Actual therapeutic efficacy of pre-transplant treatment on hepatocellular carcinoma and its impact on survival after salvage living donor liver transplantation.

A short running head: Pretreatment for HCC before LDLT

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Abstract

Background. The exact efficacy of pre liver transplant (LT) therapy for hepatocellular carcinoma (HCC) and the impact on survival after LT remain controversial in regard to salvage LT.

Materials and Methods. Of 79 patients transplanted in Nagasaki University Hospital between August 1997 and December 2007, 29 patients (36.7%) were indicated for HCC based on the Milan criteria using computed tomography and magnetic resonance imaging. Pre-LT therapy other than liver resection had been performed in 18 cases (62.1%) for 24 lesions. Treated lesions were analyzed histologically using thin slices of the whole explanted liver.

Results. Pre-LT therapy included transarterial chemoembolization (TACE) for 10 lesions, percutaneous ethanol injection (PEI)+TACE for 1 lesion, PEI in 6 lesions and ablation therapy in 7 lesions. Under preoperative imaging study, 19 lesions (79.1%) were "thought-to-be" necrotic by pre-LT therapy. However, histologically, viable HCCs were still observed in 9 lesions (9/19 47%). A median interval between the first pre-therapy and LT was 22 months, while last pre-LT therapy and LT was 11 months. No sarcomatous HCC or forced portal venous tumor thrombus was found in all cases with

residual lesions. One peritoneal recurrence has occurred after LT, in whom PEI and RFA had been performed before LDLT. The disease free survival after LDLT was comparable to that of cases without pre-LT therapy.

Conclusion. Half of the preoperatively "thought-to-be" necrotic lesions still contained viable HCC cells after the pre-LT treatment. Overall, the history of preLT therapy does not preclude or interfere following LT, although percutaneous treatment may spread disseminated tumor cell growth under immunosuppression.

Key Words: HCC, pre-LT, recurrence

Introduction

In Japan, where the availability of deceased liver donors is limited, hepatocellular carcinoma (HCC) is primarily either treated with hepatic resection, locoregional therapy and transarterial chemoembolization.[1-3] However, when HCC recurs and further treatment is no longer possible, liver transplantation (LT) may be considered as salvage LT. [4]

There are drawbacks of pre-LT treatment for HCC during the waiting period. Dissemination[5] and implantation[6] may occur after puncture of HCC and they may form tumors after the administration of immunosuppressive drugs. In addition, after incomplete locoregional therapy, sarcomatous changes have been reported.[7] With subsequent liver transplantation, damage to vital vascular structures can occur (hepatic artery, portal vein) which may affect the outcome of liver transplantation. Therefore, pre-LT therapy for HCC may increase the possibility of unfavorable changes in HCC and mask the possibility of occult HCC in a background liver, thereby compromising the outcome of LT. Therefore, the effect of pre-LT therapy on HCC and the outcome of LT for HCC, especially within the Milan criteria, have been reported with mixed results.[8-15]

A recent paper has reported the multiple occurrence and spread of HCC in a cirrhotic liver using whole liver histological examination (WLHE). However, the exact therapeutic effect of pre-LT treatment on primary HCC has not been analyzed. Explant analyses using WLHE were used, since it is the only opportunity to investigate the true viability of HCC in thought-to-be completely necrotic lesions following pre-LT therapy in imaging. This study, investigated the accuracy of pre-LT therapy for HCC and its impact on the outcome of LT.

Materials and methods

Patients

Of 79 cases transplanted in Nagasaki University Hospital before Dec. 2007, 29 case (36.7%) were indicated for HCC within the Milan criteria and for 18 cases (62.1%), pre-LT therapy other than liver resection was performed (Fig. 1). WLHE was performed with dedicated pathologists, with 5-7 mm slice for whole liver explants. Residual HCCs after pre-LT therapy were investigated histologically in combination by various factors.

This study was approved by the local Institutional Review Board and

written informed consent was obtained from all patients.

Patient characteristics

All patients were indicated for LDLT as “salvage LT”. The etiology in these cases was hepatitis C virus (HCV) infection in 12 patients and hepatitis B virus (HBV) in 6 patients. There were 7 females and 11 males, with a median age of 57 years (range, 48–61 years). The median values of alpha-fetoprotein (AFP) and protein-induced vitamin K antagonists II (PIVKA-II) were 30.25 ng/ml (range 0.8-806.1) and 23 g/ml (range, 6-247).

Liver transplantation and preoperative therapy for HCC

The median follow-up period was 24 months (range, 12-45 months). Pretreatment for HCC was performed prior to LT for 24 lesions in 18 patients, which included transarterial chemolipiodolization (TACE) in 10 lesions, radiofrequency ablation (RFA) in 7 lesions, percutaneous ethanol injection therapy (PEIT) in 6 lesions, TACE with PEIT in 1 lesion. Based on the imaging findings, all HCCs were considered to be within the Milan criteria. The clinical characteristics of the 18 patients are summarized in

Table 1.

All lesions were surveyed by based on multidetector computed tomography scanning (MDCT) and magnetic resonance imaging (MRI) done within 1 month before transplant. Preoperatively “thought-to-be” necrotic lesions were not counted as HCC lesions counted for Milan criteria. Only “thought-to-be” viable lesions evaluated by MD-CT with contrast media and MRI-SPIO were counted, which is also determined by Japanese national health insurance system. Preoperative imaging findings showed 5 patients with solitary viable HCC, 5 patients with double viable HCCs, 1 patient with triple viable HCCs and 7 patients with no viable HCCs. All patients met the Milan criteria with a solitary nodule 5 cm in size or 3 nodules 3 cm for multi-nodular HCC. [18]

Whole liver histological examination (WLHE) [16]

After explanation, the cirrhotic livers were fixed in formalin for 48 hr. The livers were then sectioned at 5-7 mm intervals and each section was carefully inspected and mapped. All sections were embedded in paraffin and all slides were made from the paraffin-embedded material and routinely stained with

hematoxylin and eosin. The median total number of slides for each patient was 116.5 (range, 64-185 slides). All slides were examined by an experienced pathologist (co-authors S.O. and H.M.). The pathological diagnoses and analyses were made according to the fourth edition of *The General Rules for the Clinical and Pathological Study of Primary Liver Cancer,* published by the Liver Cancer Study Group of Japan. [17]

Statistical analysis

A statistical comparison of survival after LT was performed using the Kaplan-Meier method and compared by the log-rank test. Results were considered statistically significant when the p values were less than 0.05.

Results

At the time of LDLT, there was no sign of extrahepatic cancer spread in any of the patients. Histologically, after pre-LT therapy, viable HCCs were still observed in 9 (47%) out of 19 “thought-to-be necrotic” lesions. A median period between the pre-therapy and LT was 22 months (range 3-58 months). In 3 lesions, residual HCCs were found in the area next to necrotic area (Fig.

3), while in 6 lesions, residual HCCs were found within the same nodule (Fig. 4). With regard to tumor differentiation, there was no sarcomatous HCC in the treated residual lesions. No “forced” portal venous tumor thrombus was found around the remaining viable HCCs.

After LDLT, no recurrence was found except for one peritoneal dissemination (1/18:5.5%) after OLT. Since the type of recurrence is unusual after OLT, this case report will be shown below.

In fact, there were 3 patients transplanted for HCC beyond Milan Criteria in which 2 patients had undergone pre-LT therapy (Fig. 1). In one patient, HCC recurred in the lung 12 months after LDLT, and he underwent partial lung resection three times. As pre-LT treatment, he had undergone liver resection and multiple TACE before LDLT. Another patient died at one month after LDLT due to hemophagocytic syndrome.

Survival after LDLT

After LT, regardless of pre-LT therapy, patient survival and recurrence free survival were comparable between the groups. (Fig. 5)

Case report

A patient underwent pre-LT treatments and subsequently developed peritoneal recurrence after LDLT. The patient was a 62-year-old male who had suffered from end-staged liver cirrhosis due to hepatitis C virus infection and was indicated for LDLT in May, 2006. Previously he underwent a caudate lobe resection of the liver for HCC in 2001 and TACE for HCC in segment 4 and 7 in 2003. Subsequently he was treated with TACE for HCC in segment 4 and 7 in 2004 and PEIT for HCC in segment 7 in 2004. RFA was procedure of a choice for an HCC in segment 2 in January, 2006. Finally, he developed end-staged liver failure and underwent a transplant in May, 2006. In the explanted liver, under WLHE, no viable HCCs were found.

However, following an increase in the AFP and PIVKA-II, two mass lesions were found in Douglas' pouch and the left lower abdomen in October, 2007. He had been on cyclosporine monotherapy as immunosuppression. Two lumpectomies were performed, which revealed moderately differentiated HCC under histological examination. It was presumed that the pre LT treatment had disseminated the HCC, which developed slowly after the LDLT (Fig. 6). In August 2008, the patient died due to the multiple

recurrence of HCC (local recurrence in the Douglas's pouch, bone metastasis, multiple liver metastases in the graft and multiple lung metastases).

Discussion

The present study demonstrated that after pre-LT therapy, 47% of the lesions still had viable HCC cells. Previously, Kim *et al.* [19] reported that a viable tumor volume ratio greater than 10% after preLT therapy was a significant prognostic factor. Pompili *et al.* [20] also reported that 58.7% of HCC had partial necrosis after percutaneous ablation procedures and the effect depended on the size of HCCs. Also, Wong *et al* reported that fifteen nodules in five patients had <75% necrosis and these were due to local/non-local recurrences or perhaps suboptimal treatment with RFA, TACE or cisplatin gel injection. [21] The mean waiting time for LT was 162.5 days. Nine of 13 patients had a different number of nodules than before pre-LT therapy, although stage changed in only three patients. The last pre-LT therapy and LDLT, which was median 11 months in our study, signified that salvage LDLT was considered and performed with 1 year for HCC bearing patients with viral hepatitis. Indeed, the outcome of LDLT even after at most pre-LT therapy could bear good disease-free-survival.

However, in the current study cohort, one case had peritoneal metastasis although PEI and RFA had been performed before LDLT. The

patient finally died 2 years after LDLT with systemic metastases following resection of 2 peritoneal metastases. In this case, there was no residual HCC in the explanted liver at the time of LDLT. This case illustrates a worst-case scenario with regard to pre-LT treatment for HCC. Therefore, physicians should keep in mind that percutaneous locoregional therapy for HCC might cause such micro-dissemination with subsequent growth under immunosuppressive therapy. The value of adjuvant chemotherapy and choice of immunosuppressive agent such as Rapamycin needs to be further determined [22]. Although previous reports on pre-LT therapy basically tend to favor the treatment, the case documented above suggested some detrimental effect of percutaneous therapy for HCC.

With regard to the recurrence-free survival after LT, there was no significant difference between the patients with or without pre-LT therapy. Recently, Yao *et al.* [23] reported that after downstaging with pre-LT therapy in 61 patients the 1- and 4-year survival after LT was 96.2 and 92.1% respectively. There was no recurrence after LT. Overall, the recurrence-free survival after LT in patients after preLT treatment was as good as in patients with T2 HCC without therapy for HCC. The study cohort in the

current review were patients who received previous treatment for HCC, namely salvage liver transplantation. In comparison to down-staged patients, salvage LT patients should have a better survival since those patients have never demonstrated a condition beyond the Milan-Criteria. Therefore, the current results showing a good disease-free survival after LDLT is warranted.

Since the purpose of this study is to investigate the pre-LT treated lesions, we did not describe too much information on untreated HCC. Investigation of untreated HCC and occult HCC were described by us recently. [16] The characteristics of the occult HCCs that were undetectable by imaging, included a minute (median size 6 mm), well-differentiated appearance (80%), with indistinct margins (85.3%) and without vascular invasion (94%). In the study, a multicentric occurrence of HCCs was demonstrated in cirrhotic livers with HCCs within the Milan criteria., although undetectable HCCs in cirrhotic livers may have no impact on recurrence after LT.

In conclusion, after pre-OLT therapy, 47% of the lesions still had viable HCC cells. However, pre-LT therapy for HCC in salvage LT had no effect on

the outcome of LT. However, one case had peritoneal recurrence probably due to percutaneous locoregional therapy under immunosuppression.

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Figure legends

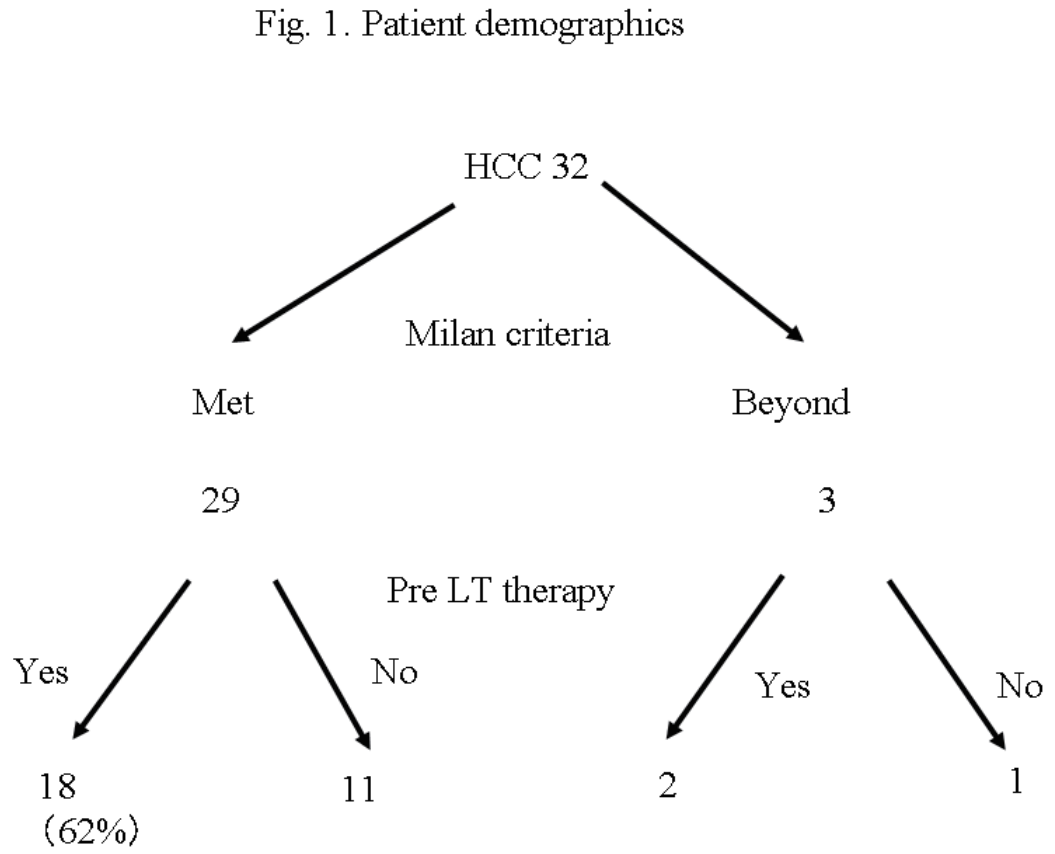

Fig. 1. Patient demographics. LT: liver transplantation, HCC: hepatocellular carcinoma.

Fig. 2. Actual effect of pre-LT therapy for HCC

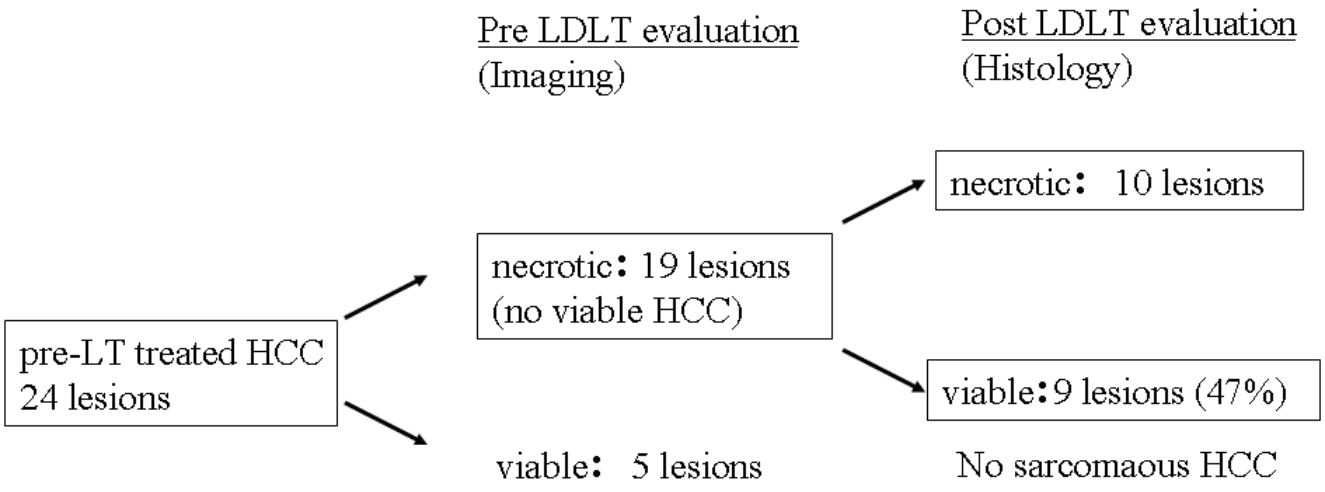

Fig. 2. Actual effect of pre-LT therapy on HCC

LDLT: living donor liver transplantation, HCC: hepatocellular carcinoma

Fig. 3. Case presentation

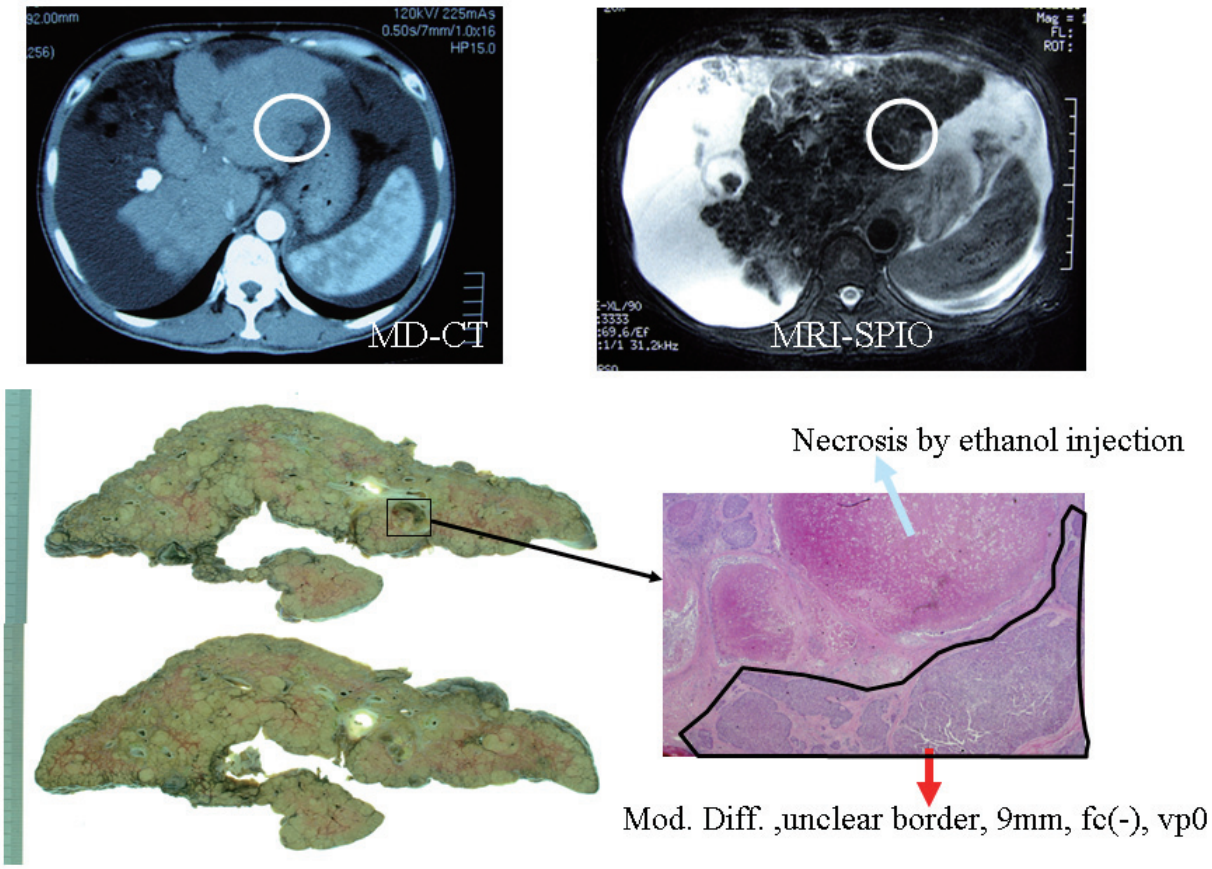

Fig. 3. A case presentation.

58 year-old male, Liver cirrhosis due to hepatitis B viral infection,

Pre-LT imaging diagnosis: HCC 0

Fig. 4. Case presentation

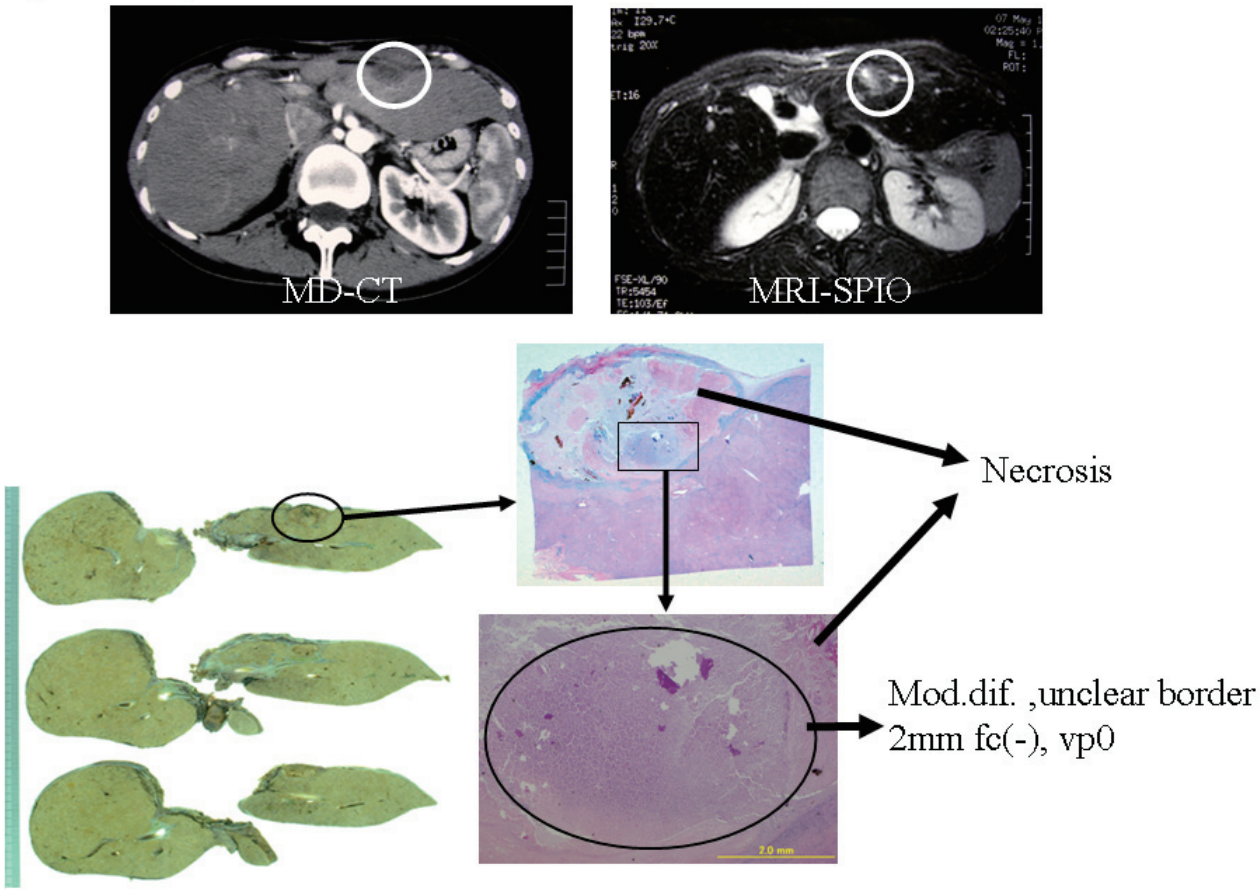

Fig. 4. A case presentation.

33 year-old Female, Liver cirrhosis due to hepatitis B viral infection,

pre-LT imaging diagnosis: HCC 0

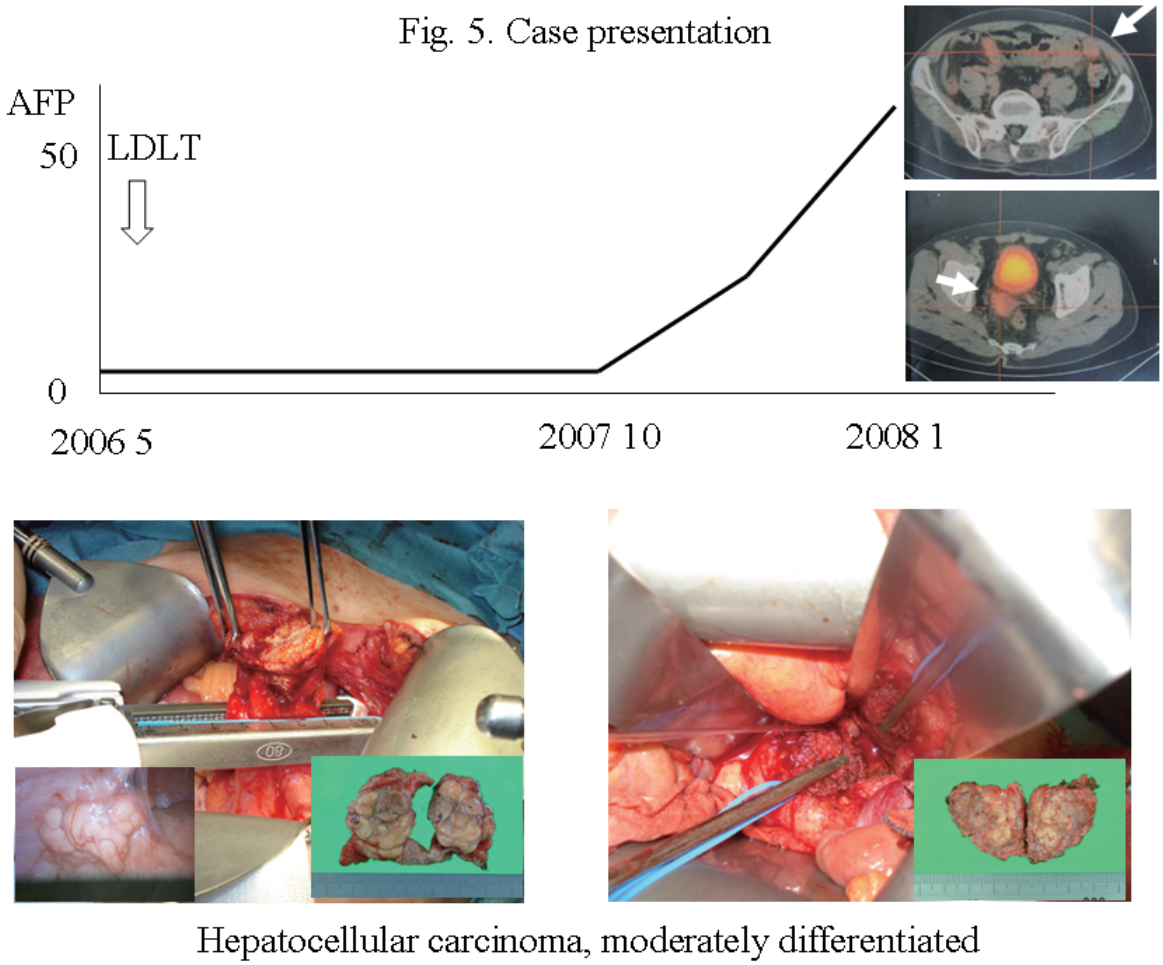

Fig. 5. A case presentation of peritoneal dissemination after LDLT.

LDLT: living donor liver transplantation.

Fig. 6. Survival after LDLT for patients with HCC within Milan criteria

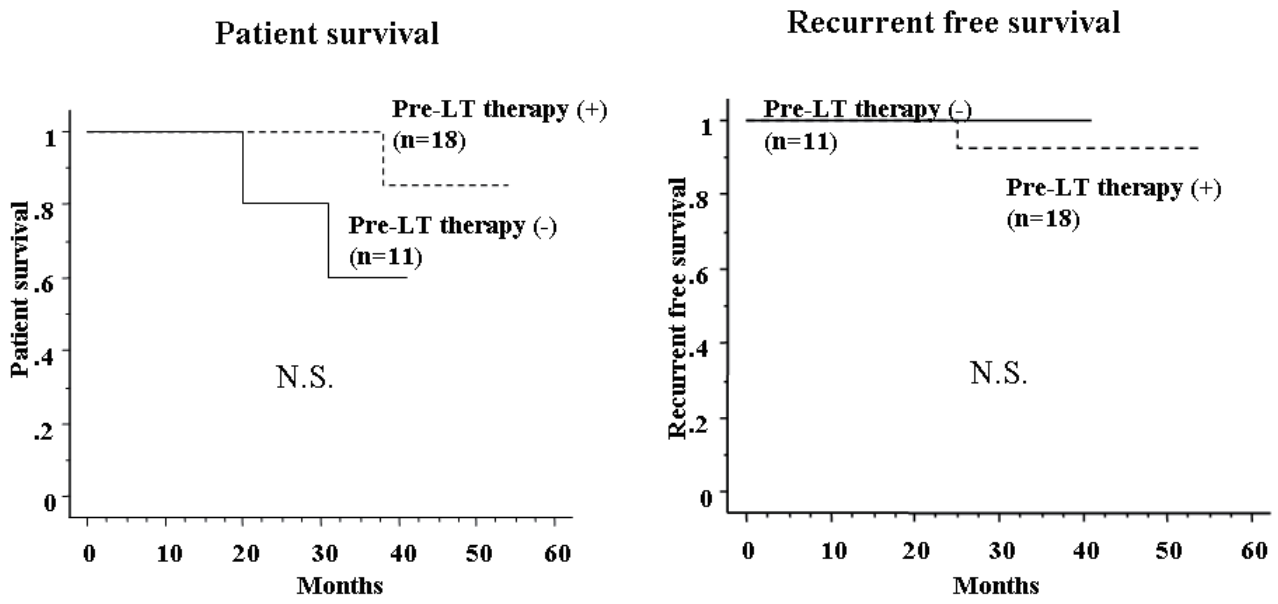

Fig. 6. Patient survival and recurrence-free survival after liver transplantation for patients with HCC within the Milan criteria.

TABLE I. Details in 18 patients with pre-LT therapy

TACE	10
Ethanol injection	6
Ablation	7
TACE + ethanol injection	1
Size of treated HCC	**18 mm (10-30)**
Number of therapy	**2 (1 - 4)**
Period between 1st therapy and LDLT	**22 months (3 - 58)**
Period between last therapy and LDLT	**11 months (3 - 58)**

TACE: transarterial chemoembolization, RFA: radiofrequency ablation, HCC: hepatocellular carcinoma, LDLT:living donor liver transplantation