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Preoperative intramuscular adipose tissue content (IMAC) is a novel prognostic predictor after hepatectomy for hepatocellular carcinoma

Yuhei Hamaguchi, Toshimi Kaido, Shinya Okumura, Takashi Ito, Yasuhiro Fujimoto, Kohei Ogawa, Akira Mori, Ahmed Hammad, Etsuro Hatano, Shinji Uemoto

Division of Hepato-Biliary-Pancreatic and Transplant Surgery, Department of Surgery, Graduate School of Medicine, Kyoto University, 54 Kawahara-cho, Shogoin, Sakyo-ku, Kyoto 606-8507, Japan

Corresponding author:

Toshimi Kaido, MD, PhD

Address: Division of Hepato-Biliary-Pancreatic and Transplant Surgery, Department of Surgery, Graduate School of Medicine, Kyoto University, 54 Kawahara-cho, Shogoin, Sakyo-ku, Kyoto 606-8507, Japan.

Phone: +81-75-751-3242 Fax: +81-75-751-4263

E-mail: kaido@kuhp.kyoto-u.ac.jp

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Abstract

Background Sarcopenia has been shown to be an independent predictor of lower disease-free and overall survival in various kinds of diseases. The quality of skeletal muscle has recently attracted much attention as a new parameter of sarcopenia.

Methods We performed a retrospective analysis of 477 patients undergoing hepatectomy for HCC between April 2005 and August 2014. The quality of skeletal muscle was evaluated by intramuscular adipose tissue content (IMAC) using preoperative CT imaging. The impact of IMAC on outcomes after hepatectomy for HCC was analyzed.

Results Patients with high IMAC showed older age, higher body mass index, higher indocyanine green retention test at 15 min, and more operative blood loss. The overall and recurrence-free survival rates were significantly lower in patients with high IMAC than in patients with normal IMAC ($P < 0.0001$, $P = 0.0012$, respectively). Multivariate analysis showed that high IMAC was the significant risk factor for death (hazard ratio [HR] = 2.942; $P < 0.0001$) and for HCC recurrence (HR = 1.529; $P = 0.0007$) after hepatectomy.

Conclusions Preoperative quality of skeletal muscle was closely correlated with postoperative mortality and HCC recurrence. IMAC could be incorporated into new selection criteria for hepatectomy for HCC.

Introduction

Sarcopenia is defined as a syndrome characterized by progressive and generalized loss of skeletal muscle mass and strength [1]. The prognostic significance of sarcopenia has been reported in various kinds of diseases including cancer [2–4]. Most previous studies have investigated only skeletal muscle mass to define sarcopenia because muscle strength and function are difficult to evaluate, though The European Working Group on Sarcopenia in Older People has recommended that the definition of sarcopenia should include not only low muscle mass, but also low muscle strength or function [1]. Recently, it has been reported that the increase in intramuscular adipose tissue (IMAT) with aging was associated with muscle weakness and poor function, one of the components of sarcopenia [5]. Kitajima et al [6]. showed that the quality of skeletal muscle could be evaluated by measuring intramuscular adipose tissue content (IMAC), and that IMAC was linked to the severity of non-alcoholic steatohepatitis (NASH). In our previous study, preoperative lower quality of skeletal muscle evaluated by IMAC was an independent and significant risk factor for poor survival in patients undergoing living donor liver transplantation [7]. Thus, the quality of skeletal muscle has recently attracted much attention as a new parameter of sarcopenia.

Hepatocellular carcinoma (HCC) is one of the most common human cancers, and its incidence is still increasing worldwide [8, 9]. HCC is a highly malignant tumor characterized by rapid progression, frequent tumor recurrence, and metastasis. To date, there has been no

successful systemic chemotherapy for patients with advanced HCC, and surgical treatment, including hepatectomy and liver transplantation, is the most effective modality for the treatment for HCC [9]. While advances in preoperative diagnosis, surgical technique, and perioperative management have improved overall and recurrence-free survival after partial hepatectomy for HCC, even in patients with curative resection, the postoperative recurrence rate remains high, with a five-year recurrence rate of up to 70% [10].

Recent study has shown that sarcopenia defined as lower skeletal muscle mass was an independent prognostic factor for mortality after hepatectomy for HCC [11, 12]. However, measuring only the area of skeletal muscle using CT imaging may not be sufficient to evaluate lean muscle mass because we could not distinguish muscle from adipose tissue with that method, which might mislead us to evaluate low lean muscle mass with much adipose tissue as normal skeletal muscle mass.

In the present study, we evaluated the quality of skeletal muscle by measuring IMAC as a new parameter of sarcopenia, and investigated the impact of sarcopenia on outcomes in patients undergoing hepatectomy for HCC.

Patients and methods

Patients

There were 547 patients who underwent hepatectomy for HCC at Kyoto University Hospital

between April 2005 and August 2014. Seventy patients who did not have preoperative plain computed tomography (CT) imaging at the umbilical level were excluded from this study. Therefore, a total of 477 patients (389 men, 88 women) were enrolled in the present study. The study was approved by the Ethics Committee of Kyoto University and conducted in accordance with the Declaration of Helsinki of 1996.

Hepatic resection was usually performed using the Cavitron Ultrasonic Surgical Aspirator (CUSA) and a bipolar cautery device equipped with a channel for water dripping, with an intermittent Pringle maneuver, or selective vascular clamping if necessary. A pathological examination was performed to evaluate tumor size, tumor number, vascular invasion, tumor differentiation, and background liver in accordance with the rules of the Liver cancer Study Group of Japan.

All patients were followed up every three months after operation with evaluation of tumor markers and CT or magnetic resonance imaging.

Imaging analysis

All CT imaging was obtained with a multidetector computed tomography scanner (Aquilion 64, Toshiba Medical Systems, Tochigi, Japan). The technical parameters used for CT were as follows: 120 kV (tube voltage), 0.5 mm × 64 row (detector configuration), tube current modulation, 0.5 sec/rotation (gantry rotation), and 7 mm reconstruction thickness.

The quality of skeletal muscle was evaluated by IMAC. IMAC was calculated as previously described:

IMAC = region of interest (ROI) of the multifidus muscle (Hounsfield units)/ROI of subcutaneous fat (Hounsfield units) [6, 7].

Subfascial muscular tissue in the multifidus muscle on the preoperative plain CT cross-sectional image at the umbilical level was precisely traced, and CT values (in Hounsfield units) were measured using the AquariusNET server (TeraRecon, Inc., San Mateo, CA, USA) (Fig. 1a). CT values were measured for ROIs of four circles on subcutaneous fat away from major vessels (Fig. 1b). The mean values of those four ROIs were used as the ROI of subcutaneous fat. In general, IMAC was shown with a negative value. Skeletal muscle with more IMAT shows higher IMAC, so higher IMAC means lower quality of skeletal muscle.

The quantity of skeletal muscle was evaluated by psoas muscle mass index (PMI). The cross-sectional areas of the bilateral psoas muscles were measured by manual tracing using preoperative CT images at the same level (Fig. 1c). The psoas muscle mass index (PMI) was calculated by normalizing the cross-sectional areas for height (cm^2/m^2).

Receiver operating characteristic (ROC) curves were calculated in order to select the optimal cut-off values of IMAC and PMI for men and women separately. The cut-off values were selected on the basis of best accuracy in relation to an outcome (death). The cut-off values of IMAC in males and females were -0.324 (area under the curve [AUC] = 0.687) and -0.138

(AUC = 0.651), respectively. The cut-off values of PMI were 6.089 (AUC = 0.548) and 4.020 (AUC = 0.523) cm^2/m^2, respectively.

Analyzed parameters

The overall and recurrence-free survival rates after hepatectomy were investigated in patients classified according to IMAC or PMI. The prognostic factors were analyzed on the basis of the following variables: patient age, sex, etiology of HCC, the presence of treatment history for HCC before hepatectomy, platelet count, indocyanine green retention test at 15 min (ICG R15), Child-Pugh classification, serum α-fetoprotein (AFP) level, des-γ-carboxyprothrombin (DCP) level, liver histology, tumor size, number of tumors, microvascular invasion (MVI), tumor differentiation, pathological stage according to the Liver Cancer Study Group of Japan, surgical procedure, duration of surgery, estimated operative blood loss, body mass index (BMI), preoperative IMAC, and preoperative PMI.

Statistical analysis

Continuous variables were nonparametrically analyzed using the Mann-Whitney U test. Categorical variables were compared using the χ^2 test or Fisher's exact test where appropriate. Correlation between continuous variables was assessed using Pearson correlation coefficients and linear regression. Cumulative overall and recurrence-free survival rates were calculated

using Kaplan-Meier methods, and differences between curves were evaluated using the log-rank test. Any variable identified as significant ($P < 0.05$) or with $P < 0.10$ in univariate analysis was considered a candidate for a multivariate Cox regression analysis. A P value < 0.05 was considered significant. All statistical data were generated using JMP 11 (SAS Institute, Cary, NC, USA) and Prism 6 (GraphPad Software, Inc., La Jolla, CA, USA).

Results

Patient characteristics

The baseline characteristics and laboratory data of the 477 patients are shown in Table 1. The median patient age was 68 years (range, 31–89 years). The Child-Pugh classifications were A and B for 437 (92%) and 40 patients (8%), respectively. Two hundreds and eighty-nine patients (61%) underwent primary hepatectomy as the initial treatment for HCC, while 189 patients (39%) had treatment history before hepatectomy such as TACE or RFA. The median tumor size was 3.5 cm (range, 0.2–25 cm). Pathological stages according to the Liver Cancer Study Group of Japan were I, II , III, and IV for 82 (17%), 181 (38%), 142 (30%), and 72 patients (15%), respectively. Microvascular invasion was identified in 152 patients (32%).

A significant positive relationship was observed between IMAC and patient age in both males ($r = 0.368$, $P < 0.0001$) and females ($r = 0.535$, $P < 0.0001$); therefore, the mean age in patients with high IMAC was significantly higher than in patients with normal IMAC ($P <$

0.0001). Similarly, BMI in patients with high IMAC was significantly higher than in patients with normal BMI ($P = 0.0002$). The patients with normal IMAC showed more hepatitis B virus (HBV) or hepatitis C virus (HCV)-related HCC than those with high IMAC ($P = 0.0350$). Among preoperative laboratory data, only ICG R15 was significantly higher in patients with high IMAC than in patients with normal IMAC ($P = 0.0485$). Tumor-associated factors were not significantly different between the two groups. Among operative factors, estimated blood loss in patients with high IMAC was significantly higher than in patients with normal IMAC ($P = 0.0225$).

Overall and recurrence-free survival rates after hepatectomy

The overall survival rate after hepatectomy for HCC was significantly lower in patients with high IMAC than in patients with normal IMAC ($P < 0.0001$; Fig. 2a). In patients classified according to PMI, the overall survival rate was not significantly different between the two groups ($P = 0.3178$; Fig. 2b). The recurrence-free survival rate was significantly lower in patients with high IMAC than in patients with normal IMAC ($P = 0.0012$; Fig. 2c). The recurrence-free survival rate in patients with low PMI was not significantly different from in patients with normal PMI ($P = 0.7468$; Fig. 2d).

Risk factors for poor outcomes in patients undergoing hepatectomy

The results of univariate and multivariate analysis of the overall survival in patients undergoing hepatectomy for HCC are shown in Table 2. A multivariate Cox regression analysis identified high serum AFP level (≥20 ng/dl), advanced TNM stage, preoperative high IMAC as independent risk factors for death after hepatectomy (Table 2).

Table 3 shows the results of univariate and multivariate analysis of the recurrence-free survival. Multivariate analysis showed that liver histology (liver fibrosis or liver cirrhosis), larger tumor size (≥5.0 cm), MVI, advanced TNM stage, and preoperative high IMAC were independent risk factors for HCC recurrence (Table 3).

Discussion

This retrospective study was the largest and first study to show that preoperative IMAC was the most significant prognostic factor for overall and recurrence-free survival in patients following hepatectomy for HCC. Recently, sarcopenia has attracted much attention as a prognostic factor for poor outcome in various kinds of diseases [2–4, 11, 12]. Although most recent studies have investigated only skeletal muscle mass to evaluate sarcopenia using CT imaging, measuring only the area of skeletal muscle using CT imaging may not be sufficient to evaluate lean muscle mass because we could not distinguish muscle from adipose tissue with that method. In other words, PMI does not always reflect actual muscle mass because the areas of psoas muscles measured in the present study include both muscle and IMAT. In some

patients with a large amount of IMAT, their PMI could be calculated as normal even if their actual amount of muscle mass was low. Therefore, only measuring PMI could not detect such patients as having sarcopenia. On the other hand, high IMAC reflects not only more IMAT but also less muscle mass because IMAC is calculated as the ratio of CT values between skeletal muscle and subcutaneous fat tissue. In addition, a recent report from the Health, Aging and Body Composition (Health ABC) Study Cohort showed that the loss of strength was surprisingly much greater than the loss of muscle mass, which indicated that decline in muscle quality contributed to loss of strength [13]. It has been also shown that fat accumulation within skeletal muscle is associated with muscle weakness, poor function, and increased risk of incidental mobility limitations because it alters muscle fiber orientation and the force-producing capabilities of the whole muscle [14–16]. On the basis of these findings, we considered that IMAC could substitute for evaluation of muscle strength or function and could be the most appropriate parameter to evaluate preoperative sarcopenia instead of measuring only the cross-sectional areas of skeletal muscle.

The mechanisms by which sarcopenia produces an increased risk of mortality and morbidity are not fully understood. In the present study, patient age, BMI, preoperative ICG R15 and operative blood loss were significantly different between high IMAC group and normal IMAC group. Operative blood loss has been reported as independent prognostic factors for death or tumor recurrence after hepatectomy for HCC [17]. Among these parameters, only

operative blood loss was shown to be a significant risk factor for death or HCC recurrence after hepatectomy as a result of univariate analysis, while it was not an independent risk factor. Recent evidence has shown that skeletal muscle loss with increasing adipose tissue leads to synthesize and secrete various kinds of pro-inflammatory adipokines such as leptin, TNF-α, interleukin (IL)-1 and IL-6 [18]. Lutz et al. [19] showed that increase of such adipokines and decrease in adiponectin or IL-15 levels in aged or sarcopenic population had effects on the immune senescence, especially natural killer lymphocytes in innate immunity. Indeed, in the present study, we found more infectious postoperative complications in patients with high IMAC than in patients with normal IMAC (22.0% vs 9.7%; $P = 0.0002$). This result is in line with previous study that showed the presence of sarcopenia increases the risk of sepsis-related death in cirrhotic patients, which might be due to impaired immunity [20]. Recently, with increased interest towards subsets of obesity-associated tumors, the role of adipose and muscle tissue interplay in carcinogenesis and cancer progression has attracted much attention [21, 22]. It has been shown that among various kinds of adipokines, anti-inflammatory cytokines such as adiponectin presents anti-tumorigenic effects *via* various mechanisms [21]. On the other hands, pro-inflammatory adipokines such as leptin has been shown to promote growth and proliferation of tumor cells *via* activation various growth and survival signaling pathways [21]. In HCC, Sharma D et al. [23] reported that adiponectin has the molecular potential to inhibit the oncogenic actions of leptin by blocking Stat 3 and Akt phosphorylation.

On the basis of these findings, we speculate that the increase in adiposity in sarcopenic patients could lead to imbalance between adipokines and myokines, which results in cancer growth and recurrence.

To improve sarcopenic status, beneficial effects can be obtained through exercise and amino acid supplementation using leucine-enriched mixture, by their regulation of mTOR signaling [24]. Today, BCAA nutrients, synbiotics and immune-modulating diet enriched with hydrolyzed whey peptide are used for all recipients as pre- and postoperative enteral nutrition in our institution. In our previous study, we found that perioperative nutritional therapy significantly improved posttransplant mortality in patients with low skeletal muscle mass measured by bioelectrical impedance analysis [25]. On the basis of these results, we are planning to prospectively evaluate the effect of preoperative nutritional therapy for improvement of IMAC, and investigate the impact of changes in IMAC on postoperative outcomes after hepatectomy for HCC.

The present study has several limitations. First, there was a possibility of selection bias for patient inclusion in the study group because as many cases were excluded from this study. We excluded 70 patients (12.8%) for the sole reason of their having undergone no CT imaging at the umbilical level. This exclusion criterion was not related to the patients' general condition and severity of disease. Indeed, the mean values of preoperative blood examinations, the distribution of the Child-Pugh classification scores, etiology of HCC, operative factors, and

tumor factors in this excluded population were almost similar to those of the study group. Therefore, there was actually little selection bias for patient inclusion in the present study. Second, we have to consider whether our cut-off values for IMAC and PMI were adequate to define sarcopenia. To date, several reports have each provided a definition of sarcopenia, but there is no criterion to define sarcopenia objectively [26, 27]. In the present study, we used ROC curves to determine the cut-off values of IMAC and PMI. The use of ROC curves is a more accurate and objective method than the use of SD for the design of cut-off values. We are now investigating IMAC and PMI considering age, sex, and BMI in healthy donors of liver transplantation; this investigation may determine more adequate cut-off values of IMAC and PMI to define sarcopenia.

In conclusion, the present study showed that the low quality of skeletal muscle evaluated by measuring IMAC was closely correlated with poor outcomes in patients undergoing hepatectomy for HCC. Preoperative IMAC could be incorporated into new selection criteria for hepatectomy for HCC.

Conflict of interest: None declared.

Author contributions: Study design: Hamaguchi, Kaido and Uemoto. Acquisition of data: Hamaguchi, Kaido, Okumura, Ito, Fujimoto, Ogawa, Mori, Hammad, Hatano. Analysis

and interpretation: Hamaguchi and Kaido. Manuscript drafted by: Hamaguchi Revision:

Kaido and Uemoto

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Figure legends

Fig. 1 Cross-sectional computed tomographic images of (**a**) subfascial muscular tissue in the multifidus muscle, (**b**) subcutaneous fat (four small circles) and (**c**) bilateral psoas muscles at the umbilical level.

Fig. 2 (**a**) Overall survival rates after hepatectomy in patients classified according to IMAC. The overall survival rate was significantly lower in patients with high IMAC than in patients with normal IMAC ($P < 0.0001$). (**b**) Overall survival rates after hepatectomy in patients classified according to PMI. The overall survival rate was not significantly different between the two groups ($P = 0.3178$). (**c**) Recurrence-free survival rates after hepatectomy in patients classified according to IMAC. The recurrence-free survival rate was significantly lower in patients with high IMAC than in patients with normal IMAC ($P = 0.0012$). (**d**) Recurrence-free survival rates after hepatectomy in patients classified according to PMI. The recurrence-free survival rate in patients with low PMI was not significantly different from in patients with normal PMI ($P = 0.7468$).

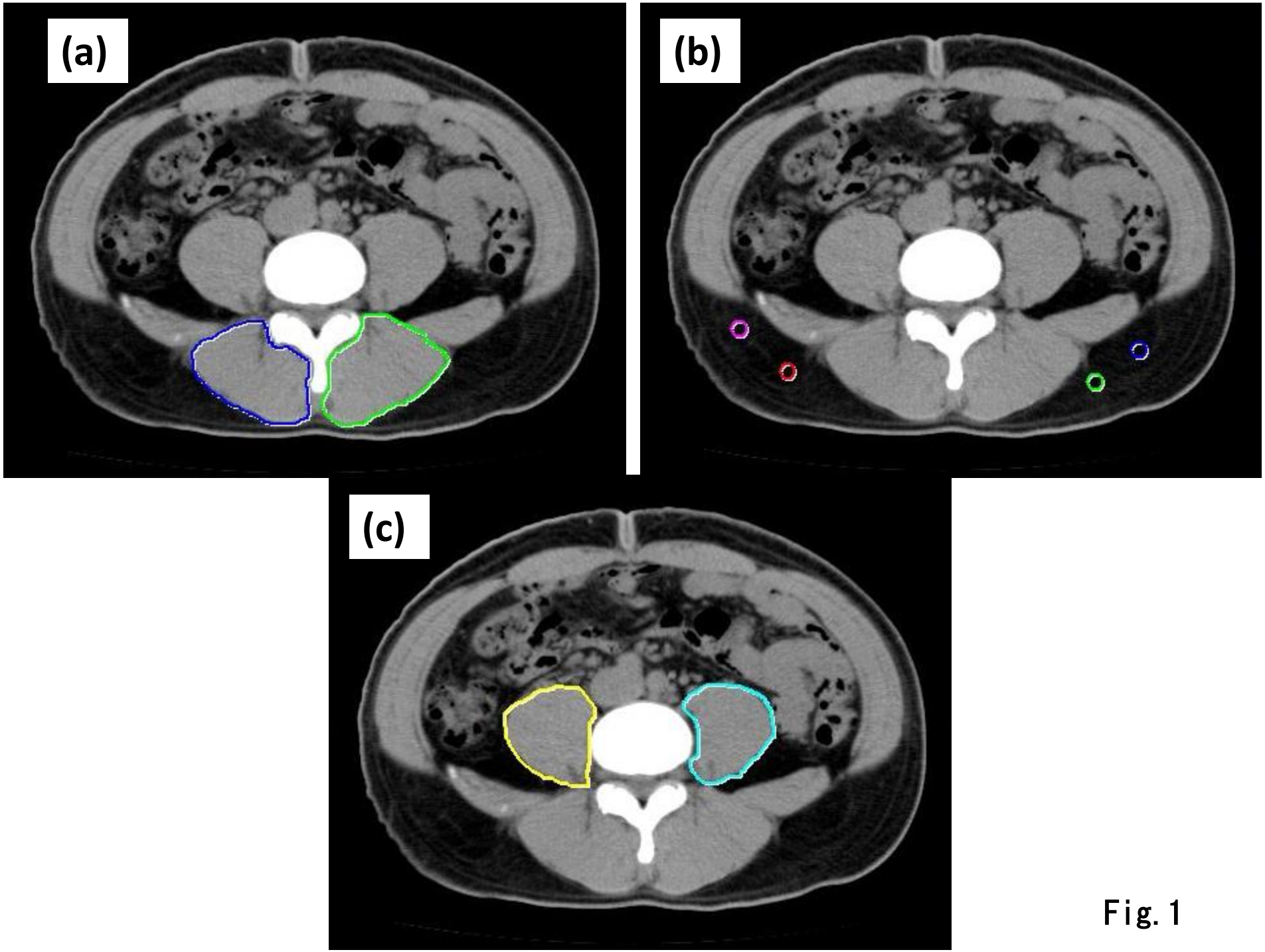
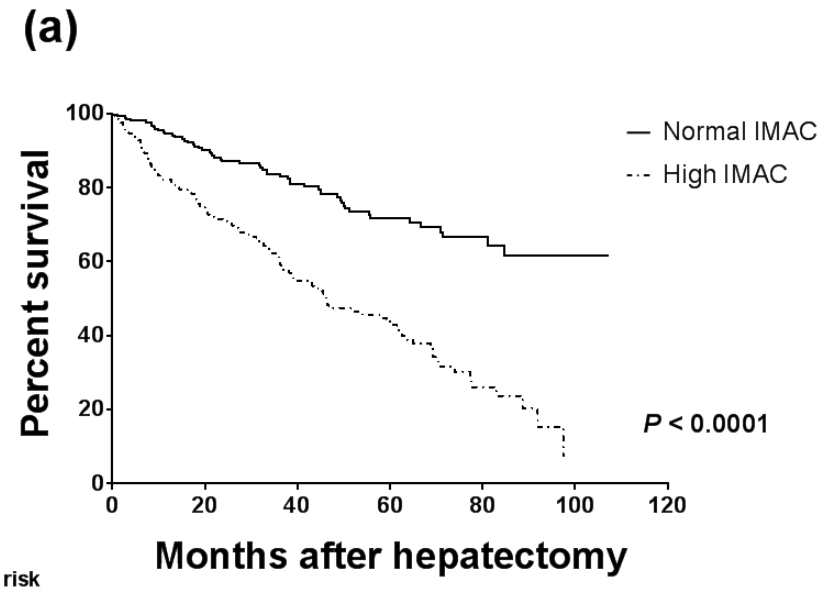
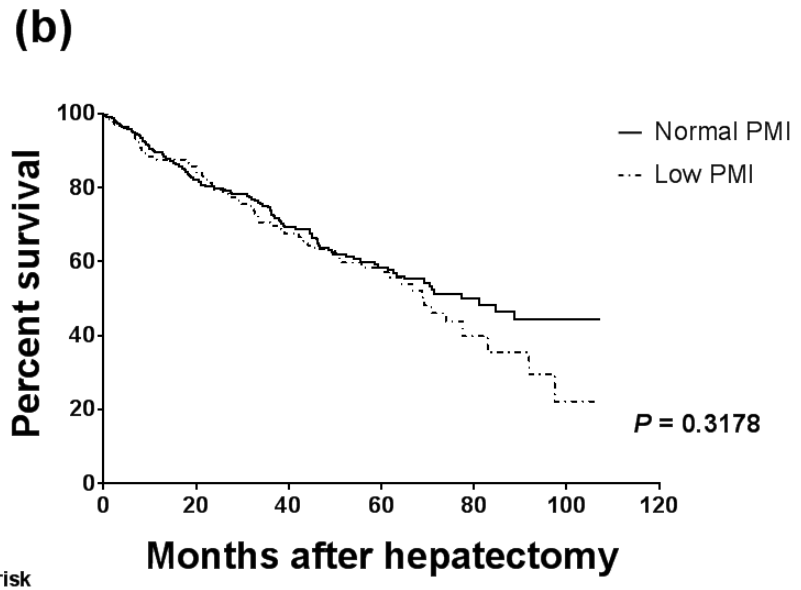
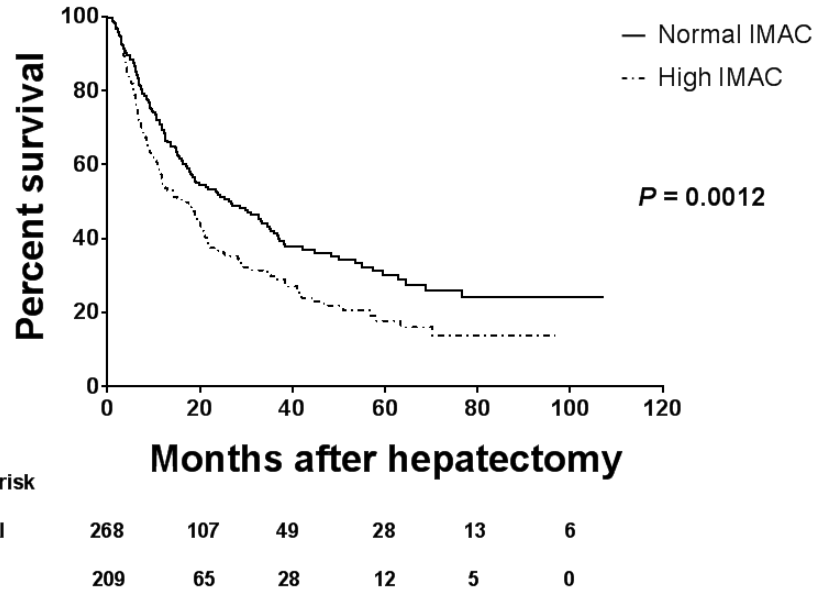


Fig. 1


(a)
Percent survival
Months after hepatectomy
Normal IMAC
High IMAC
P < 0.0001
No. at risk
Normal 268 181 121 78 30 12
High 209 131 80 47 14 0


(b)
Percent survival
Months after hepatectomy
Normal PMI
Low PMI
P = 0.3178
No. at risk
Normal 343 215 135 81 34 9
Low 134 97 66 44 11 4


(c)
Percent survival
Months after hepatectomy
Normal IMAC
High IMAC
P = 0.0012
No. at risk
Normal 268 107 49 28 13 6
High 209 65 28 12 5 0

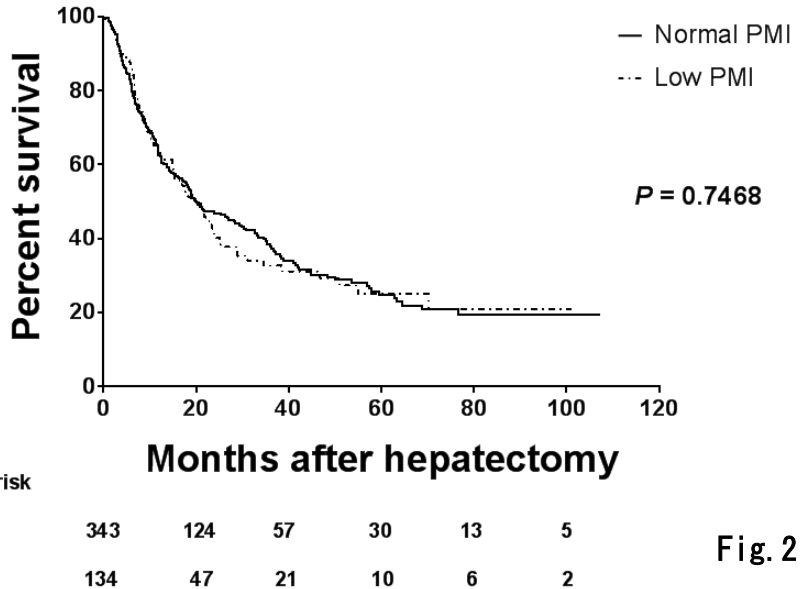

(d)
Percent survival
Months after hepatectomy
Normal PMI
Low PMI
P = 0.7468
No. at risk
Normal 343 124 57 30 13 5
Low 134 47 21 10 6 2

Fig. 2

Table 1 Characteristics of patients classified according to IMAC

Characteristic	Total (n = 477)	High IMAC (n = 209)	Normal IMAC (n = 268)	*P*
Age (years)				
Mean (SD)	67 (10)	71 (7)	64 (10)	<0.0001
Sex, n (%)				
Men	389 (82)	171 (82)	218 (81)	0.8944
Women	88 (18)	38 (18)	50 (19)	
BMI (kg/m^2)				
Mean (SD)	23.1 (3.4)	23.9 (3.6)	22.6 (3.1)	0.0002
Preoperative IMAC				
Males				
Mean (SD)	-0.339 (0.126)	-0.232 (0.091)	-0.422 (0.077)	<0.0001
Females				
Mean (SD)	-0.172 (0.170)	-0.020 (0.120)	-0.288 (0.092)	<0.0001
Preoperative PMI, n (%)				
Normal	343 (72)	145 (69)	198 (74)	0.2784
Low	134 (28)	64 (31)	70 (26)	
Etiology, n (%)				
HBV or/and HCV	319 (67)	129 (62)	190 (71)	0.0350
Others	158 (33)	80 (38)	78 (29)	
The presence of previous treatment for HCC, n (%)				
Yes	188 (39)	85 (41)	103 (38)	0.6200
No	289 (61)	124 (59)	165 (62)	
Platelet count ($\times 10^4/mm^3$)				
Mean (SD)	15.4 (7.0)	15.3(7.2)	15.4 (6.9)	0.9642
ICG R15 (%)				
Mean (SD)	17.0 (10.3)	18.0 (10.5)	16.3 (10.1)	0.0485
Child-Pugh classification, n (%)				
A	437 (92)	186 (89)	251 (94)	0.0697
B	40 (8)	23 (11)	17 (6)	
AFP (ng/ml)				
Median	17.5	17.5	17.7	0.9610
(range)	(0.9-2873490)	(1.2-167928)	(0.9-2873490)	
DCP (munits/l)				
Median	137.5	125.5	161	0.6181
(range)	(6-431000)	(6-223000)	(8-431000)	
Liver histology, n (%)				
Normal liver + chronic hepatitis	239 (50)	109 (52)	130 (49)	0.4294
Liver fibrosis + liver cirrhosis	238 (50)	100 (48)	138 (51)	
Tumor size (cm)				
Mean (SD)	4.8 (3.8)	5.0 (3.9)	4.7 (3.8)	0.3326
Number of tumors, n (%)				
Solitary	324 (68)	149 (71)	175 (65)	0.1630
Multiple	153 (32)	60 (29)	93 (35)	
MVI, n (%)				

Table 2 Prognostic factors for overall survival on univariate and multivariate analysis

	Univariate analysis		Multivariate analysis		
	MST (months)	*P*	Hazard ratio	95% CI	*P*
Age (years)					
≥65 (n = 169)	69.3	0.1541			
<65 (n = 308)	83				
Gender					
Male (n = 389)	71.4	0.9746			
Female (n = 88)	63.4				
Etiology of HCC					
HBV or/and HCV (n =319)	73.9	0.9248			
Others (n = 158)	70.4				
The presence of previous treatment for HCC					
Yes (n = 188)	62.5	0.2235			
No (n = 289)	81.1				
Platelet count ($\times 10^4/mm^3$)					
<10 (n = 101)	54	0.0093	1.237	(0.853-1.764)	0.2575
≥10 (n = 376)	84.7		Reference		
ICG R15 (%)					
≥15 (n = 245)	69.3	0.2298			
<15 (n = 232)	97.5				
Child-Pugh classification					
A (n = 437)	73.9	0.1120			
B (n = 40)	45.6				
AFP (ng/dl)					
≥20 (n = 229)	55.7	<0.0001	1.552	(1.110-2.184)	0.0099
<20 (n = 248)	97.5		Reference		
DCP (munits/l)					
≥40 (n = 338)	73.9	0.1201			
<40 (n = 139)	71.4				
Liver histology					
Normal liver or chronic hepatitis (n = 239)	84.7	0.1895			
Liver fibrosis or liver cirrhosis (n = 238)	65				
Tumor size (cm)					
≥5.0 (n =170)	60.2	0.0008	1.248	(0.864-1.801)	0.2374
<5.0 (n = 307)	83		Reference		

Number of tumors					
Multiple (n = 153)	62.5	0.0104	1.045	(0.722-1.522)	0.8155
Solitary (n = 324)	97.5		Reference		
MVI					
Positive (n = 152)	41.4	<0.0001	1.218	(0.820-1.803)	0.3283
Negative (n = 325)	84.7		Reference		
Tumor differentiation					
Well or moderate (n = 349)	81.1	<0.0001	0.698	(0.490-1.003)	0.0522
Poor (n = 113)	45.6		Reference		
TNM stage					
I or II (n = 263)	.	<0.0001	0.477	(0.309-0.735)	0.0008
III or IV (n = 214)	45.6		Reference		
Surgical procedure					
≥Lobectomy (n = 167)	46.8	0.0004	1.201	(0.810-1.782)	0.3621
<Segmentectomy (n = 310)	81.1		Reference		
Operative time (min)					
≥360 (n = 250)	52.4	0.0003	1.335	(0.938-1.915)	0.1090
<360 (n = 227)	84.7		Reference		
Operative blood loss (ml)					
≥500 (n =310)	62.5	<0.0001	1.112	(0.711-1.776)	0.6456
<500 (n = 167)	.		Reference		
BMI (kg/m^2)					
≥25 (n = 109)	71.4	0.8510			
<25 (n = 368)	73.9				
Preoperative IMAC					
High (n = 209)	46.2	<0.0001	2.942	(2.117-4.136)	<0.0001
Normal (n = 268)	.		Reference		
Preoperative PMI					
Low (n = 134)	69.3	0.3178			
Normal (n = 343)	77.3				

AFP, α-fetoprotein; *BMI* body mass index, *CI* confidence interval, *DCP* des-γ-carboxyprothrombin, *HBV* hepatitis B virus, *HCC* hepatocellular carcinoma, *HCV* hepatitis C virus, *ICG R15* indocyanine green retention test at 15 min, *IMAC* intramuscular adipose tissue content, *MST* median survival time, *MVI* microvascular invasion, *PMI* psoas muscle mass index, *TNM* Tumour Node Metastasis (stage defined by the Liver Cancer Study Group of Japan).

Positive	152 (32)	70 (33)	82 (31)	0.5010
Negative	325 (68)	139 (67)	186 (69)	
Differentiation of HCC, n (%)				
Well	48 (10)	17 (8)	31 (12)	0.5354
Moderate	301 (63)	131 (63)	170 (63)	
Poor	113 (24)	54 (26)	59 (22)	
Unknown	15 (3)	7 (3)	8 (3)	
TNM stage, n (%)				
I	82 (17)	31 (15)	51 (19)	0.5823
II	181 (38)	81 (39)	100 (37)	
III	142 (30)	62 (30)	80 (30)	
IV	72 (15)	35 (17)	37 (14)	
Surgical Procedure, n (%)				
≥Lobectomy	167 (35)	75 (36)	92 (34)	0.7237
<Segmentectomy	310 (65)	134 (64)	176 (66)	
Operative time (min)				
Mean (SD)	380 (138)	382 (140)	379 (137)	0.9118
Operative blood loss (ml)				
Mean (SD)	1264 (2210)	1399 (2568)	1160 (1884)	0.0225

AFP α-fetoprotein, *BMI* body mass index, *DCP* des-γ-carboxyprothrombin, *HBV* hepatitis B virus, *HCC* hepatocellular carcinoma, *HCV* hepatitis C virus, *ICG R15* indocyanine green retention test at 15 min, *IMAC* intramuscular adipose tissue content, *MVI* microvascular invasion, *PMI* psoas muscle mass index, *SD* standard deviation, *TNM* Tumour Node Metastasis (stage defined by the Liver Cancer Study Group of Japan).

Percentages might not add up to 100% because of rounding.

Table 3 Prognostic factors for recurrence-free survival on univariate and multivariate analysis

	Univariate analysis		Multivariate analysis		
	MST (months)	*P*	Hazard ratio	95% CI	*P*
Age (years)					
≥65 (n = 169)	20.5	0.5766			
<65 (n = 308)	17.7				
Gender					
Male (n = 389)	21	0.8394			
Female (n = 88)	18.2				
Etiology of HCC					
HBV or/and HCV (n =319)	17.7	0.1694			
Others (n = 158)	26.3				
The presence of previous treatment for HCC					
Yes (n = 188)	17.6	0.0735	1.254	(0.967-1.620)	0.0879
No (n = 289)	21.8		Reference		
Platelet count ($\times 10^4/mm^3$)					
<10 (n = 101)	14.9	0.0139	1.201	(0.876-1.623)	0.2504
≥10 (n = 376)	21.5		Reference		
ICG R15 (%)					
≥15 (n = 245)	18.9	0.2085			
<15 (n = 232)	20.2				
Child-Pugh classification					
A (n = 437)	19.8	0.6930			
B (n = 40)	18.2				
AFP (ng/dl)					
≥20 (n = 229)	14.2	0.0043	1.182	(0.914-1.530)	0.2027
<20 (n = 248)	28.1		Reference		
DCP (munits/l)					
≥40 (n = 338)	16.4	0.0008	1.343	(0.994-1.830)	0.0550
<40 (n = 139)	33.4		Reference		
Liver histology					
Normal liver or chronic hepatitis (n = 239)	24.3	0.0416	0.674	(0.521-0.871)	0.0026
Liver fibrosis or liver cirrhosis (n = 238)	17.7		Reference		
Tumor size (cm)					
≥5.0 (n =170)	10.8	0.0023	1.440	(1.063-1.920)	0.0186
<5.0 (n = 307)	23.8		Reference		

Number of tumors					
Multiple (n = 153)	11.4	<0.0001	1.299	(0.955-1.757)	0.0947
Solitary (n = 324)	26.3		Reference		
MVI					
Positive (n = 152)	8.9	<0.0001	1.491	(1.082-2.042)	0.0148
Negative (n = 325)	27		Reference		
Tumor differentiation					
Well or moderate (n = 349)	20.7	0.0043	0.956	(0.712-1.297)	0.7708
Poor (n = 113)	11.3		Reference		
TNM stage					
I or II (n = 263)	32.6	<0.0001	0.553	(0.394-0.774)	0.0006
III or IV (n = 214)	8.9		Reference		
Surgical procedure					
≥Lobectomy (n = 167)	12.5	0.0063	1.350	(0.986-1.855)	0.0613
<Segmentectomy (n = 310)	23.5		Reference		
Operative time (min)					
≥360 (n = 250)	18.1	0.2319			
<360 (n = 227)	20.1				
Operative blood loss (ml)					
≥500 (n =310)	16.4	0.0066	1.115	(0.832-1.488)	0.4647
<500 (n = 167)	26.8		Reference		
BMI (kg/m^2)					
≥25 (n = 109)	21.5	0.6060			
<25 (n = 368)	18.9				
Preoperative IMAC					
High (n = 209)	15	0.0012	1.529	(1.196-1.953)	0.0007
Normal (n = 268)	26.3		Reference		
Preoperative PMI					
Low (n = 134)	18.8	0.7468			
Normal (n = 343)	19.8				

AFP, α-fetoprotein; *BMI* body mass index, *CI* confidence interval, *DCP* des-γ-carboxyprothrombin, *HBV* hepatitis B virus, *HCC* hepatocellular carcinoma, *HCV* hepatitis C virus, *ICG R15* indocyanine green retention test at 15 min, *IMAC* intramuscular adipose tissue content, *MST* median survival time, *MVI* microvascular invasion, *PMI* psoas muscle mass index, *TNM* Tumour Node Metastasis (stage defined by the Liver Cancer Study Group of Japan).