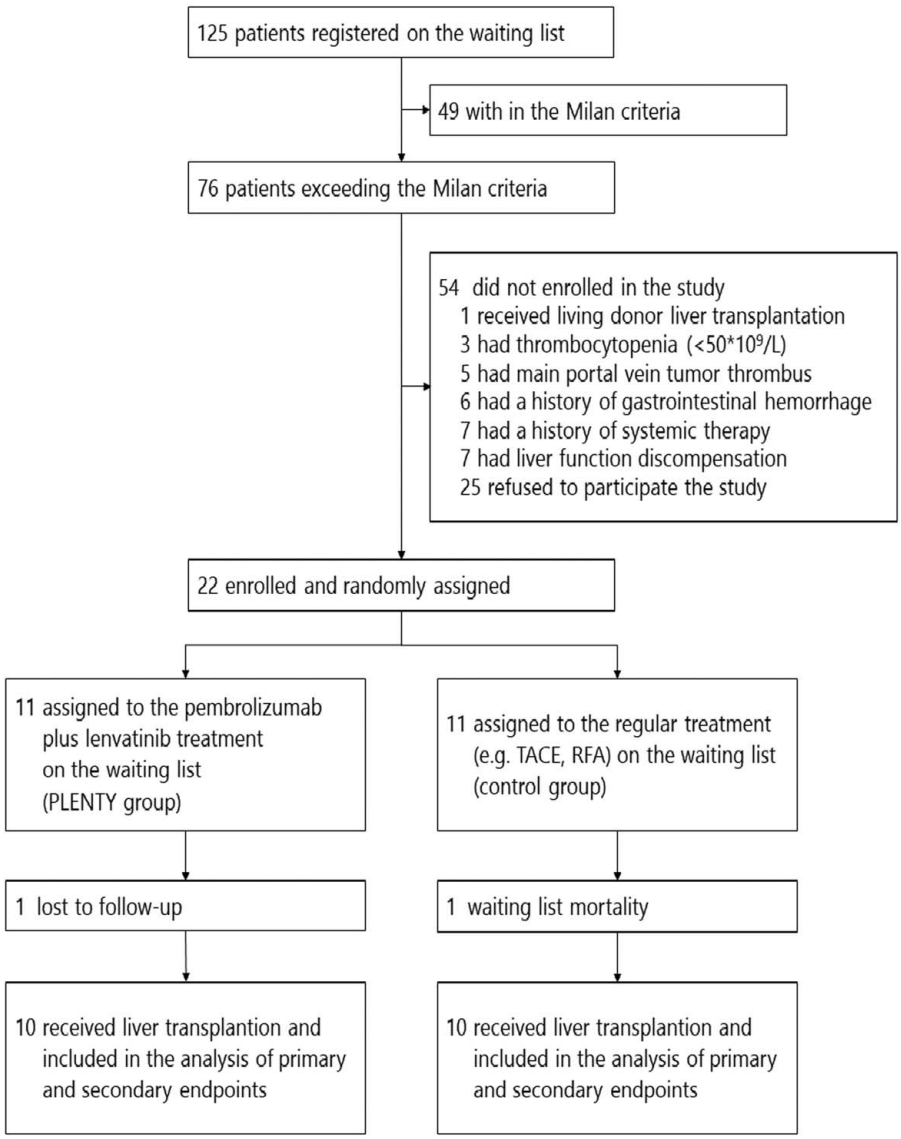

Figure 1. Trial profile.

were observed in most patients [8 (80%), 7 (70%), and 6 (60%) of 10, respectively] in the PLENTY group. However, there was no correlation between any biomarker changes over time and pathological responses (Supplement Figure, Supplemental Digital Content 3, http://links.lww.com/JS9/C978).

Adverse events

All 10 patients who received pembrolizumab plus lenvatinib treatment were included in the safety analysis. The most common adverse events were hypertension (80%), elevated transaminases (60%), diarrhea (50%), and thrombocytopenia (50%). Grade 3 adverse effects were observed in 3 (30%) patients: one had fatigue, one had diarrhea, and one had pruritus. No grade 4 or 5 adverse events were observed (Table 2). There were no treatment discontinuation or surgical cancellations due to treatment-related adverse events (TRAEs). No acute allograft rejection after LT occurred in either group.

Extension neoadjuvant ICI cohort

Up to November 2022, a total of 24 patients with HCC beyond MC had received LT after neoadjuvant ICIs with or without TKIs in our center (Fig. 4 and Table 3), including 14 extra patients during the same period in our center. Acute rejection was found in three patients, one patient was confirmed by biopsy, the other two were considered clinical suspected acute rejection due to elevated transaminases, and all of them was responded well from methylprednisolone. No allograft loss occurred (Supplementary Table 3, Supplemental Digital Content 2, http://links.lww.com/JS9/C977).

The OS rate was 33.3% and the RFS was 29.2% for these 24 patients. This is one of the largest single-center sample all over the world for neoadjuvant ICIs prior to LT, proving the application of ICIs could be a feasible and effective DS protocol for advanced HCC before LT.

Table 1

Demographics and clinical characteristics.

Characteristics	PLENTY ($n=10$)	CONTROL ($n=11$)	P
Age at randomization, years	57.5 (38–68)	51 (38–66)	0.440
Sex			0.944
Male	9 (90.0%)	10 (90.1%)	
Female	1 (10.0%)	1 (9.1%)	
ECOG performance status			0.329
0	10 (100%)	10 (90.1%)	
1	0 (0)	1 (9.1%)	
HBV background	10 (100%)	11 (100%)	/
Maximum tumor size (cm)	7.01 (1.655–16.94)	7.5 (2.3–14.5)	0.446
Number of lesions	3.5 (1–23)	3 (1–12)	0.753
PVTT			0.835
Vp0	5 (50.0%)	5 (45.5%)	
Vp1-3	5 (50.0%)	6 (54.5%)	
BCLC stage			0.528
B	5 (50%)	4 (36.4%)	
C	5 (50%)	7 (63.6%)	
Alpha-fetoprotein			0.466
≤ 400 ng/ml	3 (30.0%)	5 (45.5%)	
> 400 ng/ml	7 (70.0%)	6 (54.5%)	
Des-γ-carboxyprothrombin			0.890
≤ 400 mAU/ml	7 (70.0%)	8 (72.7%)	
> 400 mAU/ml	3 (30.0%)	3 (27.3%)	
ALT (U/l)	47 (23–88)	53 (21–111)	0.841
AST (U/l)	72 (21–99)	55 (22–162)	0.479
Child-Pugh class			0.217
A	8 (80.0%)	6 (54.5%)	
B	2 (20.0%)	5 (45.5%)	
Waiting time (Day)	114 (89–256)	33 (15–63)	0.001
Waiting list mortality			0.329
No	10 (100%)	10 (90.1%)	
Yes	0 (0)	1 (9.1%)	
Treatment cycles	4 (2–5)	/	/
Washout period (Day)	60.5 (25–193)	/	/

Data are *n* (%) or median (range).

ALT, alanine transaminase; AST, aspartate transaminase; BCLC, Barcelona Clinic Liver Cancer; ECOG, Eastern Cooperative Oncology Group; HBV, Hepatitis B Virus; LRT, loco-regional therapy; PVTT, portal vein tumor thrombus; TB, total bilirubin.

Discussion

The high recurrent rate of HCC after LT remains a considerable clinical challenge. The application of neoadjuvant ICIs before LT in patients with advanced HCC has been explored since 2019 in our center. We reported here, to our knowledge, for the first RCT providing evidence of the benefit of neoadjuvant systemic therapy in patients with HCC who are planned for LT. Although several clinical trials are ongoing, the results of these studies are not yet available (Supplementary Table 2, Supplemental Digital Content 2, http://links.lww.com/JS9/C977). This prospective trial was designed based on our retrospective study[9] and a series of literature review[17–22].

Downstaging treatments before LT have mostly been limited to LRT, including TACE, RFA, transarterial radioembolization (TARE), and combined different treatments[5,23,24]. The probability of successful DS to meet MC for patients within UNOS-DS criteria[23], was 89 and 86% with TACE and TARE, respectively[24]. However, the neoadjuvant treatment using a combination of sorafenib and TACE before LT had not significantly improved time-to-progression (71 vs. 85 days), ORR (20.8 vs. 26.9%) or DCR (66.7 vs. 73.1%) compared to TACE alone[25]. Several neoadjuvant treatment strategies with lenvatinib, instead of sorafenib[26,27], with higher DS intent that permit subsequent salvage hepatectomy due to its greater antiangiogenic effect, have been reported[28,29]. In addition, the P-161 Trial (NCT05171335), in order to explore the efficacy and safety of neoadjuvant therapy of lenvatinib plus TACE for transplant-eligible patients with large HCC is enrolled. However, high-quality evidence of pre-LT neoadjuvant systematic therapy still needs to be provided (Supplementary Table 4, Supplemental Digital Content 2, http://links.lww.com/JS9/C977).

Although the tumor was completely removed with the diseased liver, the invisible circulating tumor cells (CTCs) are believed to become the seeds of tumor recurrence in the immunosuppressive microenvironment after LT. The preoperative CTCs level has become a valuable predictor of recurrent HCC after LT[30,31]. LRT was unsatisfactory in eradicating CTCs[32,33]. More recently, a multicenter study showed that CTC levels decreased after immunotherapy in patients with metastatic renal cell carcinoma[34,35]. However, in consideration of the potential risk of post-LT rejection induced by ICIs and delays in surgery due to TRAEs, the clinical data regarding peri-operative administration of ICIs in the context of solid organ transplantation are scarce. A meta-analysis including 52 patients treated with ICIs after LT[36] discovered that acute rejection occurred in 28.8% of patients, and nearly half of them died because of graft loss. Compared to postoperative, the preoperative application of ICIs might be safer because of a controllable washout period in LT recipients. Up to now, only 25 cases of patients with HCC receiving ICIs prior to LT have been published[9,17–22]. Twelve percent had suffered severe rejection and allograft loss; two patients died[17,18], and one survived with retransplantation[19]. Besides, 16% of patients had mild to moderate rejection.[9,19,20] Schnickel and Sogbe suggested that ICIs should be terminated at least 3 months before LT[19,22]. In this pilot study, the median washout period was 2 months (range, 25–193 days); five patients (50%) had an even shorter washout period without any rejection after LT, indicating that the termination time point of ICIs needed further exploration. Referring to the prescribing information for pembrolizumab for intravenous injection, the terminal half-life (t1/2) of the drug is documented to be 22 days. Recent studies have demonstrated that the washout period for the immunotherapy agent pembrolizumab must exceed a minimum duration of 1 month to mitigate the risk of graft rejection. A literature review regarding the washout period for pembrolizumab was shown in Supplementary Table 5 (Supplemental Digital Content 2, http://links.lww.com/JS9/C977).

In the extension cohort, the median washout period was 73 days. Surprisingly, biopsy proved acute rejection (BPAR, RAI = 5) was found in the patient underwent camrelizumab treatment and the washout period was 106 days. The patient underwent tislelizumab with the washout period of 18 days and the patient underwent nivolumab with the washout period of 70 days experienced clinical suspected acute rejection (Supplementary Table 3, Supplemental Digital Content 2, http://links.lww.com/JS9/C977).

TACE and stereotactic body radiotherapy followed by TKI-ICI is also expected to be potential neoadjuvant therapy options for HCC LT. Combination therapies with locoregional therapies have also been actively explored to enhance ICI efficacy by promoting the release of tumor-associated antigens and cytokines.

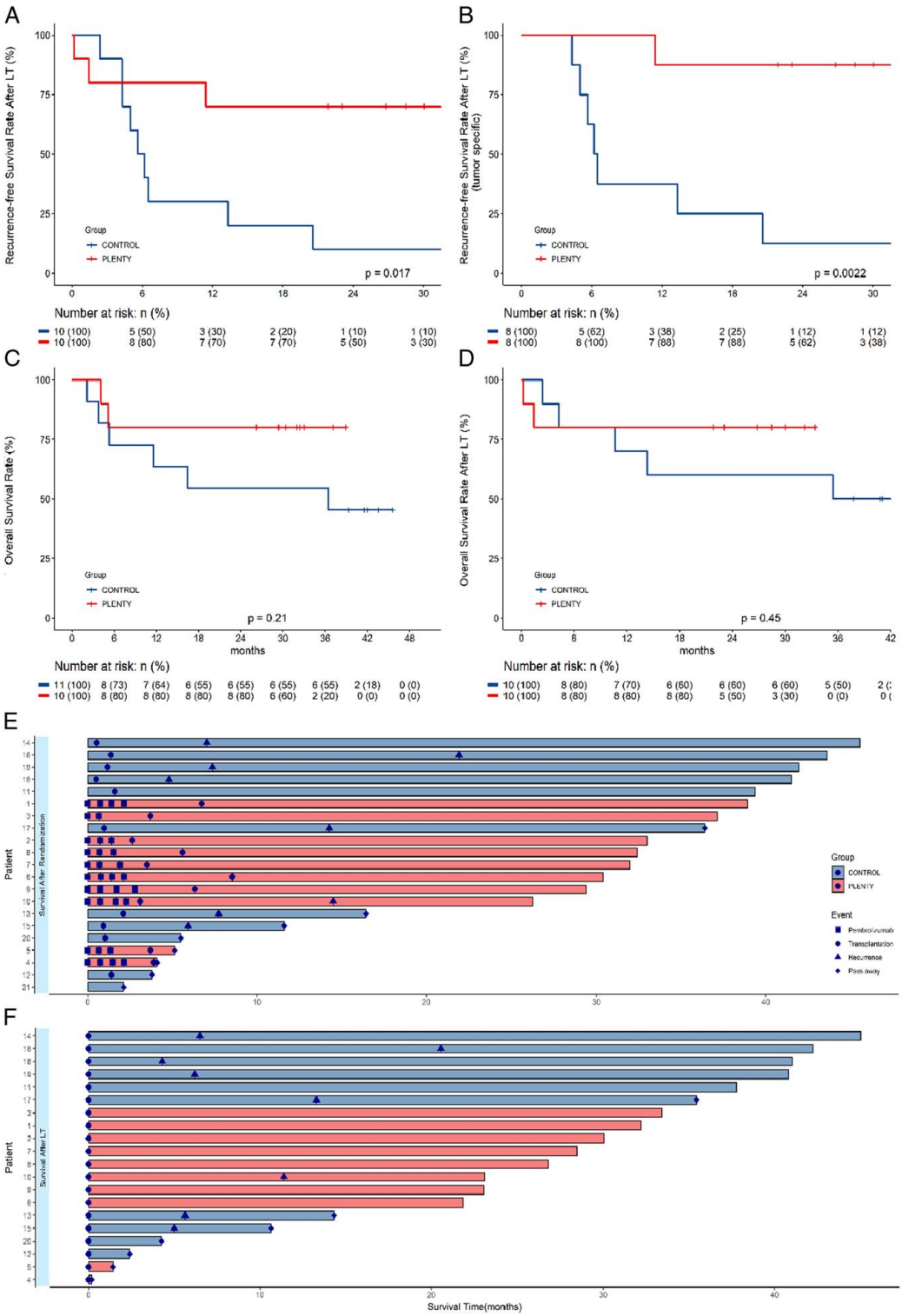

Figure 2. Survival and recurrence outcomes (A) Recurrence-free survival after liver transplantation (LT), per patient ($n = 20$). (B) Tumor-specific recurrence-free survival after LT, per patient ($n = 16$). (C) Overall survival after randomization, per patient ($n = 21$). (D) Overall survival after LT, per patient ($n = 20$). (E) Swimmer plot showing the time to treatment, recurrence and pass away after randomization. (F) Swimmer plot showing the time to recurrence and pass away after LT.

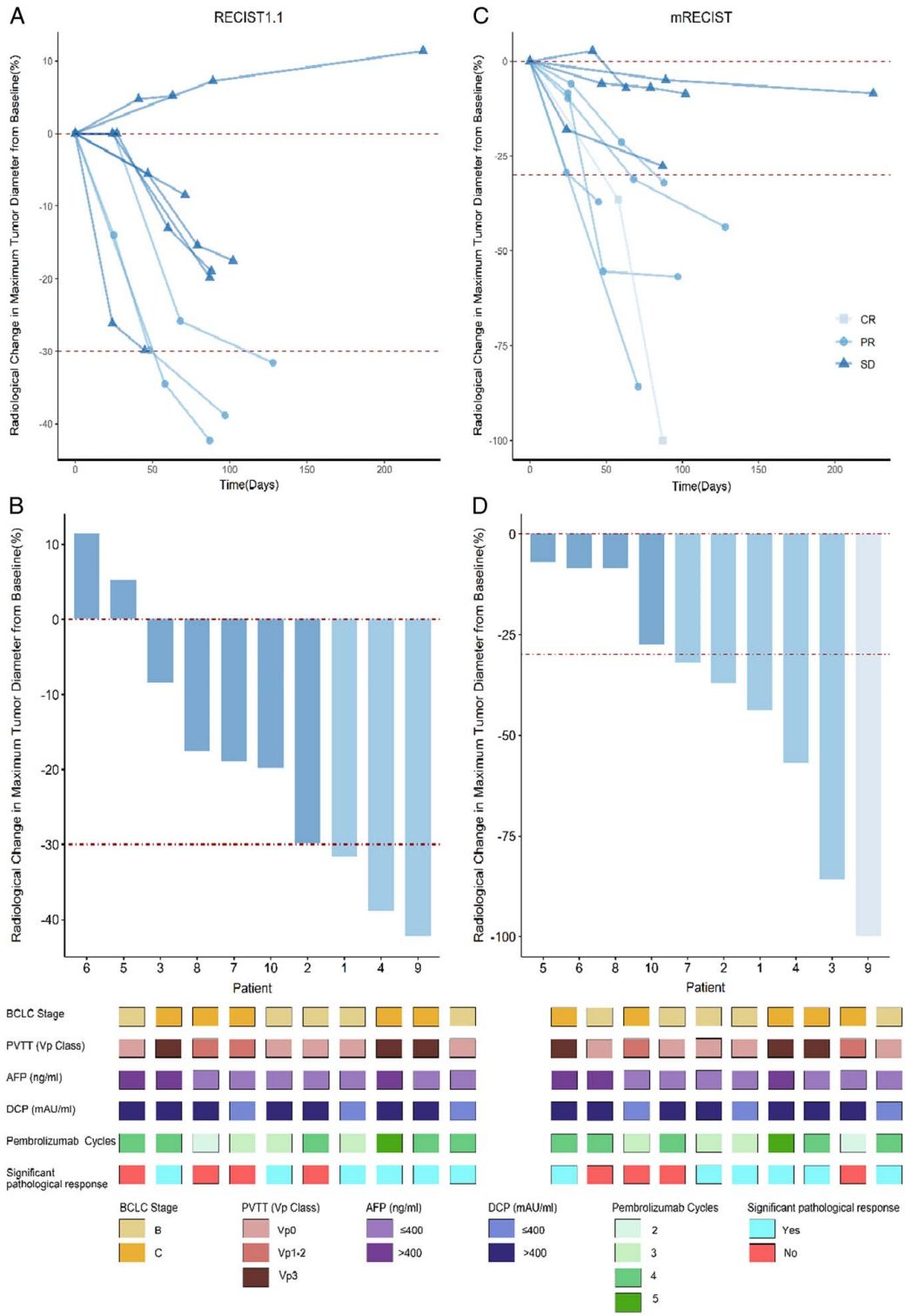

Figure 3. Tumor responses (A) Spider plot showing the percentage change from baseline in maximum diameter of target lesions over time (months) in each of the 10 patients in the PLENTY group, according to treatment response per RECIST 1.1. (B) Waterfall plot showing the percentage change from baseline in maximum diameter of target lesions in each of the 10 patients in the PLENTY group, according to treatment response per RECIST 1.1. (C) Spider plot showing the percentage change from baseline in maximum diameter of target lesions over time (months) in each of the 10 patients in the PLENTY group, according to treatment response per mRECIST. (D) Waterfall plot showing the percentage change from baseline in maximum diameter of target lesions in each of the 10 patients in the PLENTY group, according to treatment response per mRECIST.

Table 2

Treatment-related adverse events and serious adverse events.

Adverse events	Any grade	Grade 1	Grade 2	Grade 3	Grade 4–5
Any	10 (100%)	8 (80%)	5 (50%)	3 (30%)	0
Hypertension	8 (80%)	8 (80%)	0	0	0
Elevated transaminases	6 (60%)	5 (50%)	1 (10%)	0	0
Diarrhea	5 (50%)	2 (20%)	2 (20%)	1 (10%)	0
Thrombocytopenia	5 (50%)	3 (30%)	2 (20%)	0	0
Elevated bilirubin	4 (40%)	3 (30%)	1 (10%)	0	0
Fatigue	3 (30%)	0 (0)	2 (20%)	1 (10%)	0
Pruritus	2 (20%)	0	1 (10%)	1 (10%)	0
Proteinuria	2 (20%)	0	2 (20%)	0	0
Neutropenia	2 (20%)	1 (10%)	1 (10%)	0	0
Pneumonitis	2 (20%)	1 (10%)	1 (10%)	0	0
Abdominal pain	2 (20%)	1 (10%)	1 (10%)	0	0
Elevated pancreatic enzymes	2 (10%)	2 (10%)	0	0	0
Elevated creatinine	2 (20%)	2 (20%)	0	0	0
Anemia	1 (10%)	1 (10%)	0	0	0

Data are *n* (%) and represent the highest grades assigned.

The ORR were reported higher in the TACE-TKI-ICI group compared to TKI-ICI group for unresectable HCC[37,38]. In the CHANCE001 real-world study, 556 advanced HCC patients receiving either TACE-TKI-ICI or TACE monotherapy. ORR were significantly higher in the combination group (60.1 vs. 32.0%), though Grade 3/4 adverse events rate were also elevated (15.8 vs 7.5%)[39].

Our study results tentatively suggest that although immunotherapy in LT recipients is associated with an ineligible risk of rejection and even mortality, it should not be discarded in this particular fragile population. In the present study, grade 3 TRAEs were observed in 30% of patients, similar to previous reports[8,40], and did not increase the mortality, delay, or cancellation of LT waiting list by TRAEs.

The RFS benefit from neoadjuvant systemic therapy in patients with HCC has been preliminarily proved in hepatectomy. The 12-month RFS after hepatectomy was about 61.5–75% in patients with initially unresectable HCC who had received ICIs plus TKIs as conversion therapy; furthermore, achieving a pathological complete response to systemic therapy was associated with a favorable RFS after resection[35,41]. Our results showed that neoadjuvant pembrolizumab and lenvatinib could improve RFS with a low incidence of TRAEs. Despite the small sample size, it is notable that RFS differed significantly between the two groups, regardless of whether excluded 'nontumor-related' deaths ($P = 0.03$, $P = 0.0057$). The predefined primary endpoint of RFS was met, which could support further studies to investigate the efficacy of these regimens[42,43].

Cucchetti *et al.*[44] revealed that 'tumor-related death' after LT was not only correlated with tumor number and tumor size but also correlated with the effect of neoadjuvant therapy evaluated by mRECIST. Several studies have demonstrated that the level of CTCs observably decreased in patients with well-radiological response[45,46]. In our study, no patient had progressive disease after neoadjuvant therapy, although they were still exceeding the MC. Nonetheless, the goal of reducing the risk of post-LT recurrence was achieved, maybe because of the necrosis of the intrahepatic lesions and the decrease in CTCs.

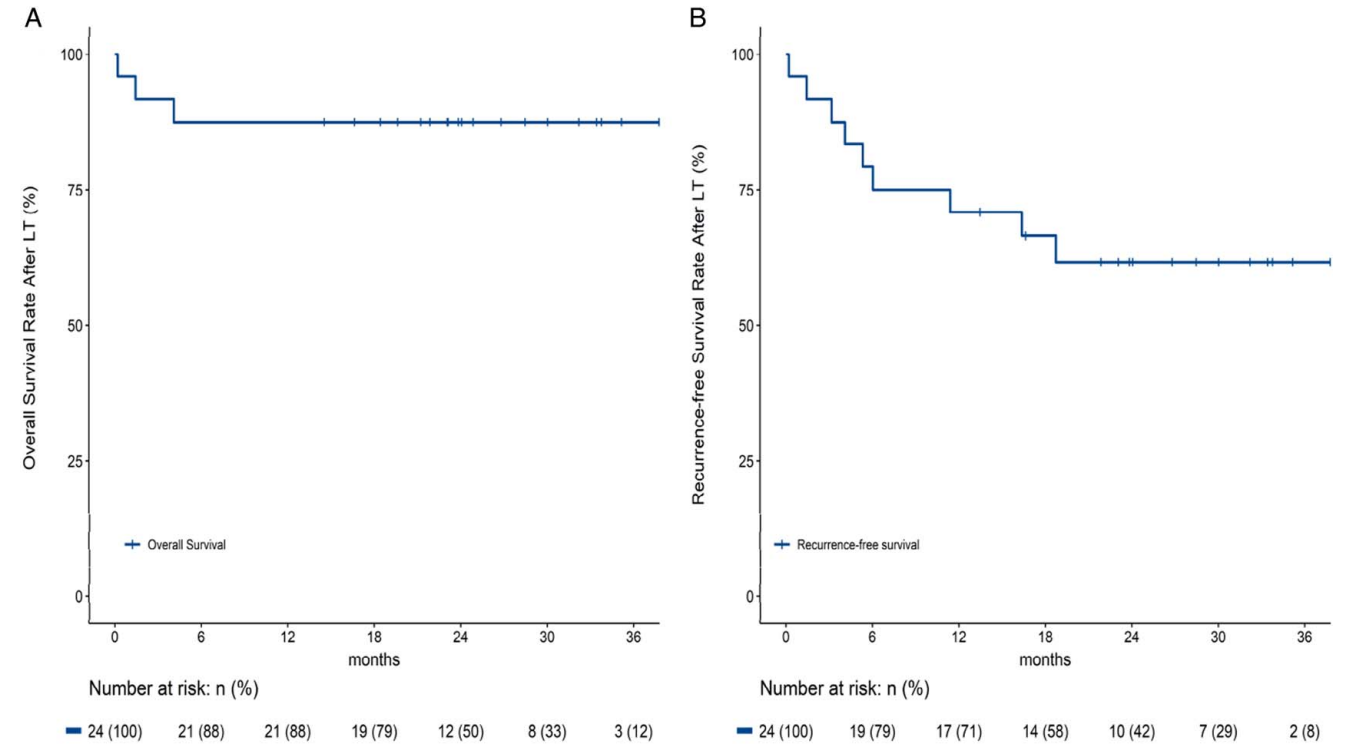

Figure 4. Overall survival and recurrence-free survival of 24 patients with hepatocellular carcinoma beyond Milan criteria who received liver transplantation after neoadjuvant immune checkpoint inhibitors in our center.

Table 3

All the patients in our center received LT after bridging/ downstaging with immunotherapy.

Characteristics	LT after immunotherapy ($n=24$)
ICI molecules	
Pembrolizumab	12 (50.0%)
Camrelizumab	7 (29.2%)
Tislelizumab	4 (16.7%)
Nivolumab	1 (4.1%)
Combination with lenvatinib	20 (83.3%)
Treatment cycles	3 (1–5)
Washout period (Day)	69.5 (18–206)
Rejection	3 (12.5%)
Recurrence	6 (25.0%)
Death	3 (12.5%)

Data are *n* (%) or median (Range).
ICI, immune checkpoint inhibitors; LRT, loco-regional therapy.

Instead of radiological responses, pathological responses have been adopted as surrogate endpoints for neoadjuvant immunotherapy trials in patients with resectable HCC[44,45]. Kaseb *et al*.[47] used an exploratory cutoff of 70% tumor necrosis as an endpoint significant tumor necrosis. Then, Marron *et al*.[48] agreed with this cutoff value and found that 20% of patients had significant tumor necrosis with two cycles of neoadjuvant cemiplimab. The results in our study suggest remarkable clinical activity of pembrolizumab plus lenvatinib for HCC with a significant pathological response rate at 60%, higher than previous studies of neoadjuvant ICIs therapy in HCC[47,48].

Limitations

Limitations of our study include the fact that it was a single-center study with a relatively small sample size. Initially, the study was designed with a sample size of 192, unluckily, the COVID-19 pandemic had hampered the enrollment of patients and the donor liver allocation. Particularly, due to the increasing time periods of neoadjuvant therapy and washout period, the follow-up time after LT was inevitably shorter in the PLENTY group than in the control group. Although a statistically significant difference was observed only in RFS but not in OS, which may be associated with the heterogeneity of participants caused by the small sample size and nonsystemic therapy-related death, the OS of the plenty group was still better than that of the control group. Additionally, the patients in this study had a larger tumor burden, and some patients had higher RETREAT scores or portal vein tumor thrombus, which may also lead to a certain bias. The extension cohort was also provided so that the transplant community could make a more comprehensive analysis of this issue. Despite the limitations, the results of this study will inform future research. On the one hand, the large-sample RCT is still recruiting and ongoing, and longer survival outcomes will be verified.

Conclusion

Our findings provide updated evidence that neoadjuvant ICIs combined with TKIs before LT is associated with better RFS for patients with HCC beyond MC, without increasing post-LT graft rejection. Systemic therapy in the preoperative setting for patients with HCC on the waiting list warrants further studies, which will promote the DS and bridging strategies into a new era.

Ethical approval

The study was reviewed and approved by the Ethical Committee of Renji Hospital(reference number KY2020-083). The trial was conducted in accordance with the principles of the Declaration of Helsinki and Good Clinical Practice. All procedures were in accordance with the STROBE guidelines.

Consent

Written informed consent on study aims, participation requirements, and the right to refuse was obtained from all participants.

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Author contribution

F.H., X.Q., and L.Z.C.: participated in the conception and design of this study; F.H.: was the project manager and coordinated patient recruitment; L.Z.C., Z.Z.J., Y.J., X.L., T.Y., T.H., L.L., Z.J., W.Y., Z.Z.G., L.Z.B., and W.T.: were involved in the acquisition, analysis, or interpretation of data; L.Z., Y.J., J.Y., and Z.Z.J.: drafted the manuscript. All the authors contributed to the critical review and final approval of the manuscript. F.H., X.Q., and L.Z.C.: accessed and verified the underlying study data. All authors were responsible for the decision to submit the manuscript.

Conflicts of interest disclosure

The authors declare no conflicts of interests.

Research registration unique identifying number (UIN)

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Guarantor

Hao Feng and Qiang Xia.

Data availability statement

The data of this study are available under a transfer agreement from the corresponding author based on a reasonable request. There will be limitations on how data can be used or how long data will be available for.

Provenance and peer review

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