J Hepatobiliary Pancreat Sci (2017) 24:49–57
DOI: 10.1002/jhbp.412

ORIGINAL ARTICLE

Significance of preoperative fluorodeoxyglucose-positron emission tomography in prediction of tumor recurrence after liver transplantation for hepatocellular carcinoma patients: a Japanese multicenter study

Yasutsugu Takada · Toshimi Kaido · Ken Shirabe · Hiroaki Nagano · Hiroto Egawa · Yasuhiko Sugawara · Akinobu Taketomi · Takeshi Takahara · Go Wakabayashi · Chikashi Nakanishi · Naoki Kawagishi · Akira Kenjo · Mitsukazu Gotoh · Yoshikazu Toyoki · Kenichi Hakamada · Masayuki Ohtsuka · Nobuhisa Akamatsu · Norihiro Kokudo · Kazuhisa Takeda · Itaru Endo · Hiroyuki Takamura · Hideaki Okajima · Hiroshi Wada · Shoji Kubo · Kaoru Kuramitsu · Yonson Ku · Kohei Ishiyama · Hideki Ohdan · Eitaro Ito · Yoshihiko Maehara · Masaki Honda · Yukihiro Inomata · Hiroyuki Furukawa · Shinji Uemoto · Hiroki Yamaue · Masaru Miyazaki · Tadahiro Takada · on behalf of the LTx-PET study group of the Japanese Society of Hepato-Biliary-Pancreatic Surgery and the Japanese Liver Transplantation Society

The author's affiliations are listed in the Appendix.

Correspondence to: Yasutsugu Takada, Department of HBP and Breast Surgery, Ehime University Graduate School of Medicine, Shitsukawa, Toon, Ehime 791-0295, Japan.
e-mail: takaday@m.ehime-u.ac.jp

DOI: 10.1002/jhbp.412

Abstract

Background In the present study, we conducted a multicenter nationwide survey to investigate the effects of preoperative fluorine-18-fluorodeoxyglucose (FDG) positron emission tomography (PET) on the prediction of hepatocellular carcinoma (HCC) recurrence after liver transplantation (LT).

Methods From 16 Japanese LT centers, data were collected on 182 recipients with HCC who underwent living donor liver transplantation (LDLT) between February 2005 and November 2013. PET-positive status was defined as increased uptake of FDG in the tumor compared to the surrounding non-tumor liver tissue. The median follow-up after LDLT was 54.5 months (range 1–125 months).

Results Postoperative HCC recurrence occurred in 23 patients. Multivariate analysis revealed that exceeding the Milan criteria (MC), alpha-fetoprotein (AFP) level ≥115 ng/ml, and PET-positive status were significant and independent risk factors for recurrence. In the over-MC group, a subgroup of patients with AFP level <115 ng/ml and PET-negative status (n = 22) had a significantly lower 5-year recurrence rate than the other patients (n = 27, 19% vs. 53%, P = 0.019).

Conclusions These results suggest that preoperative FDG-PET status offers additional information on HCC recurrence risk after LT. Over-MC patients with PET-negative status and lower AFP level may achieve successful outcome comparable to that of within-MC patients.

Keywords Biological marker · Hepatocellular carcinoma · Liver transplantation · Selection criteria

Introduction

Liver transplantation (LT) has proved to be the treatment of choice for early-stage hepatocellular carcinoma (HCC) in cirrhotic patients. The Milan criteria (MC), proposed by Mazzaferro et al. [1], have been widely accepted as an effective way of selecting patients with early-stage HCC for curative LT. However, it is well recognized that a

substantial subset of patients who exceed the MC, and who have the potential for good outcome after LT, exists. Thus, many centers have performed LT using extended criteria beyond the MC [2–9] in an attempt to afford the chance of cure to patients outside the MC.

Although these extended criteria are based on tumor number and size, which are used as an estimate of tumor burden and serve as a surrogate for tumor characteristics, these criteria are considered insufficient to reasonably predict the risk of recurrence. Accordingly, to define optimal extended criteria beyond the MC, it has been advocated that additional parameters for tumor biological features related to risk of recurrence are necessary [10, 11]. In addition, for patient selection criteria before LT, only tumor variables that can be measured in preoperative evaluation should be incorporated.

Recently, fluorine-18-fluorodeoxyglucose (^{18}F–FDG) positron emission tomography (PET) has become a standard procedure in the diagnosis and staging of several malignancies, such as malignant lymphoma, pancreatic cancer, and metastatic liver cancers [12]. It has been indicated that, although well-differentiated HCC cells exhibit an FDG metabolism comparable to that of normal liver tissue, glucose-6-phosphatase activities are much reduced in poorly differentiated HCC cells, leading to enhanced accumulation of ^{18}F–FDG similar to that in metastatic liver tumors [13]. Hence, FDG-PET is expected to describe tumor aggressiveness of HCC. In a clinical approach, preoperative FDG-PET has been shown to predict tumor differentiation and outcome after liver resection in patients with HCC [14–16]. In liver transplantation, Kornberg et al. [17] first reported that increased ^{18}F–FDG tumor uptake on preoperative PET scans is significantly associated with tumor microvascular invasion on explant pathology as well as with post-transplant outcomes in recipients with HCC. Thereafter, some studies [18–21] have suggested the usefulness of pre-transplant FDG-PET to predict the post-transplant recurrence of HCC. However, these studies were based on single-center experiences with small sample size. Therefore, we conducted a multicenter, retrospective survey to investigate the effects of FDG-PET on the prediction of HCC recurrence after living donor liver transplantation (LDLT) and to endeavor to construct new selection criteria incorporating FDG-PET.

Patients and methods

Patients

Data were collected on 182 recipients who had received preoperative FDG-PET and underwent LDLT between February 2005 and November 2013 at the following 16 Japanese institutions: Kyoto University, Kyushu University, Hiroshima University, Kobe University, Tohoku University, Yokohama City University, Iwate Medical University, Osaka University, Ehime University, Fukushima Medical University, Tokyo University, Hirosaki University, Chiba University, Kanazawa University, Osaka City University, and Kumamoto University. For all patients, viable HCC was pathologically confirmed in the explanted liver. Patient selection for LDLT was based on the criteria at each institution. However, in principle, patients with extrahepatic metastasis or radiological vascular invasion were excluded in all centers. As of the end of June 2015, the median duration of follow-up was 54.5 months (range 1–125 months). The protocol of this study was approved by the medical ethics committee of each participating institute. This study was conducted in collaboration with the Japanese Society of Hepato-Biliary-Pancreatic Surgery and the Japanese Liver Transplantation Society.

Preoperative tumor variables

Staging of HCC was based on pre-transplant imaging. Evaluation of the extent of tumor involvement using contrast-enhanced multidetector computed tomography (MDCT) was usually performed within 1 month before LDLT. Tumor staging was determined by counting only viable and enhancing nodules on MDCT. Individual HCC was staged according to MC [1], as solitary tumor ≤5 cm, or ≤3 nodules all ≤3 cm. A total of 133 patients (73%) met MC, and 49 patients (27%) exceeded the criteria.

Tumor markers such as des-gamma-carboxy prothrombin (DCP), also known as protein induced by vitamin K absence or antagonist II (PIVKA-II), and alpha-fetoprotein (AFP) were measured within 1 month prior to LDLT. Serum DCP levels were determined by electrochemiluminescence immunoassay (Picolumi PIVKA-II kit; Sanko Junyaku, Tokyo, Japan).

As extended tumor number-size criteria, the University of California, San Francisco (UCSF) criteria [5, 22] (solitary tumor ≤6.5 cm, or ≤3 nodules all ≤4.5 cm and total tumor diameter ≤ 8 cm) and the Up-To-Seven criteria [23] (sum of the size of the largest tumor and the number of tumors ≤7) were evaluated. In addition, the effects of some criteria incorporating AFP or DCP were investigated, including the Kyoto criteria [9, 24] (tumor size ≤5 cm, ≤10 nodules, and DCP ≤400 mAU/ml), the Kyushu criteria [25] (tumor size <5 cm or DCP <300 mAU/ml), and the modified Tokyo criteria [26] (≤5 nodules all ≤5 cm, AFP ≤250 ng/ml, and DCP ≤450 mAU/ml).

FDG-PET

Pre-transplant ^{18}F–FDG PET was performed according to the protocol of each institute. Patients with diabetes mellitus

or hyperglycemia at fasting, who were not considered appropriate for FDG-PET study, were excluded from the present study. The records of semiquantitative analysis of ^{18}F–FDG uptake and the maximum standardized uptake value (SUV) for tumor and non-tumor liver tissues were collected [15]. "PET-positive" status was defined as increased uptake of FDG in the tumor compared to the surrounding non-tumor liver tissue by semiquantitative analysis. In patients with multiple HCC lesions, only the one showing the highest SUVmax was included for analysis. The PET image was interpreted by the specialist in nuclear medicine (radiologist) at each institute.

Histological analysis

Histological examination was performed on the explanted liver. Viable HCC nodules were confirmed in all patients. Each HCC was histologically graded into one of three categories according to the modified Edmondson criteria: well differentiated; moderately differentiated; or poorly differentiated. When the tumor consisted of ≥2 grades of histological differentiation, the most progressive grade was used. Microvascular invasion (histological invasion to the portal and/or hepatic vein) were also investigated.

Statistical analysis

For selection of optimal cut-off values to evaluate the ability of AFP and DCP to predict postoperative recurrence, receiver operating characteristic (ROC) analysis was used, as described elsewhere [24]. Cumulative overall survival and recurrence rates were calculated using Kaplan–Meier methods, and differences between curves were evaluated using log-rank testing. The χ^2 test was used to compare differences between the two groups. The Cox proportional hazards model was used to identify the independent risk factors for recurrence. Values of $P < 0.05$ were considered significant. All statistical analyses were performed using IBM SPSS Statistics version 22.0 for Macintosh (IBM, Armonk, NY, USA).

Results

Preoperative patient profile

Patient profiles and pre-transplant clinical characteristics are shown in Table 1. The median age of the 182 study patients (118 men, 64 women) was 58 years. One hundred and fifteen patients (63%) displayed a history of previous treatment for HCC using various non-transplant methods including transcathter arterial chemoembolization (TACE), percutaneous ablation therapy, or hepatic resection. The Child–Pugh class was grade A for 21 patients, B for 88 patients, and C for 73 patients. Hepatitis C was the most common etiological factor, followed by hepatitis B. The median size and number of HCCs detected on pre-transplant imaging were 2.2 cm (range 0.5–8.8 cm) and 2 (range 1–186), respectively. The median serum AFP was 29.4 ng/ml, and the median serum DCP was 53 mAU/ml. FDG-PET status was positive for 43 patients and negative for 139 patients.

As for the donors, the median age was 40 years, and the male-to-female ratio was 103 to 79. ABO blood type mismatch transplantation was performed for 33 cases (18%).

Table 1 Patient characteristics

Variable	Value
Age (years)	58 (36–72)[a]
Sex (male/female)	118/64
History of previous treatment for HCC (yes/no)	115/67
Child–Pugh class (A/B/C)	21/88/73
MELD score	15 (3–39)[a]
Etiology	
HCV	107
HBV	49
HCV and HBV	2
Alcohol	12
PBC	3
Others	9
Tumor size (cm)	2.2 (0.5–8.8)[a]
Tumor number	2 (1–186)[a]
AFP (ng/ml)	29.4 (1–28,074)[a]
DCP (mAU/ml)	53 (5–10,950)[a]
FDG-PET status (positive/negative)	43/139
Donors	
Age (years)	40 (20–64)[a]
Sex (male/female)	103/79
ABO blood type mismatch (yes/no)	33/149

AFP alpha-fetoprotein, *DCP* des-gamma-carboxy prothrombin, *FDG-PET* fluorine-18-fluorodeoxyglucose positron emission tomography, *MELD* model for end-stage liver disease

[a]Data are given as median (range)

Postoperative patient survival

As of the end of June 2015, a total of 127 patients remained alive, with 19 patients dying of recurrent HCC and 36 patients dying of tumor-unrelated causes. The overall patient survival rate was 71% at 5 years after LDLT. As shown in Table 2, pre-transplant tumor number and tumor size were significant risk factors for overall patient survival.

Table 2 Univariate analyses of overall survival and preoperative tumor variables

Preoperative tumor variable (*n*)	*P*	Overall survival rate (%)		
		1 year	3 years	5 years
Tumor number	0.018			
≤3 (143)		85	78	75
≥4, ≤10 (33)		79	63	60
≥11 (6)		83	50	33
Tumor size	<0.001			
≤3 cm (142)		88	81	77
>3 cm, ≤5 cm (33)		79	61	57
>5 cm (7)		29	0	0
Milan criteria	0.007			
Meeting (133)		85	79	77
Exceeding (49)		82	61	54
AFP	0.104			
≤100 ng/ml (129)		85	78	74
>100 ng/ml, ≤400 ng/ml (27)		89	78	73
>400 ng/ml (26)		73	53	53
DCP[a]	0.742			
≤100 mAU/ml (110)		86	76	73
>100 mAU/ml, ≤400 mAU/ml (39)		82	71	67
>400 mAU/ml (32)		81	70	66
FDG-PET status	0.043			
Negative (139)		86	77	75
Positive (43)		77	64	58

AFP alpha-fetoprotein, *DCP* des-gamma-carboxy prothrombin, *FDG-PET* fluorine-18-fluorodeoxyglucose positron emission tomography
[a]The result was missing for one patient

Five-year survival rates were 77% for patients meeting MC and 54% for those exceeding MC ($P = 0.007$). Although AFP and DCP levels were not associated with overall survival, the 5-year survival rate for PET-negative (PET−) patients was significantly higher than for PET-positive (PET+) patients (75% vs. 58%, $P = 0.043$).

Postoperative HCC recurrence and risk factors

Postoperative recurrence of HCC was identified in 23 patients. Univariate analysis revealed that tumor number, tumor size, and MC were significant risk factors for recurrence (Table 3). The recurrence rate for patients exceeding MC was 38% at 5 years and significantly higher than for patients meeting MC (7% at 5 years, $P < 0.001$). For ROC analysis to determine cut-off values for AFP and DCP, only patients followed for ≥24 months without HCC recurrence were considered to be free of recurrence, since 19 of the 23 recurrences (83%) occurred within 24 months after LDLT. ROC analysis was performed using 23 patients with recurrence and 119 patients free of recurrence ≥24 months. The selected cut-off values were 115 ng/ml for AFP [(sensitivity, specificity) = (0.52, 0.76)] and 78 mAU/ml for DCP (0.65, 0.64). Using these cut-off values, patients with higher AFP as well as those with higher DCP levels demonstrated significantly higher recurrence rates than patients with lower levels (Table 3). In addition, pre-transplant PET status proved a significant risk factor for recurrence. The 5-year recurrence rates were 12% for PET− and 28% for PET+ patients, respectively ($P = 0.007$).

Multivariate analysis was performed by using the Cox proportional hazards model. Because the MC are most widely used as tumor number-size criteria, MC in addition to AFP, DCP, and PET status were incorporated into the analysis. As shown in Table 4, multivariate analysis demonstrated that exceeding MC ($P < 0.001$), AFP ≥115 ng/ml ($P = 0.008$), and PET+ ($P = 0.029$) were independently associated with recurrence, whereas DCP ≥78 mAU/ml was not. Exceeding MC was the strongest predictor of recurrence (relative risk = 5.309, 95% confidence interval (CI): 2.165–13.020).

New criteria incorporating AFP and PET

From the viewpoint of extension beyond MC, the study patients were stratified into three groups based on the

Table 3 Univariate analyses of hepatocellular carcinoma (HCC) recurrence rate on preoperative tumor variables

Preoperative tumor variable (*n*)	*P*	Recurrence rate (%)		
		1 year	3 years	5 years
Tumor number	<0.001			
≤3 (143)		3	7	7
≥4, ≤10 (33)		20	31	35
≥11 (6)		17	50	67
Tumor size	<0.001			
≤3 cm (142)		3	8	10
>3 cm, ≤5 cm (33)		10	26	26
>5 cm (7)		57	100	–
Milan criteria	<0.001			
Meeting (133)		3	7	7
Exceeding (49)		17	33	38
AFP[a]	0.002			
<115 ng/ml (134)		3	9	11
≥115 ng/ml (48)		16	26	29
DCP[a,b]	0.015			
<78 mAU/ml (103)		6	8	9
≥78 mAU/ml (78)		7	23	26
FDG-PET status	0.007			
Negative (139)		4	10	12
Positive (43)		15	28	28

AFP alpha-fetoprotein, *DCP* des-gamma-carboxy prothrombin, *FDG-PET* fluorine-18-fluorodeoxyglucose positron emission tomography

[a]Cut-off levels were determined based on the ROC analysis

[b]The result was missing for one patient

Table 4 Multivariate Cox proportional hazard analysis of recurrence of hepatocellular carcinoma (HCC) with preoperative variables

Preoperative variable	Relative risk	95% CI	*P*
Exceeding Milan criteria	5.309	2.165–13.020	<0.001
AFP ≥115 ng/ml	3.077	1.748–7.023	0.008
DCP ≥78 mAU/ml	1.774	0.736–4.278	0.202
FDG-PET positive	2.554	1.101–5.924	0.029

AFP alpha-fetoprotein, *CI* confidence interval, *DCP* des-gamma-carboxy prothrombin, *FDG-PET* fluorine-18-fluorodeoxyglucose positron emission tomography

[a]The analysis was performed for 181 patients after excluding one patient without DCP result

three independent risk factors for recurrence: Group A, patients within MC (n = 133); Group B, patients beyond MC but with AFP <115 ng/ml and PET− (n = 22); and Group C, patients beyond MC with AFP ≥115 ng/ml and/or PET+ (n = 27). The recurrence rates for these groups are shown in Figure 1. The 5-year recurrence rates were not significantly different between Groups A and B (6% vs. 19%, P = 0.176). However, the recurrence rate was significantly higher for Group C (53%) than for the other two groups (vs. Group A, P < 0.001; vs. Group B, P = 0.012).

Based on these results, by combining Groups A and B, we defined new extended criteria as meeting MC, or beyond MC but with AFP <115 ng/ml and PET−. The 5-year recurrence rate for patients meeting the new criteria was significantly lower than for patients beyond the new criteria (8% vs. 53%, P < 0.001). The 5-year survival rate for patients meeting the new criteria was significantly better than for patients beyond the new criteria (75% vs. 44%, P = 0.003, Fig. 2).

Histological features and outcomes

On explant pathology, histological grade was well differentiated in 28 patients (15%), moderately differentiated in 127 patients (70%), and poorly differentiated in 27 patients (15%). Microvascular invasion was present in 42 patients (23%). The 1-, 3-, and 5-year recurrence rates for patients with well or moderately differentiated tumors were significantly lower than those for patients with poorly differentiated tumors (4% vs. 20%, 10% vs. 34%, 13% vs. 34%, respectively, P = 0.002). Similarly, the 1-, 3-, and 5-year recurrence rates for patients without microvascular invasion were significantly lower than for patients with microvascular invasion (2% vs. 21%, 8% vs. 32%, 9% vs. 36%, respectively, P < 0.001).

Various criteria in relation to recurrence rates and histological features

We applied various criteria of OLT for HCC to the patients of the present study, and investigated the effects on the ability to discriminate the patients at higher risk of recurrence. As shown in Table 5, each tumor number-size set of criteria (MC, UCSF, Up-To-Seven), each set of criteria incorporating tumor markers (Kyoto, modified Tokyo), and the present new criteria incorporating AFP and PET status could significantly differentiate the patients with higher recurrence rates from those with lower rates. As for correlations between the two histological features (microvascular invasion and tumor grade) and the various criteria, the MC, UCSF, and Kyoto criteria were significantly associated with positive microvascular invasion, while the UCSF, Kyoto and modified Tokyo criteria were significantly associated with prevalence of poorly differentiated grade. Contrasting with these results, when comparing the 155 study patients who met the present criteria and the 27 study patients who exceeded the present criteria, differences in prevalence of positive microvascular invasion and poorly differentiated grade

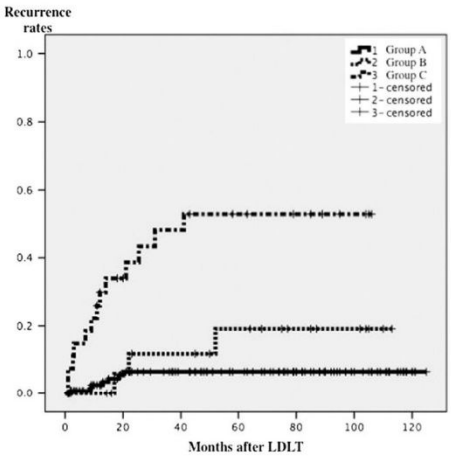

Fig. 1 Post-transplant hepatocellular carcinoma (HCC) recurrence rates. Group A: patients within Milan criteria (MC) (n = 133); Group B: patients beyond MC but with alpha-fetoprotein (AFP) <115 ng/ml and positron emission tomography (PET)− (n = 22); and Group C: patients beyond MC with AFP ≥115 ng/ml and/or PET+ (n = 27). Five-year recurrence rates were not significantly different between Groups A and B (6% vs. 19%, P = 0.176). However, the recurrence rate was significantly higher for Group C (53%) than for the other two groups (vs. Group A, P < 0.001; vs. Group B, P = 0.012)

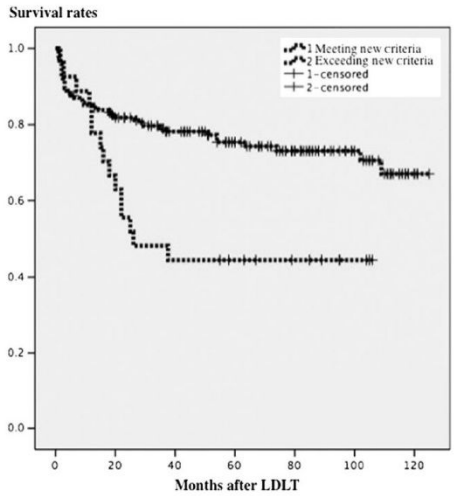

Fig. 2 New criteria and post-transplant survival rates. The new criteria were defined as meeting Milan criteria (MC), or beyond MC but with alpha-fetoprotein (AFP) <115 ng/ml and positron emission tomography (PET)−. The 5-year survival rate for the patients (n = 155) meeting the new criteria was significantly higher than for the patients (n = 27) exceeding the new criteria (75% vs. 44%, P = 0.003)

were both highly significant (P = 0.004; P = 0.003, respectively).

Discussion

Since the first report by Kornberg et al. [17], some institutions [18–21] have reported the usefulness of preoperative FDG-PET in the assessment of tumor aggressiveness and in the prediction of tumor recurrence after LT. Lee et al. [18] performed a retrospective analysis in 110 adult LDLT recipients with HCC with reference to pre-transplant PET. Nineteen (24.4%) of the 78 patients within MC had positive PET scans, while 15 (46.9%) of the 32 patients beyond MC had positive PET scans. Among the patients beyond MC, the 2-year recurrence rates were significantly higher in the PET-positive than in the PET-negative patients (80.0% vs. 29.4%, P = 0.032). In the updated report by Kornberg et al. [20], the 5-year recurrence-free survival rates for recipients (n = 20) with HCC that were over MC but were PET-negative were comparable to those for recipients within MC (n = 57, 81% vs. 86.2%), but these rates were significantly higher than the rates for over MC and PET-positive recipients (n = 14, 21%, P = 0.002). In parallel with these studies, the present study demonstrated that pre-transplant PET status is a significant and independent risk factor for post-transplant recurrence of HCC.

In the present study, among preoperative variables, exceeding MC was the strongest predictor of recurrence. However, even patients beyond MC showed a survival rate of 54% with a recurrence-free rate of 32% at 5 years. These figures suggest that a subgroup with good outcomes is included in the over-MC patients. So far, several extended criteria have been reported in an attempt to identify such a subgroup [22–30]. As shown in Table 5, the recurrence rates for patients meeting the UCSF or Up-To-Seven criteria were significantly lower than for patients beyond the criteria and similar to the recurrence rates for within-MC patients (6% and 7% at 5 years, respectively). However, the patients meeting the UCSF (n = 139) and Up-To-Seven criteria (n = 144) included only six (12%) and 11 (22%) of 49 patients beyond the MC. Recently, several studies have indicated the efficacy of selection criteria incorporating tumor markers such as AFP and DCP [23–31]. In Table 5, the Kyoto, Kyushu, and modified Tokyo criteria – three Japanese extended criteria incorporating the tumor markers – included 28 (57%), 46 (94%), and 31 (63%) of 49 patients beyond the MC, respectively,

Table 5 Various criteria in relation to recurrence rates and histological features

	Recurrence rate (%)				Positive microvascular invasion			Poorly differentiated grade		
Preoperative criteria (*n*)	1 year	3 years	5 years	*P*	%	Number	*P*	%	Number	*P*
Milan criteria										
Meeting (133)	3	7	7	<0.001	18.0	24 of 133	0.008	12.0	16 of 133	0.079
Exceeding (49)	17	33	38		36.9	18 of 49		22.4	11 of 49	
UCSF criteria										
Meeting (139)	2	6	6	<0.001	18.7	26 of 139	0.012	11.5	16 of 139	0.023
Exceeding (43)	20	37	43		37.2	16 of 43		25.6	11 of 43	
Up-To-Seven criteria										
Meeting (144)	3	7	7	<0.001	20.1	29 of 144	0.067	12.5	18 of 144	0.084
Exceeding (38)	20	39	46		34.2	13 of 38		23.7	9 of 38	
Kyoto criteria[a]										
Meeting (161)	4	10	11	<0.001	19.9	32 of 161	0.011	13.0	21 of 161	0.045
Exceeding (20)	26	46	53		45.0	9 of 20		30.0	6 of 20	
Kyushu criteria[b]										
Meeting (179)	6	14	15	0.131	22.9	41 of 179	0.671	14.5	26 of 179	0.363
Exceeding (3)	33	33	33		33.3	1 of 3		33.3	1 of 3	
Modified Tokyo criteria[c]										
Meeting (164)	5	10	12	<0.001	21.3	35 of 164	0.093	12.8	21 of 164	0.020
Exceeding (18)	18	48	48		38.9	7 of 18		33.3	6 of 18	
Present criteria[d]										
Meeting (155)	2	7	8	<0.001	19.4	30 of 155	0.004	11.6	18 of 155	0.003
Exceeding (27)	30	48	53		44.4	12 of 27		33.1	9 of 27	

UCSF University of California, San Francisco

[a]Kyoto criteria ; tumor number ≤ 10 nodules, all ≤5 cm in diameter, and DCP ≤400 mAU/ml, one patient without DCP result was excluded

[b]Kyushu criteria ; tumor size <5 cm or DCP <300 mAU/mL

[c]Modified Tokyo criteria ; fulfilling 2 or 3 among the following factors such as ≤5 nodules all ≤5 cm, AFP ≤250 ng/ml, and DCP ≤450 mAu/ml

[d]Present criteria; meeting MC, or beyond MC but with AFP <115 ng/ml and PET−

who maintained excellent outcome. In contrast, while the patients meeting the present new criteria included a smaller number of patients beyond MC (22 patients, 45%), the 27 patients exceeding the present new criteria had a significant risk of recurrence as high as 53% at 5 years. In addition, only the present new criteria were significantly associated with both microvascular invasion and tumor grade.

Microvascular invasion as well as tumor grade on explant pathology have been recognized as the gold standard for prediction of poor prognosis after LT [3, 4, 14]. Indeed, the present study confirmed that patients with microvascular invasion and those with poorly differentiated grade showed significantly higher recurrence rates, compared to patients without these pathological findings. However, it is generally thought that the presence of these pathological features cannot be ascertained before LT, even with preoperative needle biopsy. Hence, an international consensus conference on liver transplantation for HCC [32] recommended that indication for liver transplantation in HCC should not rely on these pathological features. Instead, several studies have investigated preoperative markers or criteria that are closely associated with these pathological features [17, 23]. In the present study, although AFP or PET status alone did not significantly correlate with microvascular invasion or tumor grade (data not shown), the patients exceeding the present new criteria showed a significantly higher prevalence of microvascular invasion and poorly differentiated tumors. This result also suggests that the present new criteria incorporating both AFP and PET status can reliably predict the patients at higher risk of post-transplant recurrence.

In the previous studies reporting the usefulness of pretransplant FDG-PET [17–21], PET-positive status was simply defined as standardized uptake value (SUV) of the HCC tumor being higher than that of the surrounding non-tumor parenchyma. On the other hand, Seo et al. [15] reported that the tumor/non-tumor ratio of SUV (TNR) increased stepwise according to tumor differentiation, and that there were significant differences in TNR between

well and moderately differentiated tumors and also between moderately and poorly differentiated tumors. In the initial design of the present study, we intended to collect the SUV data for both tumor and non-tumor tissues and evaluate the relationship between TNR and post-transplant HCC recurrence. However, because the survey was retrospective, the SUV data for both tumor and non-tumor tissues were obtained only for 60 patients (33%). Therefore, the PET status by semi-quantitative analysis was adopted as a variable also in the present study. Another limitation related to the retrospective design of the present study may be indicated as follows. The PET images were interpreted by the radiologist at each center and the collected reports were described in different formats. Since, in most centers, pre-transplant PET evaluation was not performed routinely for every recipient with HCC, the present study might have contained patient selection bias. In order to evaluate the significance of PET evaluation more precisely, any correlation between TNR and risk of recurrence should be investigated in further studies with a prospective design.

In conclusion, the results of the present study suggest that pre-transplant PET status offers additional information on the risk of HCC recurrence after LT and may be used for making decisions regarding transplantation for patients with HCC. Over-MC patients with negative PET status and lower AFP level may achieve successful outcome comparable to that of within-MC patients.

Conflict of interest None declared.

Appendix: author's affiliations

Yasutsugu Takada and Eitaro Ito, Department of HBP and Breast Surgery, Ehime University Graduate School of Medicine, Ehime, Japantakaday@m.ehime-u.ac.jpToshimi Kaido, Hideaki Okajima and Shinji Uemoto, Division of HBP Surgery and Transplantation, Department of Surgery, Kyoto University Graduate School of Medicine, Kyoto, Japan; Ken Shirabe, Department of Hepatobiliary and Pancreatic Surgery, Gunma University, Gunma, Japan; Hiroaki Nagano, Department of Gastroenterological Surgery, Yamaguchi University Graduate School of Medicine, Yamaguchi, Japan; Hiroto Egawa, Department of Surgery, Tokyo Women's Medical University, Tokyo, Japan; Yasuhiko Sugawara, Masaki Honda and Yukihiro Inomata, Department of Transplantation and Pediatric Surgery, Postgraduate School of Life Science, Kumamoto University, Kumamoto, Japan; Akinobu Taketomi, Department of Gastroenterological Surgery I, Hokkaido University Graduate School of Medicine, Hokkaido, Japan; Takeshi Takahara, Department of Surgery, Iwate Medical University School of Medicine, Iwate, Japan; Go Wakabayashi, Center for Advanced Treatment of Hepatobiliary and Pancreatic Diseases, Surgical Services, Ageo Central General Hospital, Saitama, Japan; Chikashi Nakanishi and Naoki Kawagishi, Department of Transplantation, Reconstruction and Endoscopic Surgery, Tohoku University Hospital, Miyagi, Japan; Akira Kenjo and Mitsukazu Gotoh, Department of Regenerative Surgery, Fukushima Medical University, Fukushima, Japan; Yoshikazu Toyoki and Kenichi Hakamada, Department of Gastroenterological Surgery, Hirosaki University Graduate School of Medicine, Aomori, Japan; Masayuki Ohtsuka, Department of General Surgery, Graduate School of Medicine, Chiba University, Chiba, Japan; Nobuhisa Akamatsu and Norihiro Kokudo, Artificial Organ and Transplantation Surgery Division, Department of Surgery,The University of Tokyo,Tokyo, Japan; Kazuhisa Takeda, National Hospital Organization Yokohama Medical Center, Kanagawa, Japan; Itaru Endo, Department of Gastroenterological Surgery, Yokohama City University Graduate School of Medicine, Kanagawa, Japan; Hiroyuki Takamura, Department of Gastroenterological Surgery, Kanazawa University, Ishikawa, Japan; Hiroshi Wada, Department of Gastroenterological Surgery, Graduate School of Medicine, Osaka University, Osaka, Japan; Shoji Kubo, Department of Hepato-Biliary-Pancreatic Surgery, Osaka City University Graduate School of Medicine, Osaka, Japan; Kaoru Kuramitsu and Yonson Ku, Hepato-Biliary-Pancreatic Surgery, Kobe University Hospital, Hyogo, Japan; Kohei Ishiyama and Hideki Ohdan, Department of Gastroenterological and Transplant Surgery, Applied Life Sciences, Institute of Biomedical & Health Sciences, Hiroshima University, Hiroshima, Japan; Yoshihiko Maehara, Department of Surgery and Science, Graduate School of Medical Sciences, Kyushu University, Fukuoka, Japan; Hiroyuki Furukawa, Division of Gastroenterologic Surgery, Department of Surgery, Asahikawa Medical University, Hokkaido, Japan; Hiroki Yamaue, Second Department of Surgery, Wakayama Medical University School of Medicine, Wakayama, Japan; Masaru Miyazaki, International University of Health and Welfare, Mita Hospital, Tokyo, Japan; Tadahiro Takada, Japanese Society of Hepato-biliary-Pancreatic Surgery, Department of Surgery, Teikyo University School of Medicine, Tokyo, Japan.

References

1. Mazzaferro V, Regalia E, Doci R, Andreola S, Pulvirenti A, Bozzetti F, et al. Liver transplantation for the treatment of small hepatocellular carcinomas in patients with cirrhosis. N Engl J Med. 1996;334:693–9.
2. Ito T, Takada Y, Ueda M, Haga H, Maetani Y, Oike F, et al. Expansion of selection criteria for patients with hepatocellular carcinoma

in living donor liver transplantation. Liver Transpl. 2007;13:1637–44.
3. Sugawara Y, Tamura S, Makuuchi M. Living donor liver transplantation for hepatocellular carcinoma: Tokyo University series. Dig Dis. 2007;25:310–2.
4. Duffy JP, Vardanian A, Benjamin E, Watson M, Farmer DG, Ghobrial RM, et al. Liver transplantation criteria for hepatocellular carcinoma should be expanded: a 22-year experience with 467 patients at UCLA. Ann Surg. 2007;246:502–11.
5. Yao FY, Xiao L, Bass NM, Kerlan R, Ascher NL, Roberts JP. Liver transplantation for hepatocellular carcinoma: validation of the UCSF-expanded criteria based on preoperative imaging. Am J Transplant. 2007;7:2587–96.
6. Soejima Y, Taketomi A, Yoshizumi T, Uchiyama H, Aishima S, Terashi T, et al. Extended indication for living donor liver transplantation in patients with hepatocellular carcinoma. Transplantation. 2007;83:893–9.
7. Guiteau JJ, Cotton RT, Washburn WK, Harper A, O'Mahony CA, Sebastian A, et al. An early regional experience with expansion of Milan criteria for liver transplant recipients. Am J Transplant. 2010;10:2092–8.
8. Ng KK, Lo CM, Chan SC, Chok KS, Cheung TT, Fan ST. Liver transplantation for hepatocellular carcinoma: the Hong Kong experience. J Hepatobiliary Pancreat Sci. 2010;17:548–54.
9. Takada Y, Uemoto S. Liver transplantation for hepatocellular carcinoma: the Kyoto experience. J Hepatobiliary Pancreat Sci. 2010;17:527–32.
10. Mazzaferro V, Chun YS, Poon RTP, Schwartz ME, Yao FY, Marsch JW, et al. Liver transplantation for hepatocellular carcinoma. Ann Surg Oncol. 2008;15:1001–7.
11. Takada Y, Tohyama T, Watanabe J. Biological markers of hepatocellular carcinoma for use as selection criteria in liver transplantation. J Hepatobiliary Pancreat Sci. 2015;22:279–86.
12. Hustinx R, Benard F, Alavi A. Whole-body FDG-PET imaging in the management of patients with cancer. Semin Nucl Med. 2002;32:35–46.
13. Khan MA, Combs CS, Brunt EM, Lowe VJ, Wolverson MK, Soloman H, et al. Positron emission tomography scanning in the evaluation of hepatocellular carcinoma. J Hepatol. 2000;32:792–7.
14. Hatano E, Ikai I, Higashi T, Teramukai S, Torizuka T, Saga T, et al. Preoperative positron emission tomography with fluorine-18-fluorodeoxyglucose is predictive of prognosis in patients with hepatocellular carcinoma after resection. World J Surg. 2006;30:1736–41.
15. Seo S, Hatano E, Higashi T, Hara T, Tada M, Tamaki N, et al. Fluorine-18 fluorodeoxyglucose positron emission tomography predicts tumor differentiation, p-glycoprotein expression, and outcome after resection in hepatocellular carcinoma. Clin Cancer Res. 2007;13:427–33.
16. Shiomi S, Nishiguchi S, Ishizu H, Iwata Y, Sasaki N, Tamori A, et al. Usefulness of positron emission tomography with fluorine-18-fluorodeoxyglucose for predicting outcome in patients with hepatocellular carcinoma. Am J Gastroenterol. 2001;96:1877–80.
17. Kornberg A, Freesmeyer M, Barthel E, Jandt K, Katenkamp K, Steenbeck J, et al. ^{18}F-FDG-uptake of hepatocellular carcinoma on PET predicts microvascular tumor invasion in liver transplant patients. Am J Transplant. 2009;9:592–600.
18. Lee SG, Ahn CS, Ha TY, Moon DB, Choi KM, Song GW, et al. Liver transplantation for hepatocellular carcinoma: Korean experience. J Hepatobiliary Pancreat Sci. 2010;17:539–47.
19. Cheung TT, Chan SC, Ho CL, Chok KSH, Chan ACY, Sharr WW, et al. Can positron emission tomography with the dual tracers [^{11}C] acetate and [^{18}F] fludeoxyglucose predict microvascular invasion in hepatocellular carcinoma? Liver Transpl. 2011;17:1218–25.
20. Kornberg A, Kupper B, Tannapfel A, Buchler P, Krause B, Witt U, et al. Patients with non-[^{18}F] fludeoxyglucose-avid advanced hepatocellular carcinoma on clinical staging may achieve long-term recurrence-free survival after liver transplantation. Liver Transpl. 2012;18:53–61.
21. Lee SD, Kim SH, Kim YK, Kim C, Kim SK, Han SS, et al. ^{18}F-FDG-PET/CT predicts early tumor recurrence in living donor liver transplantation for hepatocellular carcinoma. Transplant Int. 2013;26:50–60.
22. Yao FY, Bass NM, Watson JJ, Bacchetti P, Venook A, Ascher NL, et al. Liver transplantation for hepatocellular carcinoma: expansion of the tumor size limits does not adversely impact survival. Hepatology. 2001;33:1394–1403.
23. Mazzaferro V, Llovet JM, Miceli R, Bhoori S, Schiavo M, Mariani L, et al. Predicting survival after liver transplantation in patients with hepatocellular carcinoma beyond the Milan criteria: a retrospective, exploratory analysis. Lancet Oncol. 2009;10:35–43.
24. Fujiki M, Takada Y, Ogura Y, Oike F, Kaido T, Teramukai S, et al. Significance of des-gamma-carboxy prothrombin in selection criteria for living donor liver transplantation for hepatocellular carcinoma. Am J Transplant. 2009;9:2362–71.
25. Taketomi A, Sanefuji K, Soejima Y, Yoshizumi T, Uchiyama H, Ikegami T, et al. Impact of des-gamma-carboxy prothrombin and tumor size on the recurrence of hepatocellular carcinoma after living donor liver transplantation. Transplantation. 2009;87:531–7.
26. Shindoh J, Sugawara Y, Nagata R, Kaneko J, Tamura S, Aoki T, et al. Evaluation methods for pretransplant oncologic markers and their prognostic impacts in patient undergoing living donor liver transplantation for hepatocellular carcinoma. Transplant Int. 2014;27:391–8.
27. Yang SH, Suh KS, Lee HW, Cho EH, Cho JY, Cho YB, et al. A revised scoring system utilizing serum alphafetoprotein levels to expand candidates for living donor liver transplantation in hepatocellular carcinoma. Surgery. 2007;141:598–609.
28. Zheng SS, Xu X, Wu J, Chen J, Wang WL, Zhang M, et al. Liver transplantation for hepatocellular carcinoma: Hangzhou experiences. Transplantation. 2008;85:1726–32.
29. Toso C, Asthana S, Bigam DL, Shapiro AMJ, Kneteman NM. Reassessing selection criteria prior to liver transplantation for hepatocellular carcinoma utilizing the scientific registry of transplant recipients database. Hepatology. 2009;49:832–8.
30. Duvoux C, Roudot-Thoraval F, Decaens T, Pessione F, Badran H, Piardi T, et al. Liver transplantation for hepatocellular carcinoma: a model including α-fetoprotein improves the performance of Milan criteria. Gastroenterology. 2012;143:986–94.
31. Furukawa H, Shimamura T, Suzuki T, Taniguchi M, Nakanishi K, Yamashita K, et al. Liver transplantation for hepatocellular carcinoma: the Japanese experience. J Hepatobiliary Pancreat Sci. 2010;17:533–8.
32. Clavien PA, Lesurtel M, Bossuyt PMM, Gore GJ, Langer B, Perrer A, et al. Recommendations for liver transplantation for hepatocellular carcinoma: an international consensus conference report. Lancet Oncol. 2012;13:e11–22.