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Original Article

Increased levels of serum leptin are a risk factor for the recurrence of stage I/II hepatocellular carcinoma after curative treatment

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Obesity and related adipocytokine disbalance increase the risk of hepatocellular carcinoma. To determine the impact of increased levels of leptin, an obesity-related adipocytokine, on the recurrence of hepatocellular carcinoma, we conducted a prospective case-series analysis. Eighty-five consecutive primary hepatocellular carcinoma patients at our hospital from January 2006 to December 2008 were analyzed. Serum leptin level significantly correlated with Body Mass Index, total body fat, and the amount of subcutaneous fat. They included 33 with stage I/II, who underwent curative treatment. The factors contributing to recurrence of hepatocellular carcinoma, including leptin, were subjected to univariate and multivariate analyses using the Cox proportional hazards model. Body Mass Index ($p = 0.0062$), total body fat ($p = 0.0404$), albumin ($p = 0.0210$), α-fetoprotein ($p = 0.0365$), and leptin ($p = 0.0003$) were significantly associated with the recurrence of hepatocellular carcinoma in univariate analysis. Multivariate analysis suggested that leptin (hazard ratio 1.25, 95% CI 1.07–1.49, $p = 0.0035$) was a sole independent predictor. Kaplan-Meier analysis showed that recurrence-free survival was lower in patients with greater serum leptin concentrations (>5 ng/mL, $p = 0.0221$). These results suggest that the serum leptin level is a useful biomarker for predicting the early recurrence of hepatocellular carcinoma.

Key Words: **hepatocellular carcinoma, carcinogenesis, leptin, obesity, insulin resistance**

Hepatocellular carcinoma (HCC) is the fifth most common malignancy worldwide and is estimated to cause approximately 500,000 deaths annually.[1] HCC frequently develops and in many cases recurs in cirrhotic livers due to persistent hepatitis B virus (HBV) and hepatitis C virus (HCV) infection; this is strongly associated with poor prognosis for this particular malignancy.[2] Therefore, careful surveillance of high-risk groups for HCC is important to improve prognosis. Hence, there is a critical need to identify useful risk factors for the development of HCC. Infection with HBV and HCV, alcohol consumption, aflatoxin exposure, and immune-related hepatitis are accepted as significant risk factors for the development of primary HCC.[3] Male gender, the presence of cirrhosis, high α-fetoprotein (AFP), large tumor foci, multiplicity of tumors, pathologically high-grade atypia of tumor cells, and the presence of portal venous invasion of tumors also raise the risk for HCC recurrence.[4–8]

In addition to these factors, recent studies demonstrate that obesity[9] and related metabolic abnormalities—especially diabetes mellitus (DM) and insulin resistance[10,11]—are important risk factors for the development of HCC. For instance, insulin resistance significantly raises the risk of the recurrence of stage I HCC after curative treatment.[10] Several pathophysiological mechanisms linking obesity and HCC development have been proposed and include the emergence of insulin resistance and a state of chronic inflammation.[12,13] Adipocytokine disbalance might also be involved in obesity-related liver carcinogenesis.[14] Among the adipocytokines, it is well known that the serum levels of leptin, which regulate the homeostasis of glucose and lipid metabolism,[15] are elevated in obese individuals.[16] In addition, both *in vitro* and *in vivo* studies indicate that leptin might play a role in the development of several types of human malignancies, including HCC.[17–21] These findings suggest that the dysregulation of serum leptin levels may be a critical link between obesity and liver carcinogenesis. However, whether leptin is a significant biomarker for predicting the development and/or recurrence of HCC has not been explored.

In this study, we measured the serum leptin concentration in patients with HCC and examined whether it is correlated with obesity and insulin resistance. In addition, we designed a prospective case-series analysis to examine the recurrence-free survival in consecutive patients with stage I/II HCC, who received curative treatment by surgical resection or radiofrequency ablation (RFA), stratified by serum leptin concentrations.

Materials and Methods

Patients. From January 2006 to December 2008, 85 primary HCC patients underwent initial treatment at our hospital. We measured visceral and subcutaneous fat volume using computed tomography (CT) scans at the umbilical level according to a previously reported technique (fatAnalyses and EV Insite R, PSP Corporation, Tokyo, Japan).[22] Tumor stage was defined according to the staging system of the Liver Cancer Study Group of Japan (LCSGJ).[23] HCC nodules were detected by imaging modalities including abdominal ultrasonography (US), dynamic CT, dynamic magnetic resonance imaging (MRI), and abdominal arteriography. The diagnosis of HCC was made from a typical hypervascular tumor stain on angiography and a typical dynamic-study finding of enhanced staining in the early phase and attenuation in the delayed phase.

Treatment, follow-up, and determination of recurrence. Fifteen patients were treated with surgical resection, 41 with RFA, 19 with transarterial chemoembolization (TACE), and 10 with transarterial infusion (TAI). Among them, we selected 33 curative cases that met the following criteria: tumor stage classified as I or II; and surgical resection or RFA conducted for the initial HCC treatment. In all 33 cases, therapeutic effects were judged to be

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Table 1. Baseline demographic and clinical characteristics

Variable	Total patients (n = 85)
Sex (male/female)	54/31
Age (years)	73 [36–87]
BMI (kg/m^2)	23.2 [17.5–30.7]
Total body fat (cm^2)	188.81 [12.93–501.8]
Amount of visceral fat (cm^2)	76.43 [3.82–359.83]
Amount of subcutaneous fat (cm^2)	105.66 [9.11–265.26]
Etiology (B/C/B + C/other)*	8/55/2/20
Child-Pugh classification (A/B/C)	60/23/2
Ascites on CT imaging (present/absent)	7/78
ALB (g/dL)	3.5 [2.2–4.5]
PLT ($\times 10^4/\mu$L)	11.7 [3.0–76.4]
FPG (mg/dL)	97 [67–271]
FIRI (μU/mL)	8.115 [1.21–90.2]
HOMA-IR	2.245 [0.27–28.28]
HbA_{1c} (%)	5.3 [3.7–10.3]
Leptin (ng/mL)	5.0 [1.4–26.6]
Stage (I/II/III/IVA/IVB)	19/26/27/10/3
Initial treatment for HCC (resection/RFA/TACE/TAI)	15/41/19/10
AFP (ng/mL)	48 [0–222000]
PIVKA-II (mAU/mL)	170 [7–474000]
Follow-up period (days)	484 [14–1429]

Values are median [range]. *B means positive for hepatitis B surface antigen and C means positive for hepatitis C virus antibody. AFP, α-fetoprotein; B, hepatitis B virus; BMI, body mass index; C, hepatitis C virus; FPG, fasting plasma glucose; FIRI, fasting immunoreactive insulin; HbA_{1c}, hemoglobin A_{1c}; HCC, hepatocellular carcinoma; HOMA-IR, homeostasis model assessment of insulin resistance; PIVKA-II, protein induced by vitamin K absence or antagonists-II; RFA, radiofrequency ablation; TACE, transarterial chemoembolization; TAI, transarterial infusion.

curative using dynamic CT or MRI exhibiting a complete disappearance of the imaging characteristics of HCC described above.

Patients were thereafter followed up on a monthly outpatient basis using serum tumor markers every month, such as AFP and proteins induced by vitamin K absence or antagonists-II (PIVKA-II), and by abdominal US, dynamic CT scanning, or dynamic MRI every 3 months. Recurrent HCC was diagnosed, using the imaging modalities described earlier, by the appearance of other lesions differed from the primary lesions. The follow-up period was defined as the interval from the date of initial treatment until the date of diagnosis of recurrence or until April 2009 if HCC did not recur.

Statistical analysis. The Pearson product-moment correlation coefficient was used for measuring the linear correlation between 2 continuous variables. Recurrence-free survival was estimated using the Kaplan-Meier method, and differences between curves were examined with a log-rank test. Baseline characteristics were compared using Student's t test for continuous variables or the χ^2 test for categorical variables. There were 17 possible predictors for the recurrence of HCC after the initial curative treatment: sex, age, body mass index (BMI), total body fat, amounts of both visceral and subcutaneous fat, the presence of HCV-antibodies (HCV-Ab), Child-Pugh classification, serum albumin concentration, platelet count, homeostasis model assessment of insulin resistance (HOMA-IR = fasting plasma glucose (mg/dL) × fasting immunoreactive insulin (μU/mL)/405), hemoglobin A_{1c} (HbA_{1c}), serum tumor markers (AFP and PIVKA-II), initial treatment for HCC, tumor stage, and serum leptin concentration. Parameters that were significant as determined by univariate analysis were then subjected to multivariate analyses using the Cox proportional hazards model. Statistical significance was defined as $p<0.05$.

Results

Baseline characteristics and laboratory data of patients. The baseline characteristics and laboratory data of 85 patients (54 men and 31 women, median age 73 years) are shown in Table 1. The median follow-up period was 484 days (range, 14–1,429 days). Median BMI was 23.2 kg/m^2, which was classified in the normal range according to the WHO classification of obesity (http://www.who.int/bmi). Median free plasma glucose (FPG), free immunoreactive insulin (FIRI), HOMA-IR, and HbA_{1c} were 97 mg/dL, 8.115 μU/mL, 2.245, and 5.3%, respectively. The median serum leptin concentration was 5.0 ng/mL (range 1.4–26.6).

Association of the serum leptin concentration with obesity and insulin resistance. Four obesity-related factors were tested for possible association with the serum leptin concentration: BMI, total body fat, and the amounts of visceral and subcutaneous fat (Fig. 1). For BMI analysis, we excluded 7 patients with CT-detected ascites. The Pearson product-moment correlation coefficient and p values of BMI and the total body fat with serum leptin concentration were $r = 0.4559$ and $p<0.0001$, and $r = 0.3560$ and $p = 0.0008$, respectively; indicating that these 2 factors were significantly correlated with the serum leptin concentration. The amount of subcutaneous fat ($r = 0.5174$ and $p<0.0001$) was also strongly correlated with the serum leptin level, whereas the amount of visceral fat ($r = 0.0987$ and $p = 0.3776$) was not. In addition, no significant correlations were noted between the serum leptin concentration and insulin resistance-related factors, including FPG ($r = -0.0816$ and $p = 0.4579$), FIRI ($r = 0.1049$ and $p = 0.3378$), HOMA-IR ($r = 0.0506$ and $p = 0.6385$), and HbA_{1c} ($r = 0.0194$ and $p = 0.7820$).

Possible risk factors for the recurrence of HCC. In all 33 curative cases of stage I/II HCC, 12 patients experienced recur-

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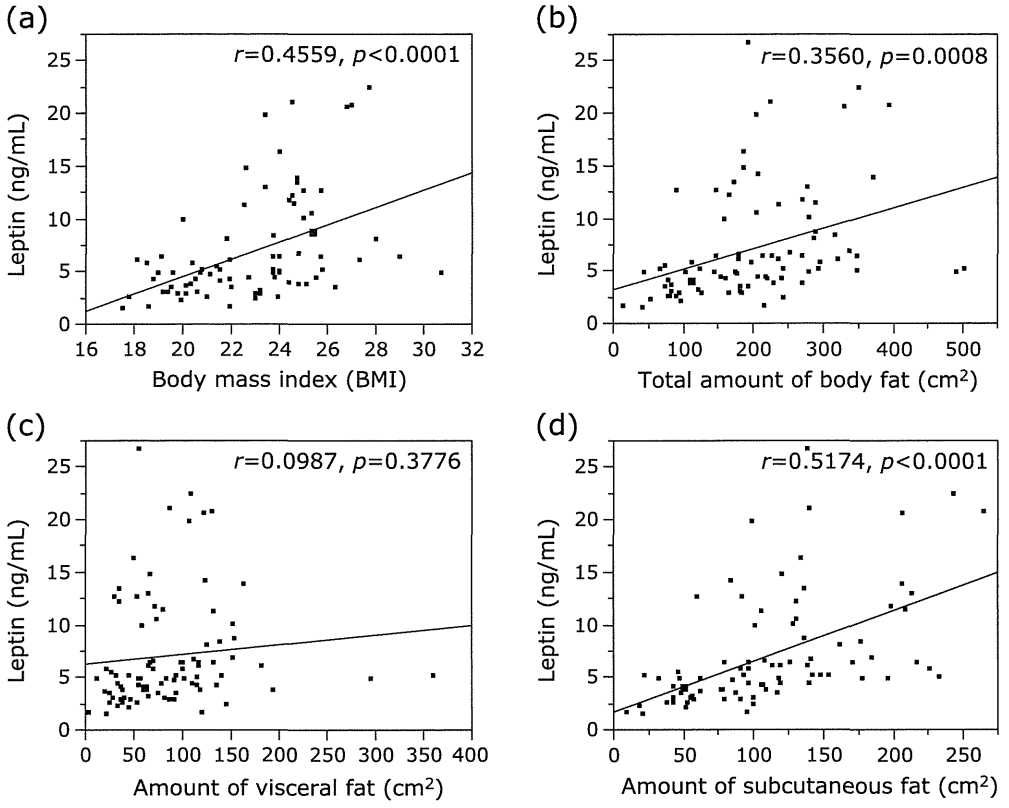

Fig. 1. Correlation between the serum levels of leptin and (a) BMI, (b) total body fat, (c) amount of visceral fat, and (d) amount of subcutaneous fat in patients with HCC (n = 85). For BMI analysis, we excluded 7 patients with CT-detected ascites.

rence in the liver, but none exhibited distant metastasis. The 1-year recurrence-free survival in the 33 patients was 79% (Fig. 2a). Fig. 2b shows Kaplan-Meier curves for recurrence-free survival divided into 2 subgroups on the basis of median serum leptin concentration (≤5 or >5 ng/mL), which results in a significant difference (p = 0.0221).

The Cox proportional hazards model was used to analyze risk factors for the recurrence of stage I/II HCC after curative treatments using the 17 variables listed in Table 2. BMI (hazard ratio 1.30, 95% CI 1.08–1.56, p = 0.0062), total body fat (hazard ratio 1.00, 95% CI 1.00–1.01, p = 0.0404), serum albumin concentration (hazard ratio 0.26, 95% CI 0.08–0.81, p = 0.0210), AFP (hazard ratio 0.99, 95% CI 0.99–0.99, p = 0.0365), and serum leptin concentration (hazard ratio 1.29, 95% CI 1.12–1.50, p = 0.0003) were identified as significant risk factors by univariate analysis. Multivariate analysis only identified serum leptin concentration (hazard ratio 1.25, 95% CI 1.07–1.49, p = 0.0035) as significant independent risk factor for the recurrence of HCC (Table 3).

Table 4 shows the baseline characteristics and laboratory data of patients divided on the basis of the serum leptin concentration (≤5 and >5 ng/mL). No significant differences were noted between the 2 subgroups, except the amount of subcutaneous fat (p = 0.0461).

Discussion

Leptin regulates body weight by signaling information to the brain regarding the availability of energy stored as fat; this negative feedback loop is disrupted in most obese individuals and results in a state known as leptin resistance.[16,24] Consistent with the results of previous studies,[16,24] the serum leptin concentration was significantly correlated with BMI and total body fat in the present study (Fig. 1 a and b). These parameters were also significant risk factors for the recurrence of HCC as determined by univariate analysis (Table 2); however, the serum leptin concentration was the most significant biomarker (p = 0.0003).

In addition, we clearly showed for the first time that patients with greater serum leptin concentrations were susceptible to HCC recurrence (Fig. 2b); thus, increased serum leptin levels are a significant independent risk factor for the recurrence of this malignancy (Table 3). This finding indicates that increased serum leptin concentration, which might link obesity with liver carcinogenesis, is a preferable and useful biomarker for screening high-risk groups for the recurrence of HCC. We previously reported that a state of insulin resistance associated with obesity is an independent risk factor for the recurrence of HCC after curative treatment.[10] Furthermore, no significant correlations between serum leptin levels and insulin resistance-related factors were noted in the present study, suggesting these two conditions might be independent from each other in HCC patients. Therefore, a combination evaluation for both the serum leptin level and insulin resistance would be more effective for screening high-risk groups for HCC, and requires future confirmation.

Several studies report that leptin is a risk factor for carcinogenesis at various organ sites, including the liver.[17–21] Leptin can stimulate cellular proliferation in various types of cancer cells such as HCC cells.[19–21,25] In addition, when focusing on the liver,

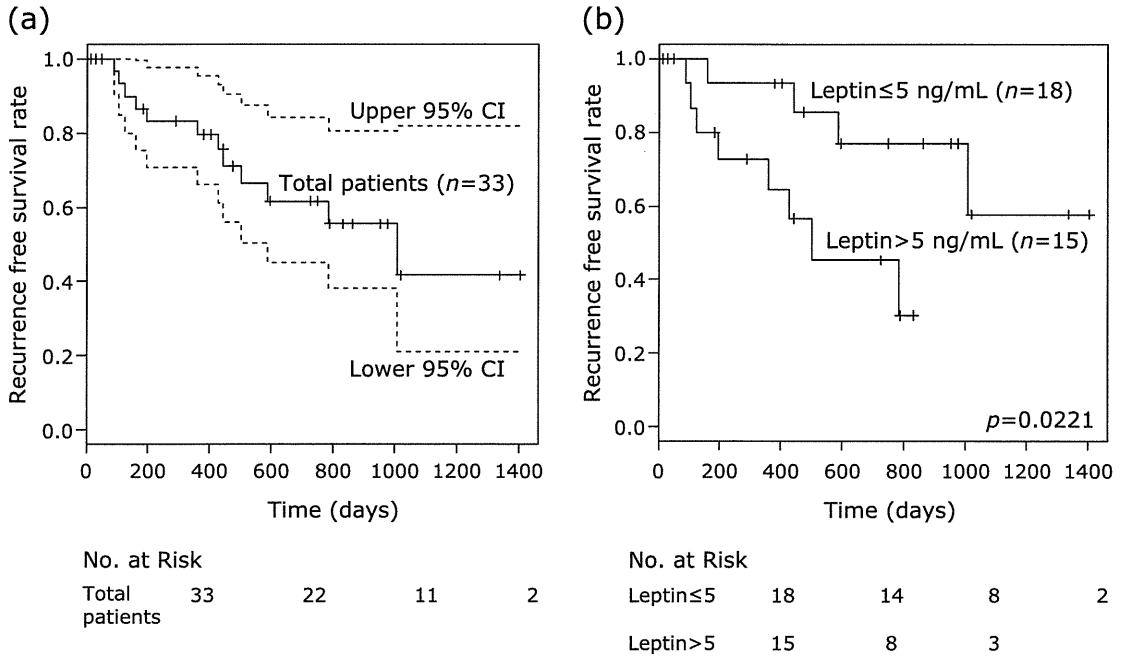

Fig. 2. Kaplan-Meier curves for recurrence-free survival in (a) total patients and in (b) subgroups divided on the basis of the serum leptin concentration (≤5 or >5 ng/mL).

Table 2. Univariate analyses of possible risk factors for recurrence-free survival of HCC using the Cox proportional hazards model

Variable	95% CI			
	HR*	lower	upper	*p* value
Sex (male vs female)	0.9	0.28	3.09	0.8726
Age (years)	0.96	0.89	1.03	0.277
BMI (kg/m^2)	1.3	1.08	1.56	0.0062
Total body fat (cm^2)	1	1	1.01	0.0404
Amount of visceral fat (cm^2)	1	0.99	1.01	0.0909
Amount of subcutaneous fat (cm^2)	1	0.99	1.01	0.0601
The presence of HCV-Ab (yes vs no)	0.42	0.12	1.98	0.2501
Child-Pugh classification (B + C vs A)	1.33	0.35	4.3	0.6482
ALB (g/dL)	0.26	0.08	0.81	0.021
PLT (×10^4/μL)	0.87	0.75	1.01	0.0714
HOMA-IR	1.03	0.94	1.1	0.4
HbA_{1c} (%)	0.87	0.37	1.6	0.7108
AFP (ng/mL)	0.99	0.99	0.99	0.0365
PIVKA-II (mAU/mL)	0.99	0.99	1	0.7448
Initial treatment for HCC (RFA vs resection)	1.61	0.42	10.5	0.5128
Stage (II vs I)	1.08	0.32	3.78	0.89
Leptin (ng/mL)	1.29	1.12	1.5	0.0003

*HR represents the values with a unit increase in continuous variables. AFP, α-fetoprotein; BMI, body mass index; CI, confidence interval; HbA_{1c}, hemoglobin A_{1c}; HCC, hepatocellular carcinoma; HCV-Ab, hepatitis C virus antibody; HOMA-IR, homeostasis model assessment of insulin resistance; HR, hazard ratio; PIVKA-II, protein induced by vitamin K absence or antagonists-II; RFA, radiofrequency ablation.

leptin is a potent profibrogenic cytokine and thus plays a key role in the progression of cirrhosis,[26] which is a precancerous condition of HCC. Indeed, increased serum leptin concentration has been documented in cirrhotic patients.[27,28] Moreover, increased leptin expression is associated with increased intratumor microvascular density. Consequently, it is hypothesized that leptin plays a stimulatory role in the development of HCC via neovascularization.[29] In addition to using leptin as a biomarker for the risk of HCC recurrence, the present findings suggest that targeting leptin might be an effective strategy for the prevention and treatment of

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Table 3. Multivariate analyses of possible risk factors for recurrence-free survival of HCC using the Cox proportional hazards model

Variable	95% CI			
	HR*	lower	upper	*p* value
BMI (kg/m^2)	1.2	0.83	1.81	0.3278
Total body fat (cm^2)	1	0.99	1.01	0.8003
ALB (g/dL)	0.54	0.12	2.28	0.4018
AFP (ng/mL)	0.99	0.99	1	0.1416
Leptin (ng/mL)	1.25	1.07	1.49	0.0035

*HR represents the values with a unit increase in continuous variables. AFP, α-fetoprotein; BMI, body mass index; CI, confidence interval; HR, hazard ratio.

Table 4. Baseline demographic and clinical characteristics of patients classified on the basis of the serum leptin concentration

Variable	Leptin ≤ 5 ng/mL (*n* = 18)	Leptin > 5 ng/mL (*n* = 15)	*p* value
Sex (male/female)	13/5	6/9	0.0604
Age (years)	72.5 [59–87]	70 [50–85]	0.2565
BMI (kg/m^2)	21.5 [17.8–30.7]	24.5 [18.5–27.7]	0.1111
Total body fat (cm^2)	167.5 [73.9–490.9]	207.3 [112.2–350.8]	0.2591
Amount of visceral fat (cm^2)	69.4 [19.9–294.4]	98.9 [21.9–181.6]	0.9479
Amount of subcutaneous fat (cm^2)	90.2 [42.0–232.3]	134.3 [79.6–242.5]	0.0461
Etiology (C/others)	14/4	11/4	0.767
Child-Pugh classification (A/B/C)	15/3/0	10/5/0	0.2657
ALB (g/dL)	3.6 [2.6–4.2]	3.3 [2.4–4.4]	0.2708
PLT (×10^4/μL)	12.45 [7.7–26.1]	9.5 [3.0–20.6]	0.0895
FPG (mg/dL)	97.5 [83–271]	105 [75–154]	0.7424
FIRI (μU/mL)	6.05 [2.57–65.2]	14.3 [7.3–27.4]	0.3657
HOMA-IR	1.51 [0.53–24.8]	3.41 [1.45–9.40]	0.641
HbA_{1c} (%)	5.3 [4.5–10.3]	5.2 [3.7–6.8]	0.3351
Stage (I/II)	7/11	9/6	0.2253
Initial treatment for HCC (resection/RFA)	6/12	2/13	0.1726
AFP (ng/mL)	8 [0–20500]	28 [1–2530]	0.1687
PIVKA-II (mAU/mL)	22.7 [8–201000]	26 [7–29800]	0.4385

Values are median [range]. AFP, α-fetoprotein; C, hepatitis C virus; FPG, fasting plasma glucose; FIRI, fasting immunoreactive insulin; HbA_{1c}, hemoglobin A_{1c}; HCC, hepatocellular carcinoma; HOMA-IR, homeostasis model assessment of insulin resistance; PIVKA-II, protein induced by vitamin K absence or antagonists-II; RFA, radiofrequency ablation.

HCC. Ribatti *et al.* state that anti-leptin antibodies reduce the angiogenic response in HCC biopsy specimens.[29] Decreases in serum leptin are also associated with the prevention of obesity-related liver tumorigenesis in obese and diabetic mice models.[14]

In conclusion, we report that patients with high serum leptin concentrations are susceptible to HCC recurrence in stage I/II cases curatively treated by surgical resection or RFA. Increased serum leptin concentration may be a useful biomarker for predicting the recurrence of HCC in high-risk patients.

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Abbreviations

AFP	α-fetoprotein
BMI	body mass index
CT	computed tomography
DM	diabetes mellitus
FPG	fasting plasma glucose
FIRI	fasting immunoreactive insulin
HbA_{1c}	hemoglobin A_{1c}
HBV	hepatitis B virus
HCC	hepatocellular carcinoma
HCV	hepatitis C virus
HOMA-IR	homeostasis model assessment of insulin resistance
LCSGJ	Liver Cancer Study Group of Japan
MRI	magnetic resonance imaging
PIVKA-II	protein induced by vitamin K absence or antagonists-II
RFA	radiofrequency ablation
TACE	transarterial chemoembolization
TAI	transarterial infusion
US	ultrasonography

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Original Article

Novel scoring system as a useful model to predict the outcome of patients with acute liver failure: Application to indication criteria for liver transplantation

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Aim: In Japan, the indication for liver transplantation in patients with acute liver failure (ALF) is currently determined according to the guideline published in 1996. However, its predictive accuracy has fallen in recent patients. Thus, we attempted to establish a new guideline.

Methods: The subjects were 1096 ALF patients enrolled in a nationwide survey. All patients showed a prothrombin time <40% of the standardized value and grade II or more severe hepatic encephalopathy. A multiple logistic regression analysis and receiver operating characteristic analysis were performed in 698 patients seen between 1998 and 2003 to identify significant parameters determining the outcome of patients. The extracted parameters were graded as numerical scores. An established scoring system was validated in patients seen between 2004 and 2008.

Results: Six parameters were identified and graded as 0, 1 and/or 2; the interval between disease onset and development of hepatic encephalopathy, prothrombin time, serum total bilirubin concentration, the ratio of direct to total bilirubin concentration, peripheral platelet count and the presence of liver atrophy. When the prognosis of the patients with total score of 5 or more was judged as "death", the predictive accuracy was 0.80 with sensitivity, specificity, positive predictive value and negative predictive value greater than 0.70. The values were similarly high in patients for validation.

Conclusion: Novel scoring system for predicting the outcome of ALF patients may be useful to determine the indication of liver transplantation, since the system showed high predictive accuracy even after validation.

Key words: acute liver failure, fulminant hepatitis, guideline, indication criteria, liver transplantation, outcome prediction

INTRODUCTION

ACUTE LIVER FAILURE is a disease entity characteristic with extensive destruction of liver parenchyma by hepatitis virus infection and other causes, and is typically represented by fulminant hepatitis. Although the outcome may differ depending on the etiology of acute liver failure, survival rates of patients receiving conventional medical care are generally low in cases with impaired liver regeneration. Hepatitis patients are diagnosed as fulminant hepatitis in Japan if grade II or deeper hepatic encephalopathy develops within 8 weeks of the onset of hepatitis symptoms due to severe abnormality of the liver function with prothrombin time lower than 40% of the standardized value. Fulminant hepatitis is further classified into two subtypes according to clinical

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course; acute type with hepatic encephalopathy developing within 10 days of disease onset and subacute type with hepatic encephalopathy at 11 days or later.[1,2] In addition, late onset hepatic failure (LOHF) is defined as a related disease of fulminant hepatitis, in which hepatic encephalopathy develops between 8 and 24 weeks after the onset of hepatitis symptoms.[2] Fulminant hepatitis in Japan is defined as acute liver failure with histological appearance of hepatic inflammation, such as lymphocyte infiltration in the liver. Thus, the etiology of fulminant hepatitis comprises viral hepatitis including persistent hepatitis B virus (HBV) carriers, autoimmune hepatitis, drug-induced/allergic hepatitis, and hepatitis with indeterminate etiologies, but excludes drug-induced/toxic liver damage, acute fatty liver in pregnancy, postoperative liver damage and ischemic liver damage. However, in Europe and the United States, the latter causes are included in the disease entity of acute liver failure, formerly called fulminant hepatic failure. Thus, for example, acetaminophen-induced liver damage is included in acute liver failure in Europe and the United States, while it is excluded from the disease entity of fulminant hepatitis in Japan.[3] Considering such differences of the definition and diagnostic criteria between fulminant hepatitis in Japan and acute liver failure in the United States and Europe, the guidelines in the latter countries to determine the indication of liver transplantation for acute liver failure are not directly applicable to fulminant hepatitis in Japan.

It is accepted worldwide that liver transplantation is the most effective therapeutic modality for patients with acute liver failure. In Japan, indication criteria for liver transplantation in patients with fulminant hepatitis were defined in 1996, as a two-step outcome prediction scoring system, by the Acute Liver Failure Study Group of Japan (Table 1).[4,5] According to this guideline, the outcome of fulminant hepatitis patients is predicted at the onset of grade II or more severe hepatic encephalopathy based on five parameters: age of patient, the interval between occurrence of hepatitis symptoms and development of hepatic encephalopathy, prothrombin time, serum total bilirubin concentration and the ratio of the serum direct to total bilirubin concentration. Then, in patients undergoing intensive medical care including artificial liver support, their prognosis is reassessed 5 days later, according to the extent of improvement of hepatic encephalopathy and prothrombin time. This guideline was prepared based on the clinical findings in fulminant hepatitis patients seen between 1988 and 1992, and was considered to be useful, since the predictive accuracy was found to be 83% in a prospective analysis conducted in patients seen between 1993 and 1995.[4] However, the predictive accuracy of the guideline fell in patients with fulminant hepatitis seen between 1998 and 2003; 68% and 78% in the acute and subacute types, respectively, and the values did not improve following reassessment at 5 days later.[6]

Table 1 Guideline to determine the indications for liver transplantation in patients with fulminant hepatitis by the Acute Liver Failure Study Group of Japan in 1996

Patients may be registered as recipients of liver transplantation when at least two of the five criteria are satisfied at the time of onset of Grade II or more severe hepatic encephalopathy.

1 Age ≥45 years.
2 Interval from the appearance of the initial symptoms to the development of hepatic encephalopathy ≥11 days.
3 Prothrombin time <10% of the standardized value.
4 Serum bilirubin concentration ≥18.0 mg/dL.
5 Ratio of the direct to total bilirubin concentration <0.67.

If liver transplantation cannot be performed within 5 days and intensive medical therapy, including artificial liver support, is undertaken, the prognosis of the patients is evaluated again. If both of the following two criteria are positive at 5 days after the onset of hepatic encephalopathy, the patients are re-predicted as "alive" and excluded from the candidate list for liver transplantation.

1 The hepatic encephalopathy shows improvement to Grade I or less or attenuation by two more grades.
2 Prothrombin time improves to over 50% of the standardized value.

English version of this guideline was published in Mochida *et al.*[5]

To improve the predictive accuracy of indication criteria for liver transplantation in patients with fulminant hepatitis, the Study Group of Intractable Hepatobiliary Diseases supported by the Ministry of Health, Labor, and Welfare of Japan organized a task force in 2006. The task force first analyzed the database obtained from patients with fulminant hepatitis and LOHF seen between 1998 and 2003 in Japan to establish the novel scoring system to predict the outcome of the patients. Then, the established system was evaluated in the patients seen between 2004 and 2008. In the present paper, we report on the usefulness of this novel scoring system. We state here that the new system is intended for use in a general cohort of acute liver failure, but is actually organized on the database of registered patients with fulminant hepatitis and LOHF. Thus, validation of

the system for acute liver failure due to other etiologies as described earlier awaits future study.

METHODS

Patients

THE STUDY SUBJECTS are 1096 patients with acute liver failure who were enrolled in the nationwide survey by the Intractable Hepato-Biliary Disease Study Group of Japan between 1999 and 2008 (formerly the Intractable Liver Diseases Study Group of Japan before 2003). All of the patients showed grade II or more severe hepatic encephalopathy and prothrombin time of less than 40% of the standardized value and were admitted to 610 hospitals of Japan specializing in hepatology between 1998 and 2008. The patients consisted of three disease types; 505 and 449 patients, respectively, with acute and subacute types of fulminant hepatitis and 88 patients with LOHF. They were divided into two cohorts; 698 patients (316, 318 and 64 patients, respectively, with acute and subacute types of fulminant hepatitis and LOHF) seen between 1998 and 2003 (the estimation cohort) and 394 patients (189, 191 and 24 patients, respectively, of each disease type) seen between 2004 and 2008 (the validation cohort). From both cohorts, the patients with incomplete records and those treated with liver transplantation were excluded. Thus, the estimation cohort included 421 patients (201 and 178 patients, respectively, with acute and subacute types of fulminant hepatitis and 41 patients with LOHF) and the validation cohort recruited 231 patients (125, 95 and 11 patients, respectively, in each disease type).

Etiologies of hepatitis in the estimation and validation cohorts are given in Table 2. Demographic and clinical features of patients in each cohort are shown in Tables 3 and 4, respectively. These features did not differ between the two cohorts, except that the ages of the patients were greater in the validation cohort than in the estimation cohort. The survival rates of patients were equivalent between two cohorts; 37.4% in the estimation cohort and 37.7% in the validation cohort.

Identification of prognostic factors responsible for the outcome of patients with fulminant hepatitis and LOHF in the estimation cohort

First, univariate logistic analysis was performed in patients of the estimation cohort to identify possible prognostic factors among demographic and clinical features at the onset of grade II or more severe hepatic encephalopathy as follows. (i) *Demographic features*; sex and age of patients, the types of disease (acute and subacute types of fulminant hepatitis and LOHF), the interval (days) between the onset of hepatitis symptoms and the development of hepatic encephalopathy. (ii) *Symptoms*; fever of 37.5°C or more, convulsion, tachycardia, disappearance of liver dullness on physical examination, flapping tremor, hepatic odor and edema. (iii) *Laboratory parameters*; prothrombin time (%), hepaplastin test (%), antithrombin III activity (%), serum concentrations of albumin (g/dL) and total and direct bilirubin (mg/dL), the ratio of direct to total bilirubin concentration, serum levels of aspartate aminotransferase (AST: IU/L) and alanine aminotransferase (AST: IU/L), serum α-fetoprotein concentration (ng/mL), blood ammonia concentration (μg/dL), plasma concentration of hepatocyte growth factor (HGF: ng/mL), peripheral platelet and white blood cell counts (/mm^3). (iv) *Imaging*; liver atrophy diagnosed by ultrasound sonography and/or computed tomography/magnetic resonance imaging (CT/MRI).

Extracted factors were subjected to multivariate logistic analyses through a stepwise elimination manner. Then a receiver operating characteristic (ROC) curve was constructed for each significant variable.

Table 2 Etiologies of fulminant hepatitis and late onset hepatic failure (LOHF) in the estimation cohort and validation cohort

	Estimation cohort 1998–2003	Validation cohort 2004–2008
HAV	33	15
HBV (acute onset)	104	51
HBV (career)	65	33
HBV (unclassified)	9	16
HCV	8	3
Other Virus	3	5
AIH	26	23
Drug	35	33
Undetermined	132	49
No record	6	3
Total	421	231

Data are expressed as the number of patients.
AIH, autoimmune hepatitis; HAV, hepatitis A virus; HBV, hepatitis B virus; HCV, hepatitis C virus.

Scoring of prognostic factors and predicted mortality of patients with fulminant hepatitis and LOHF in the estimation and validation cohorts

The grading of variables was determined as numerical scores based on the inflection points of each ROC curve.

Table 3 Demographic and clinical features of patients with fulminant hepatitis and late onset hepatic failure (LOHF) seen between 1998 and 2003 (estimation cohort)

	Total ($n = 421$)	Dead patients ($n = 260$)	Surviving patients ($n = 161$)
Sex (Male : Female)	218:202:(1)†	148:111:(1)†	70:91
Age	48.6 ± 16.3‡	53.2 ± 14.8**	41.3 ± 16.0
HBV Carrier	15.2% (64/421)	18.8%** (49/260)	9.3% (15/161)
Disease Type (FHA : FHS : LOHF)	201:178:41	86:138:36**	115:40:5
HGF (ng/mL)	6.0 ± 11.4	7.6 ± 13.9*	3.9 ± 6.0
TB (mg/dL)	14.0 ± 9.1	16.6 ± 9.6**	9.7 ± 6.2
D/T ratio	0.63 ± 0.13	0.62 ± 0.14**	0.66 ± 0.12
PT (%)	22.7 ± 12.6	22.5 ± 13.8*	23.5 ± 11.3
AT (%)	37.3 ± 20.3	35.9 ± 20.3*	41.5 ± 19.9
NH3 (μg/dL)	138.7 ± 82.8	151.6 ± 87.8**	118.1 ± 69.6
PLT (10^4/μL)	12.7 ± 7.7	12.1 ± 8.1*	13.6 ± 6.9
Liver Atrophy (present : absent)	265:156	210:50**	55:106
O-C (days)	21.2 ± 26.7	26.4 ± 29.3**	12.8 ± 19.2

*$P < 0.05$, **$P < 0.01$ versus alive.
†A value in parenthesis means the number of patients with no record regarding the sex.
‡Values are expressed as mean ± standard deviation (SD).
AT, antithrombin III; D/T ratio, ratio of direct to total bilirubin concentrations; FHA, acute type of fulminant hepatitis; FHS, subacute type of fulminant hepatitis; HBV, hepatitis B virus; HGF, hepatocyte growth factor; O-C, intervals between hepatitis onset and hepatic encephalopathy development; PLT, platelet; PT, prothrombin time; TB, total bilirubin.

The total scores were calculated in each patient belonging to the estimation cohort, and the mortality rates were evaluated depending on total scores. Then, ROC analysis was performed again to identify the cut-off value of the total score that can discriminate sharply between survived and dead patients. Finally, sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and predictive accuracy of the established scoring system were calculated when the predicted outcome of patients with the total score greater than the cut-off value was judged as "death".

Predictive accuracies of the established system were confirmed similarly in the validation cohort.

Statistical analysis

All statistical analyses were performed with JMP v7.0 for Macintosh (SAS Institute Inc., Cary, NC, USA). Univariate analyses were performed with analysis of variance (ANOVA) and χ^2 test. Multivariate analyses were performed by multiple logistic regression analysis with stepwise selection.

RESULTS

Prognostic factors responsible for the outcome of patients with fulminant hepatitis and LOHF

UNIVARIATE LOGISTIC ANALYSIS revealed 18 variables including demographic features and clinical characteristics at the appearance of grade II or more severe hepatic encephalopathy may affect the mortality of the patients; age of patients, the interval between disease onset and the development of hepatic encephalopathy, presence of tachycardia and edema, disappearance of liver dullness on physical examination and presence of liver atrophy on imaging examination, serum concentrations of albumin and total bilirubin, the ratio of direct to total bilirubin concentration, serum levels of AST and ALT, blood ammonia level, plasma HGF concentration, prothrombin time, hepaplastin test, antithrombin III activity and peripheral platelet count. These factors were subjected to multivariate logistic analysis with stepwise elimination manner, and 10 variables were identified as significant as shown in Table 5. At this step,

Table 4 Demographic and clinical features of patients with fulminant hepatitis and late onset hepatic failure (LOHF) seen between 2004 and 2008 (validation cohort)

	Total ($n = 231$)	Dead patients ($n = 144$)	Surviving patients ($n = 87$)
Sex (Male : Female)	120:111	74:70	46:41
Age	54.7 ± 15.0†	59.8 ± 12.2**	46.3 ± 15.6
HBV Carrier	14.2% (33/231)	19.4% (28/144)	5.7% (5/87)
Disease Type (FHA : FHS : LOHF)	125:95:11	61:74:9**	64:21:2
HGF (ng/mL)	6.0 ± 9.2	7.1 ± 10.4	3.2 ± 3.6
TB (mg/dL)	13.7 ± 8.9	16.6 ± 9.3**	8.8 ± 5.3
D/T ratio	0.64 ± 0.14	0.64 ± 0.13	0.63 ± 0.16
PT (%)	24.5 ± 12.7	22.0 ± 12.4**	28.4 ± 12.4
AT (%)	38.2 ± 20.4	34.4 ± 21.1*	44.3 ± 17.7
NH3 (μg/dL)	162.0 ± 141.8	191.1 ± 168.8**	116.4 ± 60.7
PLT (10^4/μL)	12.6 ± 7.2	11.5 ± 6.9**	14.3 ± 7.2
Liver Atrophy (Present/absent)	146:85	114:30**	32:50
O-C (days)	15.9 ± 17.0	19.0 ± 18.5**	10.6 ± 12.2

*$P < 0.05$, **$P < 0.01$ versus alive.
†Values are expressed as mean ± standard deviation (SD).
AT, antithrombin III; D/T ratio, ratio of direct to total bilirubin concentrations; FHA, acute type of fulminant hepatitis; FHS, subacute type of fulminant hepatitis; HBV, hepatitis B virus; HGF, hepatocyte growth factor; O-C, intervals between hepatitis onset and hepatic encephalopathy development; PLT, platelet; PT, prothrombin time; TB, total bilirubin.

"age of patients" was excluded from the list of candidate variables in order to facilitate the system to be available in pediatric patients. Then ROC curve was constructed for each variable, and six variables with the greatest area under the curve (AUC) were identified; the interval between disease onset and the development of hepatic encephalopathy, prothrombin time, serum concentration of total bilirubin, the ratio of direct to total bilirubin concentration, peripheral platelet count, and liver atrophy.

Table 5 Prognostic factors to affect the outcome of patients with fulminant hepatitis and late onset hepatic failure (LOHF): multivariate logistic analysis in those seen between 1998 and 2003 (estimation cohort)

	Odds ratio†	(95% confidence interval)	*P*-value
Liver atrophy	9.777		<0.0001
TB	1.0993	(1.043–1.168)	0.0009
D/T ratio	0.000446	(0.0001146–0.081412)	0.0062
NH3	1.007	(1.002–1.014)	0.0098
Age	1.0654	(1.010–1.136)	0.0113
PT%	0.9773	(0.959–0.995)	0.0115
HGF	1.1837	(1.049–1.374)	0.0139
O-C	1.0687	(1.014–1.141)	0.0270
ALB	0.0409	(0.129–0.906)	0.0312
PLT	0.9648	(0.931–0.999)	0.0489

†Odds ratio of dead patients to survived patients in relation to the presence or absence of liver atrophy and a unit increase of each continuous parameter.
ALB, albumin; D/T ratio, ratio of direct to total bilirubin concentration; HGF, hepatocyte growth factor; O-C, the interval between hepatitis onset and the development of hepatic encephalopathy; PLT, platelet; PT, prothrombin time; TB, total bilirubin.

Table 6 Scores for Predictive Variables Affecting the Mortality of Patients with Fulminant Hepatitis and late onset hepatic failure (LOHF)

Score	0	1	2
O-C (days)	≤5	6–10	11≤
PT (%)	20<	5< ≤20	≤5
TB (mg/dL)	<10	10≤ <15	15≤
D/T ratio	0.7≤	0.5≤ <0.7	<0.5
PLT (104/μL)	10<	5< ≤10	≤5
Liver atrophy	Absent	Present	

D/T ratio, ratio of direct to total bilirubin concentrations; O-C, the interval between hepatitis onset and the development of hepatic encephalopathy; PLT, platelet; PT, prothrombin time; TB, total bilirubin.

Scoring system to predict the possible outcome of patients with fulminant hepatitis and LOHF

Variables extracted through ROC curve analysis were graded as shown in Table 6, according to the inflection points of each curve. The interval between hepatitis onset and the development of hepatic encephalopathy, prothrombin time, serum concentration of total bilirubin, the ratio of direct to total bilirubin concentration and peripheral platelet count were classified into three grades (0, 1 and 2), and liver atrophy into two grades (0 and 1).

As shown in Table 7, the mortality rates rose in relation to total scores calculated in patients seen between 1998 and 2003 (estimation cohort). When the predictive outcome of patients showing total scores of 5 or more was judged as "death", PPV and NPV of the system were 0.84 and 0.70, respectively (Table 8), suggesting that total scores of 5 is sufficient enough as a cut-off value that can discriminate between dead and survived patients. The scoring system with such cut-off value showed sensitivity and specificity of 0.80 and 0.76, respectively, and resulted in predictive accuracy of 0.78 in patients in the estimation cohort. Predictive accuracies did not differ depending on the disease types; 0.75 in patients with acute type of fulminant hepatitis and 0.87 in those with subacute type of fulminant hepatitis.

Table 7 The outcome of patients with fulminant hepatitis and late onset hepatic failure (LOHF) seen between 1998 and 2003 (estimation cohort) depending on total scores calculated through established scoring system

Total scores	Mortality (%)	FHA/FHS/LOHF
9≤	**9/10 (90.0%)**	**2/4/4**
8	**26/27 (96.3%)**	**2/20/5**
7	**42/46 (91.3%)**	**10/30/6**
6	**71/83 (85.5%)**	**20/52/11**
5	**59/80 (73.8%)**	**26/42/12**
4	31/55 (56.3%)	32/22/1
3	12/50 (24.0%)	44/4/2
2	8/40 (20.0%)	35/5/0
1	2/25 (8.0%)	25/0/0
0	0/5 (0.0%)	5/0/0

FHA, acute type of fulminant hepatitis; FHS, subacute type of fulminant hepatitis.

Table 8 Accuracies of established scoring system in patients with fulminant hepatitis and late onset hepatic failure (LOHF) seen between 1998 and 2003 (estimation cohort) when predictive outcome of patients showing total scores of 5 or more are diagnosed as "death"

	Total scores		
	≥5	<5	Total
Number of patients			
Dead patients	207	53	260
Surviving patients	39	122	161
Total	246	175	421
Mortality	84.1%	30.3%	61.8%
The accuracies			
Positive predictive value (PPV)	207/246		0.84
Negative predictive value (NPV)	122/175		0.70
Sensitivity	207/260		0.80
Specificity	122/161		0.76
Predictive accuracy (PA)	(207+122)/421		0.78

The accuracies of the established scoring system were validated in patients with fulminant hepatitis and LOHF seen between 2004 and 2008 (validation cohort). As shown in Table 9, the mortality rate of patients in each total score was almost equivalent to that obtained in analysis with patients in the estimation cohort. Thus, predictive accuracy through analysis in the validation cohort was 0.75 with sensitivity, specificity, PPV and NPV of 0.75, 0.80, 0.86 and 0.65, respectively (Table 10).

DISCUSSION

LIVER TRANSPLANTATION IS regarded worldwide as the most effective therapeutic procedure for patients with end-stage liver diseases including acute liver

Table 9 The outcome of patients with fulminant hepatitis and late onset hepatic failure (LOHF) seen between 2004 and 2008 (validation cohort) depending on total scores calculated through established scoring system

Total scores	Mortality (%)	FHA/FHS/LOHF
9≤	4/4 (100.0%)	0/3/1
8	9/9 (100.0%)	1/6/2
7	26/30 (86.7%)	9/20/1
6	35/39 (89.7%)	9/29/1
5	33/42 (78.6%)	18/19/5
4	21/39 (53.8%)	28/10/1
3	10/30 (33.3%)	22/8/0
2	4/26 (15.4%)	26/0/0
1	2/8 (25.0%)	8/0/0
0	0/4 (0.0%)	4/0/0

FHA, acute type of fulminant hepatitis; FHS, subacute type of fulminant hepatitis.

failure. Japanese Society for the Study of Liver Transplantation revealed that survival rate at 1 year after liver transplantation was 72.7% in patients with acute liver failure,[7] while conventional medical care yielded insufficient prognosis in such patients; survival rates were 54.0% and 24.0%, respectively, in patients with fulminant hepatitis of acute and subacute types and 15.0% in LOHF patients according to the nationwide survey by the Study Group of Intractable Hepatobiliary Diseases.[8] In general, in Japan, patients with acute liver failure visit clinics or hospitals at the onset of hepatitis symptoms and derangement of liver function was diagnosed by physicians specialized in general medicine. Next, the patients were transferred to hospitals with specialists in the fields of hepatology and emergency medicine around the periods of the development of hepatic encephalopathy. Conventional medical care including artificial liver support with plasma exchange and hemodiafiltration was performed, and then the patients were introduced to transplant surgeons regarding the indication of liver transplantation. Thus, the simple criteria to predict the outcome of patients with fulminant hepatitis and LOHF with sufficient accuracies are required to facilitate communication among general physicians, hepatologists and transplant surgeons.

Table 10 Accuracies of established scoring system in patients with fulminant hepatitis and late onset hepatic failure (LOHF) seen between 2004 and 2008 (validation cohort) when predictive outcome of patients showing total scores of 5 or more are diagnosed as "death"

	Total scores		
	≥5	<5	Total
Number of patients			
Dead patients	107	17	124
Surviving patients	37	70	107
Total	144	87	231
Mortality	74.3%	19.5%	53.7%
The accuracies			
Positive predictive value (PPV)	107/144		0.75
Negative predictive value (NPV)	70/87		0.80
Sensitivity	107/124		0.86
Specificity	70/107		0.65
Predictive accuracy (PA)	(107+70)/231		0.77

In Europe and the United States, the indication of liver transplantation in patients with acute liver failure has been determined according to the guideline proposed by King's Collage Hospital[9] and Beaujon Hospital.[10] In addition, a scoring system of model for end-stage liver disease (MELD), initially designed for patients with chronic liver failure, has recently been applied also to those with acute liver failure.[11] However, these guidelines are not directly applicable to patients with fulminant hepatitis and LOHF in Japan, since social environment as well as demographic and clinical features of the patients differ among Japan, Europe and the United States; for example liver transplantation with brain death-related donor is hardly available and artificial liver support is routinely performed in Japan. Thus, novel guidelines should be established for Japanese patients with fulminant hepatitis and LOHF instead of the previous guideline proposed by the Acute Liver Failure Study Group in Japan at 1996,[4] which shows decline of predictive accuracy when applied to recent patients.[5]

In the present paper, a novel scoring system to predict the outcome of patients with fulminant hepatitis and LOHF was established based on demographic and clinical features of patients seen between 1998 and 2008. The predictive mortality rates were estimated through six variables at the occurrence of grade 2 or more severe hepatic encephalopathy; the interval between the onset of hepatitis symptoms and the development of hepatic encephalopathy, prothrombin time, serum concentration of total bilirubin, the ratio of direct to total bilirubin concentration, peripheral platelet count and presence of liver atrophy on imaging. When total scores were calculated through six variables in patients belonging to the estimation cohort, the mortality rate was 84.1% in those with scores of 5 or more, while it was 30.3% in those with scores of 4 or less. Thus, the cut-off value of total scores to discriminate possible dead

patients from surviving patients was set between 4 and 5. Consequently, excellent accuracies were obtained through analysis in patients belonging to the estimation cohort; predictive accuracy was 0.78 with either of PPV, NPV, sensitivity and specificity greater than 0.7. Such high predictive accuracy was also found through analysis in patients belonging to the validation cohort. It is noteworthy that peripheral platelet count and the presence of liver atrophy were added to the list of predictive variables in the present scoring system. Also, cut-off values to grade other variables, such as the interval between the onset of hepatitis symptoms and the development of hepatic encephalopathy, differ between the present system (Table 6) and previous guidelines (Table 1). These modifications may contribute to improve predictive accuracy of the novel scoring system when applied to recent patients.

In the present study, the age of patients was excluded from the list of predictive variables to facilitate the use of the system in pediatric patients. In our database, a patient showing total score of 6 died, while three cases with total scores of 4 or less survived, when the system was applied for patients aged less than 15 years old. Furthermore, the most recent report by Fujisawa showed 100% specificity and PPV by the scoring system in 40 pediatric patients.[12] Thus, the system seems to be useful even in such patients. Also, plasma HGF concentration was deleted from the list of predictive variables, because it is difficult to obtain the results within a day in most of the hospitals in Japan. In contrast, the presence of liver atrophy was included in the predictive variable list, but the quantitative criteria for liver atrophy were not specified in the present scoring system. The estimated liver volume is measured on CT examination, and the ratio of the value to the standardized liver volume was reported to correlate with mortality in patients with acute liver failure in Japan.[13] These problems, regarding age of patients, significance of plasma HGF concentration and diagnostic criteria to determine liver atrophy should be further investigated.

In conclusion, a novel scoring system for predicting outcome of patients with fulminant hepatitis and LOHF was established. This system may be useful to determine the indication of liver transplantation in patients with acute liver failure, since the system showed high predictive accuracies even after the validation.

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Special Report

Guideline on the use of new anticancer drugs for the treatment of Hepatocellular Carcinoma 2010 update

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The "*Guideline on the Use of New Anticancer Drugs for the Treatment of Hepatocellular Carcinoma*" was prepared by the Study Group on New Liver Cancer Therapies established by the "*Research Project on Emergency Measures to Overcome Hepatitis*" under the auspices of the Health and Labour Sciences Research Grant. The Guideline brings together data collected by the Study Group on the use and incidence of adverse events in 264 patients with advanced hepatocellular carcinoma (HCC) treated using sorafenib and in 535 patients with advanced HCC treated using miriplatin at 16 participating institutions up until 22 December 2010, as well as referring to the published studies, academic presentations, and reports from the private sector. The aim of this Guideline is to facilitate understanding and current thinking regarding the proper usage of new anticancer drugs towards actual use in therapy. In terms of the format, the Guideline presents "clinical questions" on issues pertaining to medical care, makes "recommendations" on diagnosis and treatment in response to each of these clinical questions, and provides a rationale for these recommendations in the form of "scientific statements".

Key words: hepatic arterial infusion, hepatocellular carcinoma, miriplatin, molecular targeting therapy, sorafenib

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INTRODUCTION

THE MOLECULAR-TARGETED agent sorafenib has been found to significantly prolong survival in patients with hepatocellular carcinoma (HCC).[1,2] In May 2009, sorafenib was approved in Japan for unresectable

HCC. Furthermore, miriplatin was approved in Japan for the treatment of HCC in January 2010, and clinical trials are also currently underway on a number of other promising new anticancer agents. Treatment of HCC is thus undergoing a period of major transition, but the role of these anticancer drugs and conventional therapies remains unclear, leading to concerns about the risk of serious adverse events (SAEs).

The Study Group on New Liver Cancer Therapies (the Study Group) was formed as part of the "Research Project on Emergency Measures to Overcome Hepatitis" sponsored by the Health and Labour Sciences Research Grant, with the overall purpose of formulating a guideline to facilitate understanding on the practical usage of new anticancer drugs.

The Study Group collected information on the use of new anticancer drugs, sorafenib and miriplatin at 16 affiliated institutions and compiled current opinions regarding the proper use of these drugs based on published studies, academic conference papers and reports from the private sector. These results have now been compiled in the form of a guideline.

However, of note is that this guideline is provisional and has been prepared to expedite the provision of proper information because information on these new anticancer drugs is constantly being updated.

STUDY METHODS, SUBJECTS AND PARTICIPATING INSTITUTIONS

Basic statistics

THE STUDY GROUP'S "New Liver Cancer Therapies" (NLCT) study was based on data from patients with advanced HCC treated using sorafenib or miriplatin up until 22 December 2010 at the participating institutions. Clinical data were recorded by each institution in case report files (CRFs) created by the Study Group. Of the patients enrolled in this study, 264 were treated with sorafenib and 535 were treated with miriplatin. Any input variables that were unclear were excluded from the analyzed data. After analyzing collecting data on the use of these drugs, the Study Group compiled current opinions on proper use based on published papers, academic conference papers and reports from the private sector. The Study Group proposed a series of "clinical questions" (CQ) on issues pertaining to practical medical care and summarized the current evidence in response to each of these CQ in the form of "scientific statements", as well as making "recommendations".

Participating institutions

The 16 institutions that participated in this study were: Kinki University; Chiba University; Yamaguchi University; Kurume University; Kyorin University; Showa University; Ehime University; Okayama University; Kyoundo Hospital; Tohoku University; Osaka University; Gifu University; Hyogo College of Medicine; Toranomon Hospital; Saitama Medical University; and Kanazawa University.

RESULTS

Sorafenib therapy

Indications

CQ1-1 For which patients with HCC is sorafenib therapy indicated?

Recommendation Sorafenib therapy is indicated in HCC patients with good performance status (PS) and Child–Pugh class A for whom surgical resection, local ablation therapy (LAT), and transcatheter arterial chemoembolization (TACE) are not possible or not indicated.

The safety and efficacy of sorafenib has not been established in Child–Pugh class B/C patients.

Furthermore, the usefulness of sorafenib as adjuvant chemotherapy after resection, LAT, or TACE of HCC has not been demonstrated.

Scientific statement Two randomized, placebo-controlled trials demonstrating the usefulness of sorafenib were conducted on patients in whom surgical resection, LAT and TACE were not indicated or who were unresponsive to TACE.[1,2]

The Japan Society of Hepatology provides the following definitions for impossible and refractory cases to TACE.[3]

Definition of "Impossible cases to TACE"

1 Deterioration of treated vessel resulting in inability to select catheter for insertion into the nutrient vessel;
2 Deterioration in hepatic function to Child–Pugh class C due to repeated treatment;
3 Patients with tumor thrombus in main trunk or first branch of portal vein;
4 Patients with large arterio-portal shunts.

Definition of "Refractory cases to TACE"

(1) Intrahepatic lesion(s)
  (i) Poor Lipiodol deposits (≤50%) observed on at least two consecutive occasions in computed tomography (CT) assessment of therapeutic response immediately after (>1 month) correctly performing TACE;

(ii) Multiple new lesions observed on at least two consecutive occasions in CT assessment of therapeutic response immediately after (>1 month) TACE;
(2) Appearance of vascular invasion;
(3) Appearance of distant metastasis;
(4) Tumor markers.
(i) Continued increase in tumor markers with transient decrease only immediately after TACE procedure.

In the present NLCT study, as many as 91% of patients underwent prior treatment, in whom 29% received hepatic arterial infusion chemotherapy (HAIC). Comparison of the characteristics of the remaining NLCT study patients with those of previous clinical trials[1,2,4–6] is presented in Table 1.

An adverse event (AE) report on all-patient special drug use surveillance (SDUS) conducted in Japan[7] contains analysis and reporting of AEs for 777 patients for whom CRFs were collected up until 19 December 2009.

That report compared the clinical characteristics for 51 of these 777 patients who died within 30 days of treatment ("early death group") and the 382 patients who survived for ≥61 days ("control survival group"). The data indicate that the prevalence of Eastern Cooperative Oncology Group (ECOG) PS grades ≥2 tended to be high among patients in the "early death group" at 5.9% compared with those in the "control survival group" at 0.5%, suggesting the need to carefully follow the course of patients with poor PS. In the NLCT study, 98% of patients had a PS score of 0–1.

In terms of hepatic function, two randomized, placebo-controlled trials demonstrating the usefulness of sorafenib were conducted on Child–Pugh class A patients.[1,2]

Meanwhile, in the NLCT study, 81% of evaluable patients were Child–Pugh class A, and 94% had a Child–Pugh score of ≤7. Comparison of treatment results of Child–Pugh class A and B patients did not reveal any difference in tumor control rates (46% vs. 50%; $P = 0.52$), but overall survival (OS) was inferior in Child–Pugh class B patients (median OS: 11.5 months vs. 5.2 months; $P < 0.01$).

In a Phase I trial conducted in Japan, no clear increase in toxicity was observed in Child–Pugh class B patients compared with Child–Pugh class A patients.[8] On the other hand, the aforementioned SDUS found that hepatic functional reserve was poor in the "early death group" compared to the "control survival group".[4]

A Phase II study of sorafenib therapy in HCC patients including those with Child–Pugh class B is currently underway in Japan (UMIN [University Hospital Medical Information Network] 000002972). Another study currently being conducted worldwide is the Global Investigation of therapeutic decisions in HCC and of its treatment with sorafenib (GIDEON); a large-scale prospective study on actual sorafenib therapy of patients with unresectable HCC. The GIDEON study is recruiting 3000 patients from over 400 sites in more than 40 countries in the Asia-Pacific region, Europe, USA, Latin America, and Japan.[9] The study's first interim analysis has been released and the findings of 511 recruited patients including those in Child–Pugh class B have been examined. No significant difference in grade 3 or 4 AEs was found to exist between Child–Pugh class A and B patients, at 31% and 38%, respectively.[10] Future GIDEON study analyses are expected to provide crucial information concerning the safety of sorafenib for Child–Pugh class B patients.

A Phase III study of post-TACE adjuvant sorafenib chemotherapy versus placebo conducted in Japan and South Korea failed to demonstrate the usefulness of sorafenib administration.[11] In addition, a Phase III placebo-controlled trial of adjuvant sorafenib chemotherapy following radical treatment (either surgical resection or LAT) of HCC (STORM Trial) is currently underway.[12]

The NLCT study did not include any patients treated with sorafenib as adjuvant chemotherapy.

Method of administration

CQ1-2 What is the optimal dosage regimen for sorafenib therapy?

Recommendation The standard dosage regimen for sorafenib therapy is 400 mg administered twice daily (800 mg/day).

The safety and efficacy of sorafenib therapy in combination with other anti-neoplastic agents or TACE have not been established.

Scientific statement In the two aforementioned randomized, placebo-controlled trials demonstrating the usefulness of sorafenib, a single 400 mg dose of sorafenib was administered twice daily (800 mg/day),[1,2] and usefulness was not observed at a reduced dosage. A high-fat diet reportedly lowers the plasma concentration of sorafenib so administration should be avoided from 1 h before to 2 h after meals.

Reduced dose regimen due to AEs was conducted in the abovementioned studies as follows:
Step-down dose (step 1): 400 mg once a day
Step-down dose (step 2): 400 mg every another day

Table 1 Characteristics of patients receiving sorafenib therapy

	NLCT Study ($n = 264$) %	SDUS[4,6] ($n = 777$) %	SHARP Trial[1] ($n = 299$) %	Asia-Pacific Trial[2] ($n = 150$) %	Sorafenib phase II[5] ($n = 137$) %
Age (years)					
Median	70		64.9 ± 11.2	51	69
Range	33–87		(mean ± SD)	23–86	28–86
Gender					
Male	79		87	84.7	71
PS					
0	83	69.5	54	25.3	50
1	15	26.5	38	69.3	50
Child–Pugh class					
A	81	88.2	95	97.3	72
B	19	9.9	5	2.7	28
HBs antigen					
Positive	20	24.6	19	70.7	17
HCV antibody					
Positive	62	52.2	29	10.7	48
Prior treatment					
Yes	91	91.2	49		
Resection	31		19		
LAT	47		15		
TACE	78		29		
HAIC	29				
Advanced vascular invasion					
Yes	18		36	36.0	
Extrapulmonary lesion(s)					
Yes	51	54.4	53	68.7	–
Lymph node(s)	22	15.4	30	52	–
Lung(s)	26	30.6	22	30.7	–
Maximum tumor size (mm)	34				
Range	7–170				
≥30 mm	59				
Stage	†	‡	§	§	‡
I	1	1.2			0
II	9	4.8			3
III	30	20.7	B: 18	B: NE	31
IV A	17	23	C: 82	C: 95.3	66
IV B	43	47.6			
T-Bil (mg/dL)					
Median	0.8		0.7		
Range	0–7.7		0.1–16.4		
Alb (g/dL)					
Median	3.5		3.9		
Range	1.7–4.8		2.7–5.3		
AFP (ng/mL)					
Median	218		44.3		
Range	0.8–252150		0–2080000		
≥10	84			77.3	76

†Japanese Classification of Liver Cancer.
‡UICC classification.
§BCLC classification.
AFP, α fetoprotein; Alb, albumin; HAIC, hepatic arterial infusion chemotherapy; HBs, Hepatitis B surface antigen; HCV, hepatitis C virus; LAT, local ablation therapy; NLCT, New Liver Cancer Therapies; PS, performance status; SD, standard deviation; SDUS, special drug use surveillance; SHARP, sorafenib hepatocellular carcinoma assessment randomized protocol; TACE, transcatheter arterial chemoembolization; T-Bil, total bilirubin.

In the NLCT study, 77% of patients received the standard dosage regimen of 400 mg twice daily, while 21% were started on a reduced dose.

Comparison of the group started on the standard dose of 800 mg/day and the group started on a reduced dose did not reveal any significant differences in either duration of treatment (117 days vs. 81 days; $P = 0.05$) or number of dosing days (107 days vs. 78 days; $P = 0.10$). Furthermore, dosage was subsequently increased in 22% of the reduced initial dose group. Daily dosage intensity (DI) was 615 mg in the standard-dose group and 387 mg in the reduced-dose group.

It is conceivable to start sorafenib therapy at a reduced dose according to the condition of the patient or prevention of AEs. Because efficacy at reduced doses has not been demonstrated, as long as no AEs are encountered in the course of treatment, consideration should be given to increasing the dose to the standard dosage regimen.

With regard to sorafenib combination therapies, Phase I and Phase II studies on systemic chemotherapy in combination with sorafenib therapy have been published for radiotherapy,[13,14] doxorubicin,[15] tegafur/uracil,[16] and octreotide.[17] Several Japanese clinical trials are also being conducted on combination therapy, specifically low-dose cisplatin/fluorouracil HAIC (UMIN000004315), cisplatin HAIC (UMIN000001496), and S-1 chemotherapy (UMIN000002418, UMIN000002590). Therapies combining sorafenib with other anti-neoplastic agents are therefore still in the research stage, and their efficacy is yet to be demonstrated.

In terms of sorafenib combined with LAT, a Phase III placebo-controlled trial of adjuvant sorafenib chemotherapy following radical treatment (surgical resection or LAT) of HCC (STORM Trial) is presently underway.[12] Meanwhile, sorafenib combined with TACE has been investigated in a Phase III study of post-TACE adjuvant sorafenib chemotherapy versus placebo conducted in Japan and South Korea, but the study failed to demonstrate the usefulness of sorafenib administration.[11] Another Phase II trial on TACE in combination with sorafenib is presently being carried out in Japan (TACTICS; UMIN 000004316).

Discontinuation criteria

CQ1-3 How and when should sorafenib therapy be discontinued?

Recommendation Administration of sorafenib should be discontinued immediately in the event of SAEs.

Discontinuation should also be considered when disease progression is confirmed by radiological imaging or on the basis of patient symptoms.

Scientific statement In the two randomized, placebo-controlled trials demonstrating the usefulness of sorafenib therapy, administration was discontinued upon confirmation of radiologic or symptomatic progression or in the event of SAEs.[1,2]

In the NLCT study, sorafenib therapy was discontinued in 185 patients with 63% due to disease progression and 22% due to AEs. Moreover, 60% of discontinued patients did not undergo post-treatment.

No data are currently available on the efficacy/safety of continued administration of sorafenib after disease progression.

Adverse events

CQ1-4 What are the adverse events associated with sorafenib therapy?

Recommendation Some form of AE has appeared in almost all patients treated with sorafenib.

These AEs vary, and have even included serious adverse events (SAEs) resulting in death. Familiarity with these AEs is therefore essential, to carefully monitor patient progress while taking the necessary precautions, and to respond rapidly when an AE occurs.

The following AEs are known to occur frequently in patients treated with sorafenib.

1 Hand-foot skin reaction (HFSR);
2 Rash/desquamation;
3 Diarrhea;
4 Anorexia;
5 Hypertension;
6 Fatigue;
7 Alopecia;
8 Nausea.

While infrequent, life-threatening SAEs include hepatic failure, interstitial pneumonia, and gastrointestinal hemorrhage.

In addition, the following blood test abnormalities are known to occur frequently in patients treated with sorafenib.

1 Leukopenia;
2 Neutropenia;
3 Anemia;
4 Thrombocytopenia;
5 Hepatic impairment (elevated AST [aspartate aminotransferase], ALT [alanine aminotransferase], ALP [alkaline phosphatase], γ-GTP [γ-glutamyltransferase], T-Bil [total bilirubin]);