Table 3. Patient characteristics

	mICC (n=14)	pHCC (n=22)	*P*
Size (cm), mean±SD (min–max)	5.0±2.4 (1.5–7.5)	7.5±4.7 (2.0–16.5)	0.066
Shape			
Lobulated	8 (57)	5 (23)	0.036
Round/oval	6 (43)	17 (77)	
Intratumoral hemorrhage	4 (29)	11(50)	0.204
Fat	0 (0)	11 (50)	0.002
Central hypointensity on T2WI	6 (43)	2 (9)	0.018
Fibrous capsule	0 (0)	13 (59)	<0.001
Arterial enhancement			
Ring-like	11 (79)	11 (50)	0.087
Other	3 (21)	11 (50)	
Late enhancement	10 (71)	3 (16)	<0.001
Uptake on hepatobiliary base	4 (19)	2 (9)	0.126
ADC value ($\times 10^{-3}$ mm²/s), mean±SD	0.85±0.18	0.87±0.20	0.722
Vascular invasion	2 (14)	4 (18)	0.760
Intrahepatic metastasis	3 (21)	8 (36)	0.343

Data are presented as n (%), unless otherwise noted.

mICC, mass-forming intrahepatic cholangiocarcinoma; pHCC, poorly differentiated hepatocellular carcinoma; SD, standard deviation; T2WI, T2-weighted imaging; ADC, apparent diffusion coefficient.

($P < 0.001$, Fig. 1); hypointense rim on precontrast T1-weighted image and T2-weighted image were detected in 10 and five cases, respectively. Rim enhancement on the dynamic late phase was seen in three cases. A hypointense area on T2-weighted image was seen at the center of the tumor in six of 14 mICC cases (43%) and two of 22 pHCC cases (9%), respectively ($P = 0.018$). Late enhancement was more commonly observed in mICC (n=10, 71%) than in pHCC (n=3, 14%) ($P < 0.001$) (Figs. 2, 3). No significant difference was observed in terms of intratumoral hemorrhage, arterial enhancement pattern (Figs. 2, 3), uptake of contrast on the hepatobiliary phase, ADC value, vascular invasion, or intrahepatic metastasis.

In pathological evaluation, central fibrous desmoplasia was observed to a greater or lesser degree in all mICC cases, while it was not observed in any pHCC cases. Pathologically, a fibrous capsule was detected in none of 14 mICC cases and 21 of 22 pHCC cases (95%). Of these 21 pHCC cases with pathologically identified fibrous capsule, MRI could identify the fibrous capsule during precontrast T1-weighted phase in 10 patients (47.6%), T2-weighted phase in five patients (23.8%), and dynamic late phase in three patients (14.3%).

Discussion

In this study MRI revealed interesting differences between mICC and pHCC, particularly relating to the presence or absence of intratumoral fat and fibrous capsule, and late enhancement at three minutes after hepatobiliary contrast agent injection.

It is well known that HCC has various degrees of fatty metamorphosis. However, researchers have focused on its relationship in the course of early stage of hepatocarcinogenesis. Fatty change is an important marker for the transformation of premalignant lesions to hepatocellular carcinoma (16). There are few reports regarding the fatty change of pHCC. Pathological evaluation by Kutami et al. (6) revealed frequencies of fatty change in well differentiated HCC, well-to-moderately differentiated HCC, moderately differentiated HCC, and moderately-to-poorly differentiated HCC as 42.0%, 37.5%, 6.0%, and 0%, respectively. Our study showed that there is a relatively high frequency of fatty change in pHCC (50%). The mechanism of fatty change in pHCC has not been reported previously. In case of small HCC (mainly well differentiated HCC), fatty change is closely related to an insufficient development of the arterial tumor vessels (6). Arterial blood supply significantly decreases as the histological grade increases in the late stage of HCC development (from moderately differentiated HCC to pHCC) (7), which may be related to the high frequency of fatty change in pHCC.

The imaging features of ICC have been reported by many researchers (3, 4). Most of these focused on imaging characteristics in relation to the internal desmoplastic change. Maetani et al. (4) observed central hypointensity on T2-weighted images in 27 of 50 cases (54%) of ICC and suggested that this finding, which reflects severe fibrosis, may be a characteristic marker of ICC. In our study, a central hypointense area was seen on T2-weighted images in six of 14 cases (42.9 %), which is concordant with their results. The number of cases exhibiting hypointensity on T2-weighted images (n=6) was lower than that of late enhancement (n=10) in the present study. Coagulation necrosis shows both high and low signal intensity on T2-weighted images and can intermingle with the fibrous stroma, which can affect the internal signal intensity. Another potential cause of this discrepancy may have been the scanning slice thickness (7 mm vs. 3 mm) and contrast resolution.

The standard gadolinium dose for gadoxetic acid (0.025 mmol/kg) is one-fourth that of gadopentetate dimeglumine (0.1 mmol/kg). As the T1 relaxivity of gadoxetic acid is 1.8 times that of gadopentetate dimeglumine (17), the expected T1 relaxation effect would be expected to be one-half that of gadopentetate dimeglumine (18). Biodistribution studies of gadoxetic acid have shown dose-independent renal (41.6%–51.2%) and biliary (43.1%–53.2%) elimination and an enterohepatic recirculation rate of approximately 4% (19). Even though

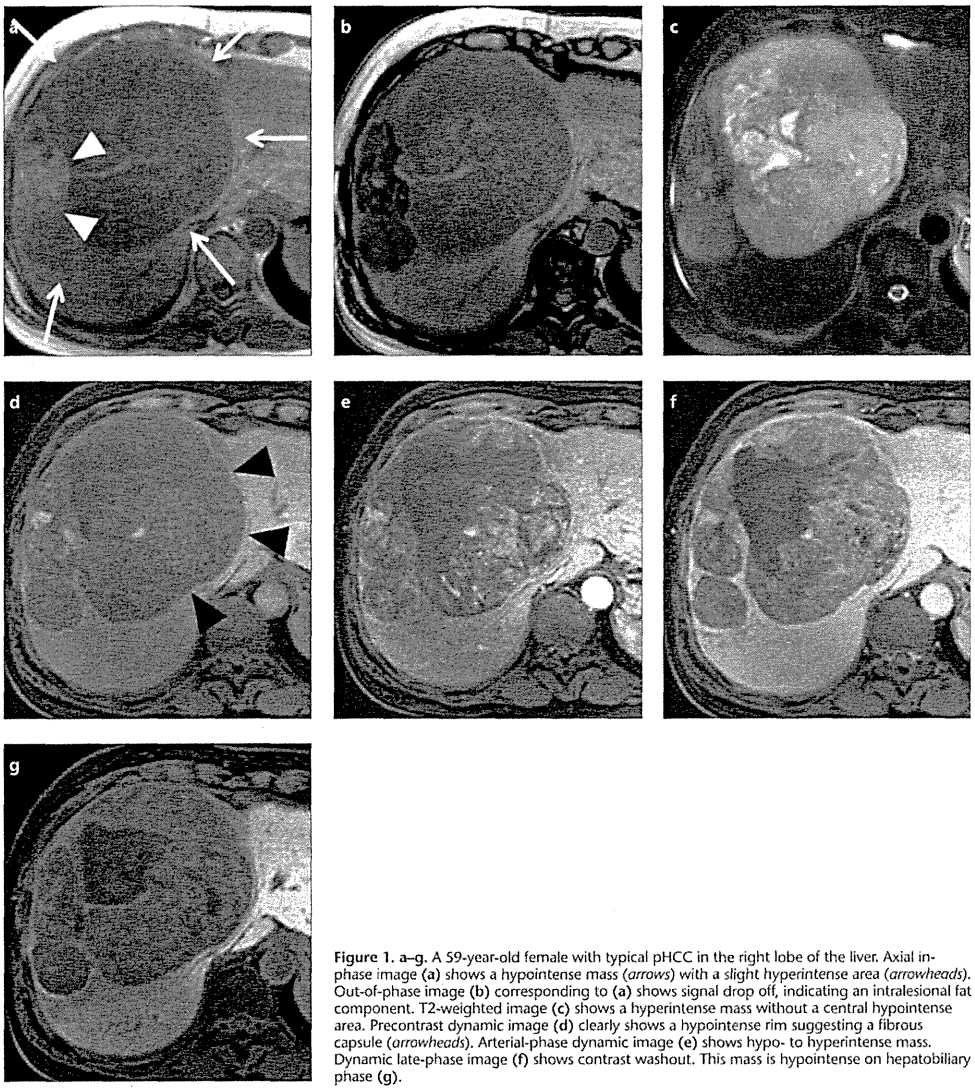

Figure 1. a–g. A 59-year-old female with typical pHCC in the right lobe of the liver. Axial in-phase image **(a)** shows a hypointense mass (*arrows*) with a slight hyperintense area (*arrowheads*). Out-of-phase image **(b)** corresponding to **(a)** shows signal drop off, indicating an intralesional fat component. T2-weighted image **(c)** shows a hyperintense mass without a central hypointense area. Precontrast dynamic image **(d)** clearly shows a hypointense rim suggesting a fibrous capsule (*arrowheads*). Arterial-phase dynamic image **(e)** shows hypo- to hyperintense mass. Dynamic late-phase image **(f)** shows contrast washout. This mass is hypointense on hepatobiliary phase **(g)**.

the effect of recirculated or extracellular distribution of gadolinium might be less in gadoxetic acid-enhanced MRI than in gadopentetate dimeglumine-enhanced MRI, our result suggests that the dynamic late phase can give us useful information, similar to delayed enhancement, using an extracellular contrast agent. In addition, despite the absence of functional hepatocytes in mICC, uptake of contrast was seen in four mICC cases on hepatobiliary phase. This is probably due to the remaining contrast in the extracellular space of the tumor. After 2–5 minutes of contrast injection, extracellular contrast agent returns from the interstitial space to the vascular space due to decreased concentration in the vascular space caused by excretion into urine (20). Furthermore, because of the

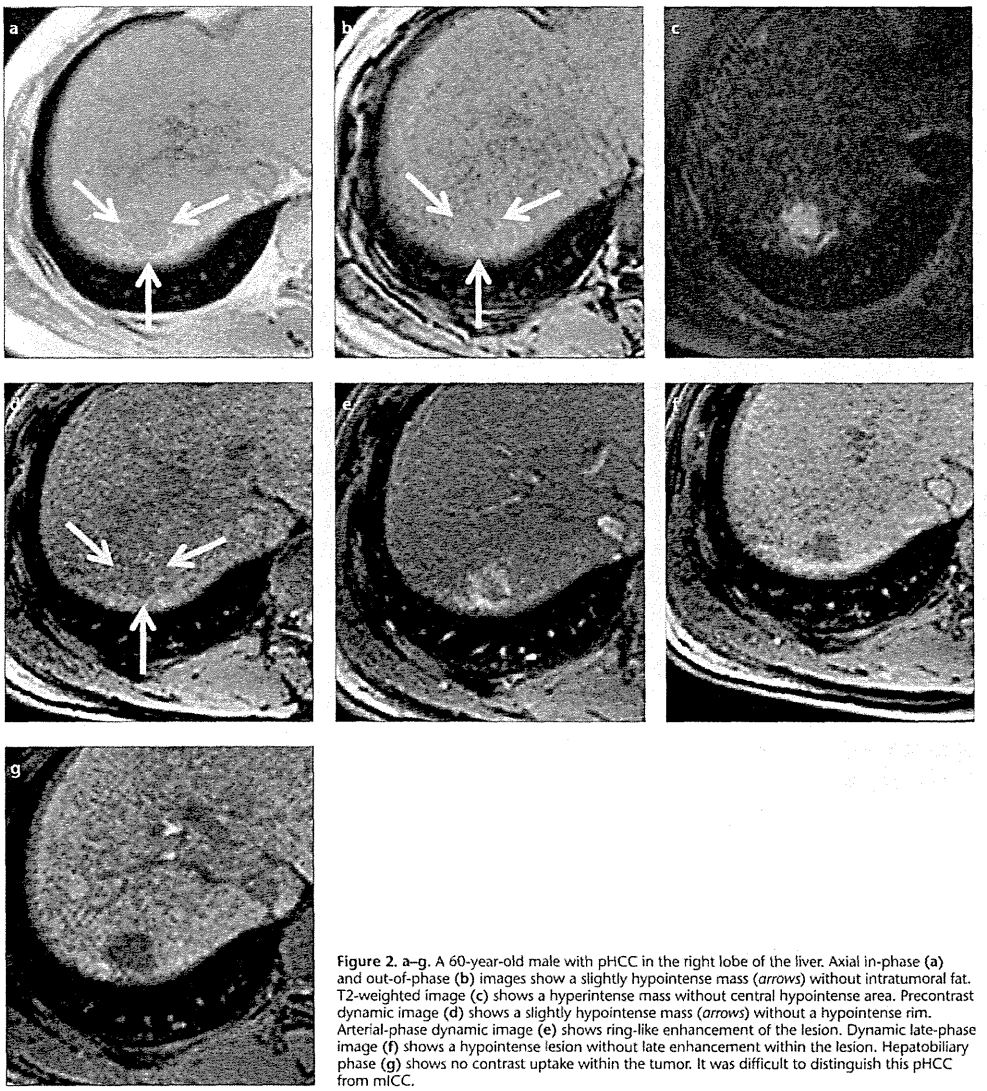

Figure 2. a–g. A 60-year-old male with pHCC in the right lobe of the liver. Axial in-phase **(a)** and out-of-phase **(b)** images show a slightly hypointense mass (*arrows*) without intratumoral fat. T2-weighted image **(c)** shows a hyperintense mass without central hypointense area. Precontrast dynamic image **(d)** shows a slightly hypointense mass (*arrows*) without a hypointense rim. Arterial-phase dynamic image **(e)** shows ring-like enhancement of the lesion. Dynamic late-phase image **(f)** shows a hypointense lesion without late enhancement within the lesion. Hepatobiliary phase **(g)** shows no contrast uptake within the tumor. It was difficult to distinguish this pHCC from mICC.

stronger enhancement of the liver parenchyma on hepatobiliary phase of gadoxetic acid-enhanced image (21), most mICC did not show contrast distribution, in contrast to dynamic late phase. In case of pHCC, the uptake of contrast was observed only in 9% of cases (2 of 22) on hepatobiliary phase, because the expression of uptake transporter (organic anion-transporting polypeptide 8 (OATP8) may decrease as the tumor grade advances (22). Hepatobiliary phase images were not helpful in differentiating mICC from pHCC in our study, however, hepatobiliary phase is very useful in detection of satellite nodules or intrahepatic metastases (21). In addition, radiologists should pay attention to the pseudo-washout sign (23), which shows relatively low signal intensi-

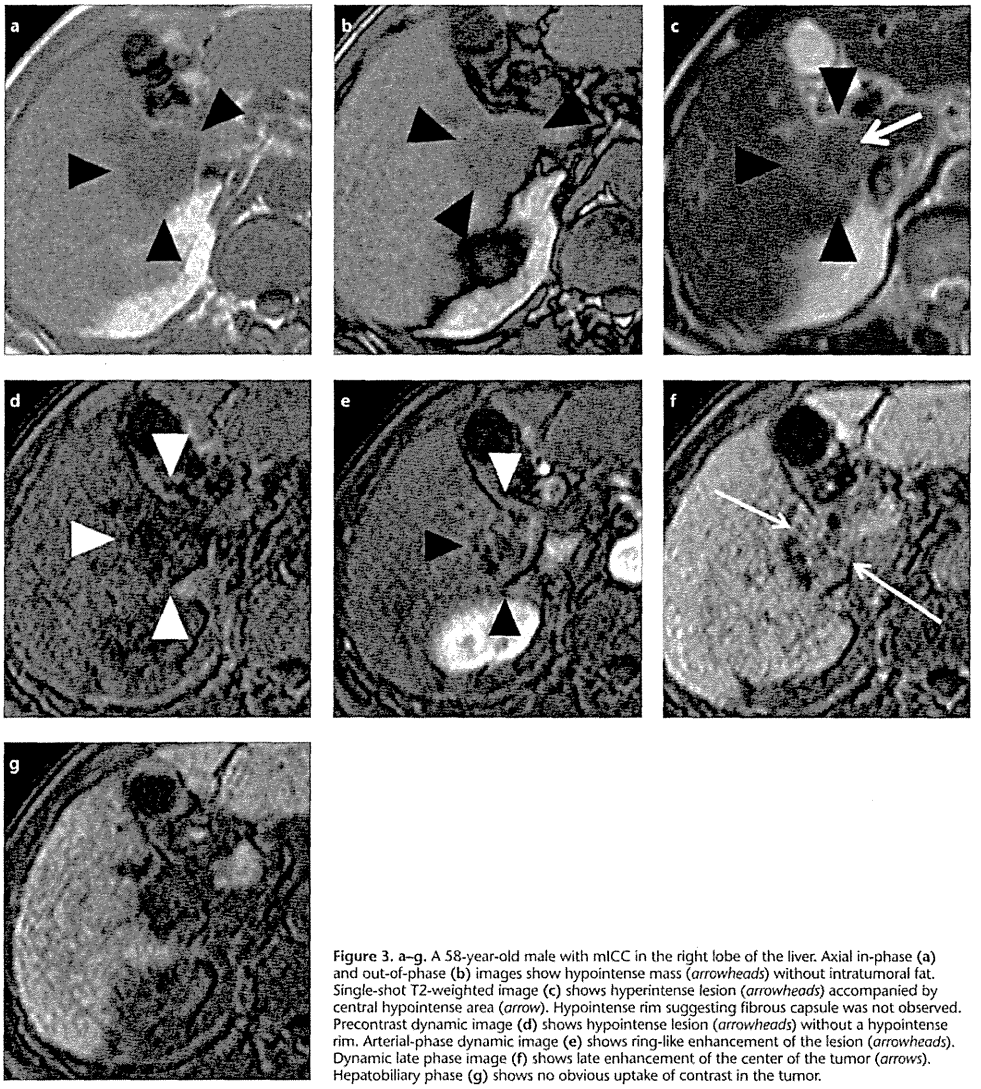

Figure 3. a–g. A 58-year-old male with mICC in the right lobe of the liver. Axial in-phase (**a**) and out-of-phase (**b**) images show hypointense mass (*arrowheads*) without intratumoral fat. Single-shot T2-weighted image (**c**) shows hyperintense lesion (*arrowheads*) accompanied by central hypointense area (*arrow*). Hypointense rim suggesting fibrous capsule was not observed. Precontrast dynamic image (**d**) shows hypointense lesion (*arrowheads*) without a hypointense rim. Arterial-phase dynamic image (**e**) shows ring-like enhancement of the lesion (*arrowheads*). Dynamic late phase image (**f**) shows late enhancement of the center of the tumor (*arrows*). Hepatobiliary phase (**g**) shows no obvious uptake of contrast in the tumor.

ty of hypervascular tumor because of continuous contrast uptake in the surrounding normal hepatic parenchyma during the equilibrium phase.

MRI detection of fibrous capsule using an extracellular contrast agent is reported to be most sensitive on the delayed-phase (24). Our results showed that precontrast T1-weighted image is the most sensitive at detecting fibrous capsule, while dynamic late phase had the lowest detection rate. Even though there is a fair amount of contrast distribution into the fibrous capsule, increasing signal intensity due to uptake of contrast by the hepatocytes and excretion into bile ducts in the surrounding noncancerous parenchyma may obscure the pseudocapsule (Fig. 4).

Nishie et al. (25) reported that the mean ADC of pHCC is significantly low-

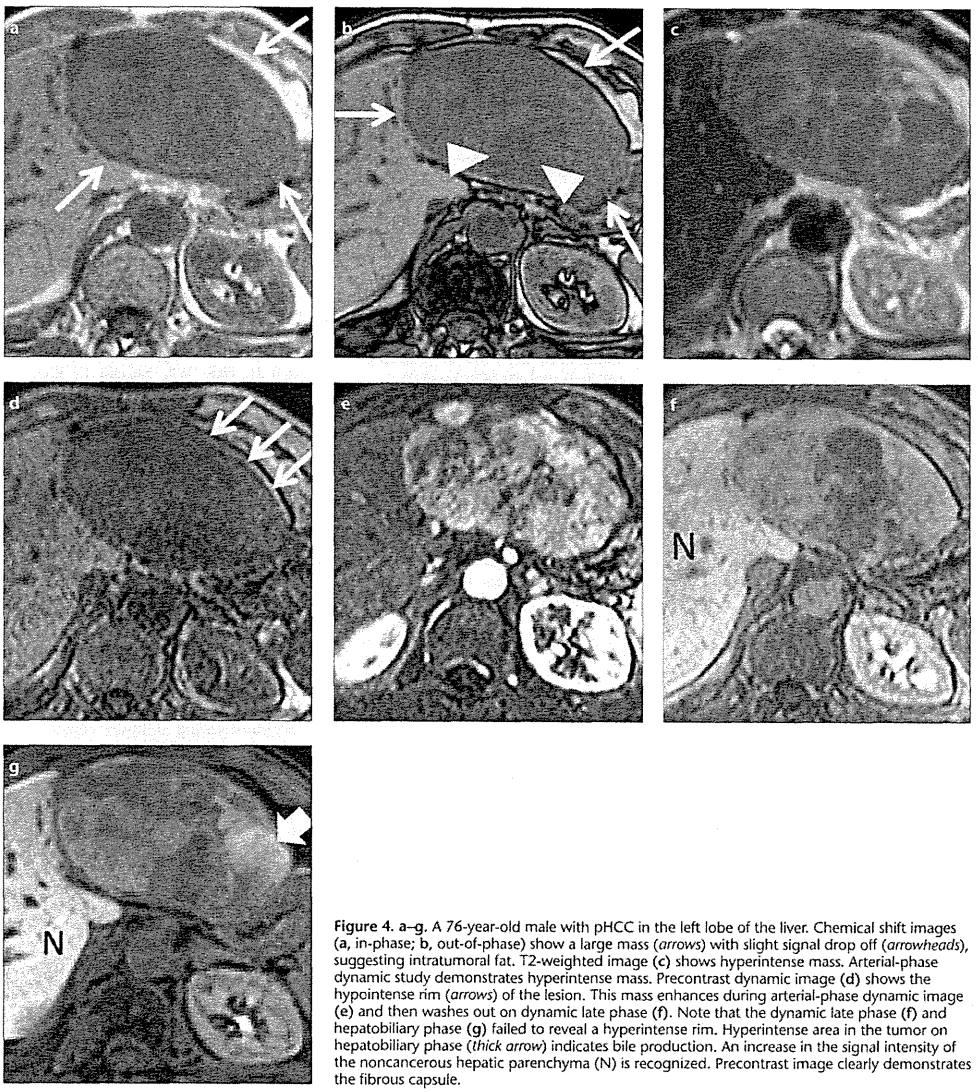

Figure 4. a–g. A 76-year-old male with pHCC in the left lobe of the liver. Chemical shift images (**a**, in-phase; **b**, out-of-phase) show a large mass (*arrows*) with slight signal drop off (*arrowheads*), suggesting intratumoral fat. T2-weighted image (**c**) shows hyperintense mass. Arterial-phase dynamic study demonstrates hyperintense mass. Precontrast dynamic image (**d**) shows the hypointense rim (*arrows*) of the lesion. This mass enhances during arterial-phase dynamic image (**e**) and then washes out on dynamic late phase (**f**). Note that the dynamic late phase (**f**) and hepatobiliary phase (**g**) failed to reveal a hyperintense rim. Hyperintense area in the tumor on hepatobiliary phase (*thick arrow*) indicates bile production. An increase in the signal intensity of the noncancerous hepatic parenchyma (N) is recognized. Precontrast image clearly demonstrates the fibrous capsule.

er than those of well and moderately differentiated HCC. To our knowledge, no previous reports focusing on the ADC of mICC have been reported. In our study, the mean ADC of mICC (0.85±0.18 $\times 10^{-3}$ mm^2/s) was almost the same as that of pHCC (0.87±0.20 $\times 10^{-3}$ mm^2/s). We can deduce from these data that it is difficult to differentiate mICC from pHCC by means of ADC.

There were several limitations to this study. First, our study population was small because mICC and pHCC are not common diseases. Second, we did not compare the area of late enhancement to that of pathological fibrosis directly. Third, in daily practice, dynamic CT is routinely performed for evaluating liver tumor; thus, delayed enhancement can be easily evaluated. We did not directly compare the

diagnostic performance of gadoxetic acid-enhanced MRI and dynamic CT or extracellular gadolinium contrast agent. Fourth, we did not compare with other tumors such as liver metastasis or inflammatory pseudotumor in noncirrhotic liver. In particular, liver metastasis in noncirrhotic liver will be encountered in the daily practice. It goes without saying that patients' medical history is important in making the differential diagnosis.

In conclusion, the absence of fat and fibrous capsule, and presence of enhancement at 3 min are more indicative for mICC than for pHCC.

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Conflict of interest disclosure

The authors declared no conflicts of interest.

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Comparative Study of Living and Deceased Donor Liver Transplantation as a Treatment for Hepatocellular Carcinoma

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BACKGROUND: Living donor liver transplantation (LDLT) is an important treatment option for unresectable hepatocellular carcinoma (HCC), but whether recurrence and survival in LDLT differ from those in deceased donor liver transplantation (DDLT) remains controversial.

STUDY DESIGN: A retrospective analysis was performed between patients with HCC who underwent LDLT in a Japanese institute (n = 133) and those who underwent DDLT in a United States institute (n = 362).

RESULTS: Although there was a difference in patient background characteristics (eg, body mass index, donor age, Model for End-Stage Liver Disease [MELD] score), tumor aggressiveness represented by Milan criteria and microscopic vascular invasion were comparable between the 2 groups. The cumulative 5-year recurrence rates of the LDLT group and the DDLT group were similar (14.8% vs 19.0%, $p = 0.638$), but overall survival in the LDLT group was significantly better than that in the DDLT group (84.2% vs 63.5%, $p < 0.0001$). Separate multivariate analysis identified different preoperative predictive factors for HCC recurrence (salvage transplantation and Des-gamma-carboxy prothrombin >300 in the LDLT group, beyond Milan criteria in the DDLT group). Combined multivariate analysis of the 2 groups identified recipient's body mass image >30 kg/m^2 as an independent risk factor for overall survival; the technique of transplantation (LDLT or DDLT) was not found to be a risk factor.

CONCLUSIONS: When compared between the institutes where LDLT or DDLT were the first treatment choices for unresectable HCC, recurrence rates were comparable. Living donor liver trasplantation is a viable treatment option for unresectable HCC, providing recurrence rates similar to those achieved with DDLT. (J Am Coll Surg 2015;220:297–304. © 2015 by the American College of Surgeons)

The efficacy of liver transplantation (LT) as a treatment option for unresectable hepatocellular caricinoma (HCC) is well established because it removes both the tumor and the cirrhotic liver that is at risk of developing future malignancy.[1] The Milan criteria (1 nodule with a maximal diameter of 5 cm or up to 3 nodules with a maximal diameter of 3 cm) are widely accepted for selection of patients with HCC for LT, and using them helps achieve post-transplant long-term survival comparable to that in patients without HCC.[1]

In the United States, approximately 7,000 new patients with HCC are put on the waiting list for deceased donor liver transplantation (DDLT) each year, and 15% die during the waiting period without receiving an LT due to the relative shortage of deceased donors.[2] Because long waiting time for DDLT increases the risk of tumor progression and drop out from the waiting list, living donor liver transplantation (LDLT) has been proposed as an alternative.[3,4] However, the impact of the source

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Abbreviations and Acronyms

BMI = body mass index
DCP = des-gamma-carboxy prothrombin
DDLT = deceased donor liver transplantation
HCC = hepatocellular carcinoma
LDLT = living donor liver transplantation
LT = liver transplantation
MELD = model for end-stage liver disease
MSMC = Mount Sinai Medical Center

of the graft (ie, DDLT or LDLT) on the treatment outcome remains controversial. Some previous reports have demonstrated worse overall and disease-free survivals for patients treated by LDLT for HCC[5-7]; others have shown similar outcomes between LDLT and DDLT.[8,9] Because most published studies were conducted in Western countries, where the large majority of LT are DDLT, a possible bias for selection of treatment may exist such that patients within Milan criteria are preferentially treated with DDLT and those beyond Milan criteria are relegated to LDLT; this is clearly the case in the United States, where the organ allocation system assigns higher priority to patients with HCC within Milan criteria.[10]

In many Asian countries, especially Japan, the availability of organs from deceased donors is quite limited, so the first treatment option for patients with unresectable HCC is LDLT.[4,11] We aimed to compare the treatment outcomes between LDLT and DDLT in settings when they were the first treatment choice for unresectable HCC. Therefore, we compared LDLT in 133 patients from Kyushu University (Japan) and DDLT in 362 patients from Mount Sinai Medical Center (NY), both major transplant centers in their respective countries. The primary endpoint was recurrence rate after LT, because it is generally the most important factor that determines long-term outcomes after LT.

METHODS

Patients

Between January 2002 and December 2010, 386 patients with a diagnosis of HCC underwent primary DDLT at Mount Sinai Medical Center (New York, NY), and 133 patients underwent primary LDLT at Kyushu University Hospital (Fukuoka, Japan). After approval by the Mount Sinai Medical Center (MSMC) Institutional Review Board and Kyushu University Ethical Committee, data were extracted from database records and from hospital and office charts. Only patients with histologically proven HCC in their explants were included in this study.

At Kyushu University Hospital, the eligibility criteria for LDLT at the beginning of the study period were: no modality except LT available to cure HCC, end-stage liver disease; no extrahepatic metastasis; and no major vascular invasion such as portal vein or hepatic vein. There was no restriction on the tumor size or tumor number. Because initial data demonstrated that patients with both HCC > 5 cm and serum des-gamma-carboxy prothrombin (DCP) levels >300 mAU/mL had poor prognosis,[1] the policy changed in 2007 to exclude such patients from transplant candidacy.

At MSMC, the eligibility criteria for DDLT were: unresectable HCC within Milan criteria and tumor beyond these limits that was down-staged by nonsurgical treatment and maintained within Milan criteria for 6 months.

Because the series performed in MSMC included 24 patients (6.3%) with pathologic T4b tumors, but none were included in the Kyushu University Hospital cohort, these 24 patients were excluded from the comparative analysis. Therefore, 362 DDLT patients from MSMC (DDLT group) and 133 LDLT patients from Kyushu University Hospital (LDLT group) were enrolled in this study.

Preoperative assessment for hepatocellular carcinoma

Preoperative diagnosis and staging of HCC was with thoracic and abdominal CT and/or MRI. Routine biopsies were not performed. Tumors were staged according to the American Liver Tumor Study Group modified Tumor-Node-Metastasis (ALTSG-TNM) classification.[12]

Donor evaluation and selection

The selection criteria for partial liver graft from living donor in Kyushu University was based on volumetric analysis, and details are described elsewhere.[13,14] Briefly, the left lobe was initially considered for the graft. The right lobe was chosen if the estimated left lobe with the caudate lobe volume of the donor was less than 35% of the standard liver volume of the recipient. The person was excluded as a donor candidate if the remnant liver volume was less than 35% of the total liver volume. If the CT or ultrasound study showed the possibility of steatosis in the donor liver, short-term intensive treatment for hepatic steatosis was prescribed before surgery.[15]

Postoperative management and follow-up

The transplantation procedures of both institutes have been described previously.[13,16] In both institutes, postoperative immunosuppressive therapy consisted of a triple-drug regimen of cyclosporine or tacrolimus in

Table 1. Clinical Characteristics of Patients with Hepatocellular Carcinoma Receiving Living vs Deceased Donor Liver Transplantation

Variables	LDLT group (n = 133)	DDLT group (n = 362)	p Value
Recipient male sex,%	58.2	78.7	<0.0001
Recipient age, y, mean ± SD	57.6 ± 7.1	58.3 ± 7.4	0.330
Recipient BMI, kg/m^2, mean ± SD	23.9 ± 3.1	27.6 ± 5.4	<0.0001
Cause,%	–	–	0.001
HBV	15.6	14.1	–
HCV	75.4	58.6	–
Others	9.0	27.3	–
MELD score, mean ± SD	11.9 ± 4.9	15.9 ± 8.3	<0.0001
Waiting time, days, median (range)	44 (4–236)	196 (0–3996)	<0.0001
Salvage transplantation,%	67.2	62.5	0.379
Log AFP, mean ± SD	1.60 ± 0.97	1.20 ± 0.70	<0.0001
DCP, mAU/mL, mean ± SD	394 ± 1404	–	–
Beyond Milan criteria,%	41.4	34.3	0.162
Largest tumor diameter, cm, mean ± SD	2.4 ± 1.1	2.8 ± 1.8	0.013
No. of tumor nodules	4.8 ± 7.9	2.6 ± 2.2	<0.0001
Bilobar HCC,%	49.3	36.9	0.017
Pathologic ALTSG–TNM,%	–	–	0.005
T1	14.2	12.1	–
T2	40.3	52.5	–
T3	10.5	14.8	–
T4a	35.1	20.7	–
≥T3	45.5	35.5	0.041

AFP, alpha fetoprotein; ALTSG-TNM, American Liver Tumor Study Group modified Tumor-Node-Metastasis classification; BMI, body mass index; DCP, des-gamma-carboxy prothrombin; DDLT, deceased donor liver transplantation; LDLT, living donor liver transplantation; MELD, model for end-stage liver disease.

combination with corticosteroids and mycophenolate mofetil. The dose of corticosteroids was gradually tapered to discontinuation at 3 to 6 months after LT. The detailed postoperative follow-up policy in each institute has been described elsewhere.[17,18] Briefly, abdominal and chest CT scans were performed at 3- to 6-month intervals. Additionally, hepatic angiography, MRI, or bone scintigraphy examination was performed if there was deterioration in the graft function, or an increase in the alpha fetoprotein (AFP) or DCP levels was noted, although DCP was not measured in the MSMC cohort.

Statistical analysis

Descriptive statistics are expressed as mean ± standard deviation (SD) or percentages, and compared using a 2-tailed, unpaired Student's *t*-test. Categorical data were compared using the chi-square test. Nonparametric variables were expressed by median (range), and compared using the Mann-Whitney *U* tests. Overall survival and recurrence rates were estimated using Kaplan-Meier method and compared using the log-rank test. Deaths from all causes were included in the calculation of survival. Cox proportional hazard models were created to evaluate the risk associated with prognostic variables. Statistical analyses were performed using JMP 9 software (SAS Institute). A value of $p < 0.05$ was considered significant.

RESULTS

Characteristics of LDLT and DDLT recipients

Table 1 shows the baseline characteristics of the 133 LDLT and 362 DDLT recipients at the time of LT. No significant differences existed between the groups in terms of recipient age and the proportion who underwent salvage transplantation. The percentage of male sex, body mass index (BMI), and Model for End-Stage Liver Disease (MELD) score were significantly higher in the DDLT group than in the LDLT group. The percentage of recipients with hepatitis C virus as the underlying cause of cirrhosis was significantly higher in the LDLT group. Regarding preoperative tumor characteristics, no significant difference was found in the proportion of patients outside the Milan criteria. Log alpha fetoprotein levels, number of tumor nodules, and the proportion of patients

with bilobar HCC or pathologic American Liver Tumor Study Group modified Tumor-Node-Metastasis classification stage ≥T3 were significantly greater in the LDLT group than in the DDLT group; the largest tumor diameter was greater in the DDLT group than in the LDLT group.

Characteristics of LDLT and DDLT donors and operative variables

Table 2 shows that age and BMI of the donor and cold ischemic time were significantly higher in the DDLT group than in the LDLT group. The maximum age of the donor in the LDLT group was 58 years; that in the DDLT group was 86 years. In the DDLT group, 34.8% of donors were older than 60, but none of the donors in the LDLT group was older than 60. The DDLT group included 23 right lobe grafts (6.4%) as a result of split liver transplantation. The graft weight and graft-to-recipient weight ratio (GRWR) were significantly higher in the DDLT group than in the LDLT group. Operation time, amount of blood loss, and requirement for any type of blood transfusion were significantly greater in the LDLT group than in the DDLT group. At liver explant pathology, no significant difference was found between the groups for microscopic vascular invasion. The LDLT group had a higher incidence of poor histologic grade compared with that in the DDLT group.

Post-transplant survival and hepatocellular carcinoma recurrence

Figure 1A shows the overall survival after LT in all patients. The survival rate for LDLT recipients was significantly better than that for DDLT recipients. After a median post-transplant follow-up of 6.3 years in the LDLT group and 5.6 years in the DDLT group, the 1-, 3-, and 5-year overall survivals were 93.2%, 87.0%, and 84.2% in the LDLT group, and 83.1%, 72.5%, 63.5% in the DDLT group, respectively ($p < 0.0001$). Cumulative recurrence rates (Fig. 1B) after LT in all patients were not different between the groups (1-, 3-, and 5-year recurrence: 7.7%, 13.5%, and 14.8% in the LDLT group, and 6.0%, 13.8%, 19.0% in the DDLT group, respectively) ($p = 0.638$).

Table 2. Characteristics of Living vs Deceased Donors and Operative Variables

Variables	LDLT group (n = 133)	DDLT group (n = 362)	p Value
Donor			
Sex, male,%	68.9	54.4	0.004
Age, y, mean ± SD	33.7 ± 9.6	51.7 ± 18.3	<0.0001
BMI, kg/m^2, mean ± SD	23.9 ± 3.1	27.5 ± 5.5	<0.0001
Graft type, %	LL, 63.9; RL, 35.3; RPL, 0.8	Whole liver, 93.6; split liver, 6.4	
Graft weight, g, mean ± SD	475 ± 99	1394 ± 394	<0.0001
GRWR, %, mean ± SD	0.77 ± 0.16	1.82 ± 0.71	<0.0001
Blood type compatibility, %			<0.0001
Identical	77.3	93.9	
Compatible	15.9	5.5	
Incompatible	6.8	0.6	
Cold ischemic time, min, mean ± SD	80.1 ± 49.5	466.1 ± 186.5	<0.0001
Warm ischemic time, min, mean ± SD	39.6 ± 9.0	37.9 ± 9.4	0.125
Operative time, min, median (range)	792 (545–1,489)	425 (211–894)	<0.0001
Blood loss, mL, median (range)	5188 (150–35,000)	2000 (250–43,200)	0.009
Blood transfusion, U, median (range)			
PRBC	10 (0–100)	7 (0–139)	0.001
FFP	15 (0–150)	7 (0–75)	<0.0001
PC	20 (0–150)	0 (0–44)	<0.0001
Microscopic vascular invasion, %	39.9	40.3	0.922
Tumor differentiation, %			<0.0001
Well	7.5	40.8	
Moderate	63.9	49.7	
Poor	28.6	9.7	

BMI, body mass index; DDLT, deceased donor liver transplantation; FFP, fresh frozen plasma; GRWR, graft-to-recipient weight ratio; LDLT, living donor liver transplantation; LL, left lobe; PC, platelet concentrate; PRBC, packed red blood cells; RL, right lobe; RPL, right posterior lobe.

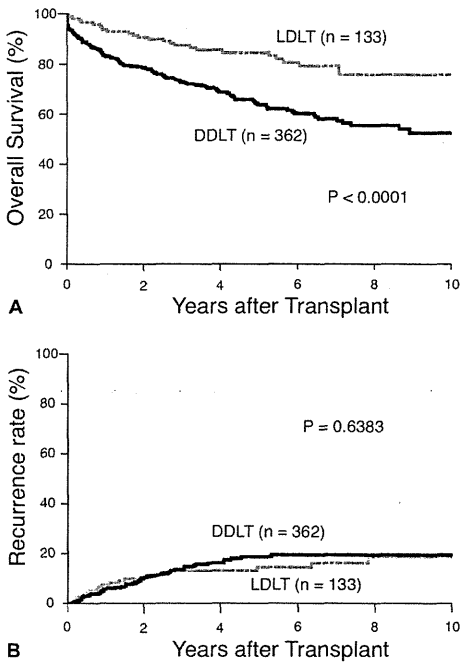

Figure 1. Kaplan-Meier curves of (A) patient overall survival and (B) HCC recurrence rate after liver transplantation for HCC patients. Comparison between LDLT performed in Kyushu University Hospital in Japan (n = 133) and DDLT performed in the Mount Sinai Medical Center in New York (n = 362). DDLT, deceased donor liver transplantation; HCC, hepatocellular carcinoma; LDLT, living donor liver transplantation.

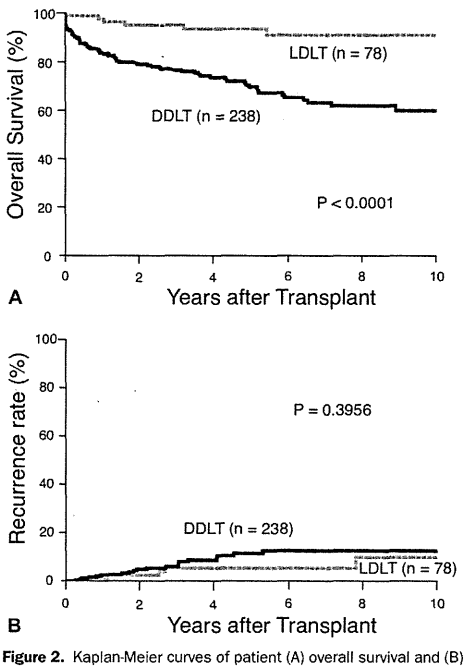

Figure 2. Kaplan-Meier curves of patient (A) overall survival and (B) HCC recurrence rate after liver transplantation for HCC patients within Milan criteria. Comparison between LDLT performed in Kyushu University Hospital in Japan (n = 78) and DDLT performed in the Mount Sinai Medical Center in New York (n = 238). DDLT, deceased donor liver transplantation; HCC, hepatocellular carcinoma; LDLT, living donor liver transplantation.

A similar trend was noted on comparison according to Milan criteria (Figs. 2 and 3). In the DDLT group, 124 of 362 patients (34.3%) were outside the Milan criteria, as compared with 55 of 133 patients (41.4%) in the LDLT group. In patients who fulfilled Milan criteria, the 1-, 3-, and 5-year overall survival rates were significantly better in the LDLT group than in the DDLT group (97.4%, 94.9%, and 93.3% in the LDLT group [n = 78] vs 83.5%, 76.4%, and 69.5% in the DDLT group [n = 238], respectively) ($p < 0.0001$).

Risk factors for hepatocellular carcinoma recurrence after LDLT and DDLT

Supplementary Table 1 (online only) shows the results of univariate analysis for factors associated with HCC recurrence in each group separately. Factors associated with more aggressive tumor were similarly included in both groups, but there were some factors that were specific to 1 group or the other, such as salvage transplantation in the LDLT group, and higher recipient BMI or more transfusion in the DDLT group.

On multivariate analysis, among the preoperative variables in the LDLT group, salvage transplantation and DCP levels over 300 mAU/mL were found to be independent predictive factors for HCC recurrence. Meanwhile, in the DDLT group, HCC beyond the Milan criteria was an independent preoperative predictive factor for HCC recurrence (Table 3).

Factors associated with post-transplant survival after LDLT and DDLT

Supplementary Table 2 (online only) shows the results of separate univariate analysis for variables associated with

post-transplant survival in each group. Factors associated with more aggressive tumors, greater blood loss, and microscopic vascular invasion were similarly included in both groups. In the DDLT group, higher recipient age, hepatitis C virus-related disease, higher MELD score, and more fresh frozen plasma/packed red blood cell transfusions were associated with decreased overall survival.

On multivariate analysis, HCC beyond the Milan criteria was an independent prognostic factor for survival in both groups (Supplementary Table 3, online only).

We performed univariate (Supplementary Table 4, online only) and multivariate (Table 4) analysis of the groups combined to identify any risk factor associated with the difference in survival between the LDLT and DDLT groups. On multivariate analysis, recipient BMI higher than 30 kg/m^2 was an independent predictive factor for survival ($p = 0.017$). Although mode of LT (ie, LDLT vs DDLT) was identified as a significant prognostic factor on univariate analysis, it was not prognostic for survival on multivariate analysis.

DISCUSSION

Among concerns regarding the use of LDLT to treat HCC is the tendency to include patients with higher tumor stage because LDLT tends to be applied to patients who are excluded from DDLT. In order to reduce this source of selection bias, we compared the results from 2 centers where each respective modality was the first treatment choice for unresectable HCC.

In a small single-center study of 60 HCC patients, Lo and colleagues[5] reported higher recurrence rates after LDLT when compared with DDLT. They suggested this difference was because of different baseline characteristics in the 2 groups, for example, more salvage transplantations in the LDLT group (10 of 43 LDLT vs 1 of 17 DDLT).[5] In our series, although multivariate analysis detected salvage transplantation as an independent predictor of tumor recurrence in the LDLT group, no difference was found for rates of salvage transplantation and recurrence between the LDLT and DDLT groups. Therefore, we concur with Lo and associates[5] that the higher recurrence in their series was caused by differences in baseline characteristics between groups. Another hypothetical concern with LDLT is that the rapid regeneration after partial liver grafting in LDLT may result in increased HCC recurrence due to effects on cell adhesion, angiogenesis, and cell migration.[19,20] In this study, we found similar rates of recurrence after LDLT and DDLT, despite the preferential use of smaller left-sided partial grafts for LDLT at Kyushu University.[21,22] In a recent multicenter study from the US (A2ALL cohort), Kulik and colleagues[23] reported that HCC recurrence rates in recipients receiving transplants after the introduction of MELD prioritization were similar between LDLT and DDLT after adjustment for tumor characteristics, although the unadjusted crude data were significantly different (38% in LDLT vs 11% in DDLT). These results support the idea that higher recurrence rates observed after LDLT in Western countries in other reports may be due to differences in baseline tumor characteristics. Although baseline characteristics were actually different in our series, comprehensive estimation of tumor aggressiveness, as represented by Milan criteria and microscopic vascular invasion, was similar between the 2 groups; it is therefore not surprising that recurrence rates were similar.

On multivariate analysis in the LDLT group, DCP level greater than 300 mAU/mL was an independent risk factor for HCC recurrence. Our previous study

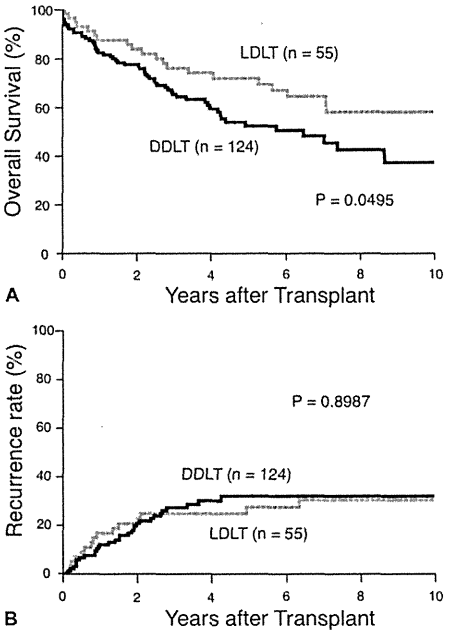

Figure 3. Kaplan-Meier curves of patient (A) overall survival and (B) HCC recurrence rate after liver transplantation for (B) HCC patients beyond Milan criteria. Comparison between LDLT performed in Kyushu University Hospital in Japan (n = 55) and DDLT performed in the Mount Sinai Medical Center in New York (n = 124). DDLT, deceased donor liver transplantation; HCC, hepatocellular carcinoma; LDLT, living donor liver transplantation.

Table 3. Multivariate Analysis of Factors Associated with Hepatocellular Carcinoma Recurrence after Living vs Deceased Donor Liver Transplantation for Hepatocellular Carcinoma Patients

	LDLT group (n = 133)		DDLT group (n = 362)	
Variables	**Risk ratio (95% CI)**	**p Value**	**Risk ratio (95% CI)**	**p Value**
Recipient BMI > 30, kg/m^2	–	–	1.96 (0.77–5.19)	0.158
Salvage transplantation	4.31 (1.19–27.7)	0.024	–	–
DCP > 300, mAU/mL	4.62 (1.70–12.4)	0.003	–	–
Beyond Milan criteria	2.02 (0.66–6.96)	0.220	3.37 (1.27–9.90)	0.014
PRBC transfusion >7, U	–	–	1.14 (0.42–3.43)	0.796
Microscopic vascular invasion	1.22 (0.41–3.75)	0.722	3.05 (1.09–9.05)	0.033
Poor differentiation	2.78 (1.01–8.18)	0.048	4.08 (1.27–11.3)	0.021

BMI, body mass index; DCP, des-gamma-carboxy prothrombin; DDLT, deceased donor liver transplantation; LDLT, living donor liver transplantation; PRBC, packed red blood cells.

demonstrated that high levels of DCP correlated with the presence of microscopic vascular invasion of HCC, and was therefore a powerful predictor of HCC recurrence after LDLT.[4] This might be a reason why microscopic vascular invasion, a well-known risk factor of HCC recurrence, was not predictive for HCC recurrence on multivariate analysis in the LDLT group, although it was in univariate analysis. Since 2007, Kyushu University has included the level of DCP as one of the eligibility criteria for LDLT treatment of HCC patients.

In contrast to similar tumor recurrence between LDLT and DDLT in our series, the overall survival of LDLT patients was significantly better than that of DDLT patients. Because tumor recurrence rates were comparable, the survival difference between LDLT and DDLT must be attributable to nontumor-related factors. Recipient BMI was significantly higher in the DDLT group, and it was detected as an independent risk factor for survival on multivariate analysis. Our group previously showed that higher recipient BMI was associated with significantly worse outcomes after LDLT.[24] Therefore, at Kyushu University, eligibility for LDLT recipients and donors was restricted to those with BMI preferably less than 30 kg/m^2. So, more stringent eligibility criteria for undergoing LDLT might have contributed to better survival after LDLT.

Table 4. Combined Multivariate Analysis of Factors Associated with Survival after Liver Transplantation for Hepatocellular Carcinoma Patients

Variables	**Risk ratio (95% CI)**	**p Value**
Recipient age over 60 y	1.10 (0.46–2.74)	0.826
Recipient BMI over 30, kg/m^2	4.62 (1.31–15.9)	0.017
MELD score	1.26 (0.33–3.76)	0.709
Beyond Milan criteria	2.09 (1.00–4.43)	0.052
Donor age over 60 y	1.28 (0.22–5.87)	0.769
Cold ischemic time, h	1.21 (0.92–1.62)	0.184
Blood loss over 3L	0.98 (0.41–2.33)	0.964
DDLT vs LDLT	0.26 (0.02–2.88)	0.279

For both living and deceased donors, n=495.
BMI, body mass index; DDLT, deceased donor liver transplantation; LDLT, living donor liver transplantation; MELD, model for end-stage liver disease.

The MELD score of the recipients in the DDLT group at listing was significantly higher than that in the LDLT group. While the pre-LT condition of the recipient certainly is a factor in post-LT survival, the mean MELD in the DDLT group was only 16, not a level associated with significantly decreased post-LT survival.

The primary reason for decreased survival in the DDLT group appears to be the effect of recurrent HCV infection of the allografts (data not shown). Donor age is a well-known determinant of the severity of post-LT, and 35% of the donors in the DDLT group were older than age 60. Other donor-related issues including, for example, steatosis, were not evaluated in this study but may also have contributed to increased impact of HCV recurrence after DDLT.

There are several limitations to our study. Because comparisons were between 2 centers in different countries, the differences in baseline characteristics were significant. Also, there might have been selection bias in both centers. Therefore, it should be noted that current results do not necessarily represent superiority of LDLT over DDLT in terms of survival benefits of HCC patients receiving LT. This is especially true when considering issues of donor risk in patients subjected to LDLT. However, important tumor characteristics such as Milan criteria and microscopic vascular invasion were not significantly different between the 2 groups. Therefore, we believe that it was acceptable to analyze, at least, the outcomes of HCC patients in these 2 population groups.

CONCLUSIONS

When compared between 2 centers where LDLT or DDLT was performed as the first treatment choice for

unresectable HCC, recurrence rates were similar and survival rates in the LDLT group were better compared with those in the DDLT group. Living donor liver transplantation is a viable treatment option for unresectable HCC, providing similar recurrence rates to those achieved with DDLT.

Author Contributions

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Analysis and interpretation of data: Ninomiya, Shirabe, Facciuto, Schwartz, Florman, Yoshizumi, Ikegami

Drafting of manuscript: Ninomiya, Facciuto, Schwartz, Harimoto, Ikegami, Uchiyama

Critical revision: Shirabe, Florman, Yoshizumi, Maehara

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Supplementary Table 1. Univariate Analysis of Factors Associated with Hepatocellular Carcinoma Recurrence after Living and Deceased Donor Liver Transplantation

	LDLT group (n = 133)		DDLT group (n = 362)	
Variables	**Risk ratio (95% CI)**	**p Value**	**Risk ratio (95% CI)**	**p Value**
Recipient male sex	0.92 (0.38–2.28)	0.854	0.99 (0.52–2.10)	0.986
Recipient age, y	0.98 (0.93–1.05)	0.634	1.02 (0.98–1.06)	0.310
Recipient BMI, kg/m^2	1.04 (0.89–1.22)	0.588	1.09 (1.01–1.18)	0.026
Etiology				
HBV	1.34 (0.38–3.66)	0.609	0.84 (0.35–1.75)	0.672
HCV	0.69 (0.27–1.94)	0.453	1.25 (0.72–2.22)	0.429
Others	1.47 (0.23–5.11)	0.623	0.84 (0.43–1.54)	0.590
MELD score	0.96 (0.86–1.05)	0.339	0.98 (0.94–1.02)	0.320
Salvage transplantation	4.76 (1.37–29.9)	0.011	1.31 (0.66–2.76)	0.447
Log AFP > 1.2	1.74 (0.70–4.91)	0.243	2.20 (1.27–3.83)	0.005
DCP > 300, mAU/mL	4.54 (1.83–11.0)	0.002	–	–
Beyond Milan criteria	4.92 (1.90–15.16)	<0.0001	3.42 (1.97–6.08)	<0.0001
Largest tumor diameter, cm	1.83 (1.30–2.52)	0.001	1.14 (1.03–1.24)	0.016
No. of tumor nodules	1.10 (1.07–1.14)	<0.0001	1.12 (1.03–1.19)	0.011
Bilobar HCC	3.23 (1.25–9.94)	0.015	1.58 (0.86–2.90)	0.142
Donor age, y	0.95 (0.90–1.00)	0.068	1.00 (0.98–1.01)	0.553
Donor BMI, kg/m^2	0.94 (0.79–1.10)	0.457	0.97 (0.90–1.03)	0.286
Cold ischemic time, min	0.99 (0.97–1.00)	0.206	1.00 (0.99–1.00)	0.550
Warm ischemic time, min	0.95 (0.88–1.02)	0.173	1.01 (0.98–1.04)	0.637
Blood loss, L	0.92 (0.78–1.05)	0.239	0.95 (0.79–1.07)	0.463
Blood transfusion, U				
PRBC	0.98 (0.94–1.02)	0.413	1.02 (1.00–1.04)	0.030
FFP	1.01 (0.99–1.03)	0.337	1.02 (0.99–1.05)	0.202
PC	0.99 (0.96–1.02)	0.641	1.01 (0.95–1.05)	0.726
Microscopic vascular invasion	3.29 (1.34–8.75)	0.009	5.79 (3.13–11.5)	<0.0001
Poor differentiation	4.21 (1.74–10.8)	0.002	3.66 (1.88–6.67)	0.001

AFP, alpha fetoprotein; BMI, body mass index; DCP, des-gamma-carboxy prothrombin; DDLT, deceased donor liver transplantation; FFP, fresh frozen plasma; LDLT, living donor liver transplantation; MELD, Model for End-Stage Liver Disease; PC, platelet concentrate; PRBC, packed red blood cells.

Supplementary Table 2. Univariate Analysis of Factors Associated with Survival after Living and Deceased Donor Liver Transplantation for Hepatocellular Carcinoma Patients

	LDLT group (n = 133)		DDLT group (n = 362)	
Variables	**Risk ratio (95% CI)**	**p Value**	**Risk ratio (95% CI)**	**p Value**
Recipient male sex	1.69 (0.76–4.13)	0.202	0.79 (0.54–1.20)	0.267
Recipient age, y	1.01 (0.96–1.07)	0.667	1.03 (1.00–1.05)	0.020
Recipient BMI, kg/m^2	1.10 (0.96–1.26)	0.174	1.04 (0.99–1.09)	0.094
Etiology				
HBV	0.72 (0.17–2.07)	0.577	0.59 (0.32–1.01)	0.057
HCV	1.00 (0.42–2.73)	0.996	1.42 (1.00–2.05)	0.050
Others	1.57 (0.37–4.51)	0.491	0.87 (0.58–1.28)	0.486
MELD score	1.02 (0.94–1.10)	0.584	1.02 (1.00–1.04)	0.048
Salvage transplantation	2.16 (0.88–6.48)	0.097	0.71 (0.48–1.08)	0.108
Log AFP > 1.2	1.66 (0.75–4.05)	0.220	1.27 (0.88–1.83)	0.207
DCP > 300, mAU/mL	3.49 (1.56–7.56)	0.003	–	–
Beyond Milan criteria	4.87 (2.07–13.3)	0.001	1.60 (1.13–2.26)	0.009
Largest tumor diameter, cm	1.39 (1.03–1.83)	0.033	1.06 (0.98–1.14)	0.153
No. of tumor nodules	1.06 (1.03–1.08)	0.001	1.05 (0.98–1.11)	0.181
Bilobar HCC	2.75 (1.20–7.04)	0.015	1.23 (0.82–1.85)	0.313
Donor age, y	1.01 (0.97–1.05)	0.716	1.00 (0.99–1.02)	0.172
Donor BMI, kg/m^2	0.92 (0.78–1.06)	0.235	0.96 (0.92–1.01)	0.096
Cold ischemic time, min	1.00 (0.98–1.01)	0.412	1.00 (1.00–1.01)	0.099
Warm ischemic time, min	1.00 (0.95–1.06)	0.861	0.99 (0.98–1.01)	0.554
Blood loss, L	1.10 (1.01–1.17)	0.028	1.08 (1.02–1.12)	0.008
Blood transfusion, U				
PRBC	1.02 (0.99–1.04)	0.289	1.02 (1.01–1.03)	0.003
FFP	1.00 (0.97–1.01)	0.682	1.02 (1.00–1.04)	0.025
PC	1.00 (0.98–1.02)	0.783	1.01 (0.98–1.04)	0.563
Microscopic vascular invasion	3.08 (1.40–7.24)	0.005	1.86 (1.31–2.64)	0.001
Poor differentiation	1.51 (0.66–3.28)	0.319	1.35 (0.74–2.28)	0.309

AFP, alpha fetoprotein; BMI, body mass index; DCP, des-gamma-carboxy prothrombin; DDLT, deceased donor liver transplantation; FFP, fresh frozen plasma; HBV, hepatitis B virus; HCV, hepatitis C virus; LDLT, living donor liver transplantation; MELD, Model for End-Stage Liver Disease; PC, platelet concentrate; PRBC, packed red blood cells.

Supplementary Table 3. Multivariate Analysis of Factors Associated with Survival after Living and Deceased Donor Liver Transplantation for Hepatocellular Carcinoma Patients

	LDLT group (n = 133)		DDLT group (n = 362)	
Variables	**Risk ratio (95% CI)**	**p Value**	**Risk ratio (95% CI)**	**p Value**
Recipient age > 56 y	–	–	1.49 (0.64–3.63)	0.353
Etiology HCV	–	–	0.78 (0.32–2.17)	0.606
MELD score >18	1.02 (0.94–1.10)	0.584	1.56 (1.24–5.87)	0.584
DCP > 300, mAU/mL	2.05 (0.84–4.95)	0.111	–	–
Beyond Milan criteria	3.46 (1.37–9.97)	0.008	3.62 (1.44–10.5)	0.005
Blood loss >3 L	1.35 (0.58–3.54)	0.498	1.76 (0.73–4.71)	0.213
Microscopic vascular invasion	1.55 (0.62–4.05)	0.349	2.26 (0.96–5.65)	0.063

DCP, des-gamma-carboxy prothrombin; DDLT, deceased donor liver transplantation; HCV, hepatitis C virus; LDLT, living donor liver transplantation; MELD, Model for End-Stage Liver Disease.

Supplementary Table 4. Combined Univariate Analysis of Factors Associated with Survival after Liver Transplantation for Hepatocellular Carcinoma Patients

Variables	Risk ratio (95% CI)	p Value
Recipient male sex	1.12 (0.78–1.62)	0.550
Recipient age, y	1.03 (1.01–1.05)	0.015
Recipient BMI, kg/m^2	1.08 (1.03–1.12)	0.001
Etiology		
HBV	0.62 (0.35–1.00)	0.050
HCV	1.20 (0.86–1.68)	0.284
Others	1.08 (0.73–1.54)	0.701
MELD score	1.03 (1.01–1.05)	0.004
Salvage transplantation	0.87 (0.60–1.26)	0.452
Log AFP > 1.2	1.18 (0.85–1.64)	0.332
Beyond Milan criteria	1.78 (1.30–2.43)	0.001
Largest tumor diameter, cm	1.10 (1.02–1.17)	0.018
No. of tumor nodules	1.04 (1.01–1.06)	0.003
Bilobar HCC	1.32 (0.93–1.89)	0.124
Donor age, yr	1.01 (1.00–1.02)	0.004
Donor BMI, kg/m^2	0.99 (0.95–1.03)	0.714
Cold ischemic time, h	1.09 (1.04–1.14)	0.001
Warm ischemic time, min	0.99 (0.98–1.01)	0.590
Blood loss, L	1.07 (1.02–1.11)	0.004
Blood transfusion, U		
PRBC	1.01 (1.00–1.03)	0.022
FFP	1.00 (0.98–1.01)	0.611
PC	0.98 (0.97–1.02)	0.035
Microscopic vascular invasion	2.00 (1.45–2.75)	<0.0001
Poor differentiation	1.06 (0.67–1.61)	0.799
DDLT vs LDLT	2.28 (1.52–3.55)	<0.0001

For both living and deceased donors, n=495.
AFP, alpha fetoprotein; BMI, body mass index; DDLT, deceased donor liver transplantation; FFP, fresh frozen plasma; HBV, hepatitis B virus; HCV, hepatitis C virus; LDLT, living donor liver transplantation; MELD, model for end-stage liver disease; PC, platelet concentrate; PRBC, packed red blood cells.

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ORIGINAL ARTICLE

Preventive effect of *Goshajinkigan* on peripheral neurotoxicity of FOLFOX therapy (GENIUS trial): a placebo-controlled, double-blind, randomized phase III study

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Abstract
Background Peripheral sensory neurotoxicity is a frequent adverse effect of oxaliplatin therapy. Calcium and magnesium (Ca/Mg) infusions are frequently used as preventatives, but a recent phase III trial failed to show that they prevent neurotoxicity. We therefore conducted a multicenter randomized phase III trial to compare fluorouracil, leucovorin, and oxaliplatin (mFOLFOX6) with and without *Goshajinkigan* (GJG), a traditional Japanese herbal medicine (Kampo), to determine GJG's potential for reducing peripheral neuropathy in patients with colorectal cancer.
Methods Patients with colon cancer who were undergoing adjuvant therapy with infusional mFOLFOX6 were randomly assigned to GJG (7.5 mg three times daily) or placebo in a double-blind manner. The primary endpoint was the time to grade 2 or greater neuropathy, which was determined at any point during or after oxaliplatin-based therapy using version 3 of the National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE).
Findings An interim analysis was performed when 142 of the planned 310 patients had been enrolled and the safety assessment committee recommended that the study be

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discontinued. One hundred eighty-two patients were evaluable for response. They included 89 patients in the GJG group and 93 patients in the placebo group. The incidence of grade 2 or greater neurotoxicity was 50.6 % in the GJG group and 31.2 % in the placebo group. A Cox proportional hazards analysis indicated that the use of GJG was significantly associated with the incidence of neuropathy (hazard ratio, 1.908; $p = 0.007$).

Conclusion *Goshajinkigan* did not prevent oxaliplatin-associated peripheral neuropathy in this clinical trial. The clinical study was therefore terminated.

Keywords Adjuvant chemotherapy · Colon cancer · Colorectal cancer · *Goshajinkigan* · Herbal medicine, Kampo · Peripheral neuropathy

Introduction

In recent years, the standard chemotherapy for advanced/recurrent colorectal cancer has been continuous intravenous infusion of 5-fluorouracil (5-FU) or oral 5-FU derivatives combined with either oxaliplatin [CapeOX, FOLFOX4, or modified FOLFOX6 (mFOLFOX6)] or irinotecan (CapeIRI, FOLFIRI) [1–5]. In a pivotal phase III trial, FOLFOX was found to be superior to fluorouracil and leucovorin (FU/LV) in patients with resected stage III colon cancer in disease-free survival and overall survival [6]. However, peripheral neuropathy is a complication of oxaliplatin therapy, and therefore the oxaliplatin dose must be limited to avoid toxicity.

The first strategy for avoiding oxaliplatin-linked neuropathy involves stop-and-go regimens such as the OPTIMOX series, which includes oxaliplatin-free intervals to reduce grade 3 sensory neuropathy [7]. This stop-and-go regimen avoids the problem of oxaliplatin-induced neurotoxicity by using the dose-intense FOLFOX7 regimen for a defined period, stopping the therapy before severe neurotoxicity develops and then reintroducing the same regimen. However, this is unsuitable as an adjuvant treatment, and is instead used for patients with recurrent or nonresectable cancer. As an alternative, neurologic symptoms may be reduced by administering agents such as calcium (Ca) and magnesium (Mg) preparations [8]. Gamelin et al. [9, 10] reported that the administration of calcium gluconate and magnesium sulfate (Ca/Mg) before and after oxaliplatin therapy alleviated peripheral neurotoxicity. Other similar treatments have been described, which include carbamazepine [11] and glutathione [12], but no effective remedy for oxaliplatin-induced peripheral neurotoxicity has been established to date.

Goshajinkigan (GJG), a traditional Japanese herbal medicine (Kampo), is composed of 10 crude herbs: *Rehmanniag lutinosa*, *Achyranthes* spp. root, *Cornus officinalis*, *Dioscorea* spp. rhizome, *Plantago* spp. seed, *Alisma orientale*, *Porica cocos*, *Moutan* cortex, *Cinnamomum cassia*, and aconite tuber [13–15]. In Japan, GJG is primarily used to improve symptoms such as numbness, cold sensation, and limb pain associated with diabetic neuropathy [13, 16]. *Goshajinkigan* is also reportedly useful for coping with paclitaxel- and oxaliplatin-induced peripheral neuropathy [17, 18]. Furthermore, Kono et al. [19] recently reported that oxaliplatin-induced peripheral neurotoxicity was relieved by the administration of GJG in patients with advanced colorectal cancer who were receiving FOLFOX therapy. The GJG group experienced an improvement in peripheral neurotoxicity, and these patients tended to receive more oxaliplatin before peripheral neurotoxicity developed [19]. To confirm the preventive effect of GJG on oxaliplatin-induced peripheral neurotoxicity, we conducted a double-blind, placebo-controlled, multicenter, randomized phase III trial of GJG as an adjuvant therapy for patients with resected stage III colon cancer.

Methods

Eligibility criteria

Patients who had histologically confirmed adenocarcinoma of colorectal cancer, with the lower edge of the tumor located at a site above the pouch of Douglas, were included if they met all of the following criteria: they had an Eastern Cooperative Oncology Group performance status (ECOG PS) of 0 or 1; their cancer status was pathological stage III (based on the 7th edition of the Japanese Classification of Colorectal Carcinoma) [20]; they had undergone complete radical resection (R0); the surgery had taken place within 8 weeks; and they had adequate hepatic, renal, respiratory, and bone marrow function. All patients provided written informed consent before their enrollment in the study. Patients were not allowed to participate in the trial if they had pre-existing peripheral neuropathy of any grade. The study was approved by the institutional review boards of all

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participating institutions, and was registered in the University Hospital Medical Information Network (UMIN) clinical trial registry (UMIN000004282).

Treatment schedule and investigational medicinal products

Patients and all clinical study personnel who interacted with them were blinded to the treatment arm. Patients were allocated randomly to a 12-course mFOLFOX6 regimen with GJG at 7.5 g/day (Tsumura and Co., Akasaka, Japan) or an identical placebo. The quality of the investigational medicinal products (i.e., the placebo and GJG) was approved by the Quality Assurance Unit of Tsumura in accordance with current good manufacturing practice (cGMP). Tsumura prepared and stored GJG. The placebo was prepared and stored by Yamato Logistics (Tokyo, Japan) in a room in which humidity and temperature were managed by a pharmacist during the study period. The designated numbered drug was sent to each institution at every registration. The drug was handled by a management representative (but not the researchers) in each institution. Storage, shipment, receipt at the company, management, use, and remaining checks at each institution were managed by an electronic data capture (EDC) system. *Goshajinkigan* and the placebo were administered on the first day of mFOLFOX6 therapy and continued to be administered orally before meals or between meals on a daily basis until the end of the 12 courses. Other sensory neuromodulatory agents such as calcium–magnesium infusions or antiepileptic agents were forbidden. The mFOLFOX6 chemotherapy regimen consisted of a 2-h intravenous infusion of oxaliplatin (85 mg/m^2) combined with l-LV (100 mg/m^2), followed by a rapid intravenous infusion of 5-FU (400 mg/m^2), and then a 46-hour continuous infusion of 5-FU (2400 mg/m^2). This regimen comprised one course of therapy and was repeated once every 2 weeks. Oxaliplatin dose modifications and skipping were not allowed for patients who experienced grade 1 neurotoxicity. However, a dose reduction of oxaliplatin to 75 mg/m^2 was allowed for patients who experienced persistent grade 2 sensory neurotoxicity.

Endpoints

The primary endpoint was the time to the onset of grade 2 or greater sensory neurotoxicity [i.e., time to neuropathy (TTN)] during therapy. Primary neuropathy was assessed using the National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE; version 3.0); the Neurotoxicity Criteria of Debiopharm (DEB-NTC) was used for comparison [21]. Standardized questions regarding symptoms of neurotoxicity and examples of answers were used to facilitate the more accurate classification of patient-reported symptoms as grade 1, 2, 3, or 4. These grades were determined by physicians from patient records.

Statistical considerations

Eligible patients were assigned randomly to receive GJG or placebo in the ratio 1:1. Treatment allocation was stratified by institution, sex, age (<65 and >65 years). The EDC system was used to screen patients for treatment allocation. Patient and drug identification numbers were allocated sequentially in the order in which the patients were enrolled.

A two-arm, randomized, placebo-controlled, double-blind, phase III design was employed. The TTN was compared between groups using Kaplan–Meier survival curves and log-rank testing. In the MOSAIC trial, among the patients receiving FOLFOX, and 44 % experienced grade 2 and 48 % experienced grade 1 sensory neurotoxicity, respectively. In Japan, the Kyushu Study Group of Clinical Cancer 0501 (KSCC0501) (FOLFOX4) and SWIFT2 (mFOLFOX6) prospective studies reported grade 2 or greater sensory neurotoxicity in 43 % and 26 % of the patients, respectively. Thus, the cumulative incidence of peripheral neuropathy was approximately 40 % after the 12-course chemotherapy regimen among patients in the placebo group in this study. We expected the cumulative incidence of peripheral neuropathy in the GJG group to be 25 %. Ninety-five cases per arm were required for the log-rank test to detect a difference of 15 % with 80 % power, and 291 cases were needed to achieve this during the 6-month follow-up. The original study design thus included 155 patients per arm. The secondary endpoints were of the proportions of patients who reported adverse events and the dose intensity of oxaliplatin, both of which were also compared between the groups. The EDC system was used for patient enrollment and for the preparation of a case report form (CRF) from start to finish. The EPS Corporation (Tokyo, Japan) conducted central monitoring every 6 months to manage quality control and guarantee quality. The statistical section of the EPS Corporation was in charge of statistical analyses under the direction of YT (who was the representative of statistics management). An interim analysis was scheduled, based on the data for one-half of the patients (i.e., 150 patients). The purpose of the interim analysis was to curtail the recruitment when we found that the *Goshajinkigan* arm was superior to the control arm, with the significance level determined by the Lan–DeMets alpha spending function (i.e., O'Brein–Fleming type), or the futility of the *Goshajinkigan* arm given the low value of the Bayesian predictive power. Stopping boundaries according to the Lan–DeMets function were computed using the East software, version 5.3 (Cytel, Cambridge, MA, USA). Statistical analysis of the study data was conducted using SAS software, version 9.2 (SAS Institute, Cary, NC, USA).