Risk factors of recurrence and poor survival in curatively resected hepatocellular carcinoma with microvascular invasion

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Advances in Clinical and Experimental Medicine, ISSN 1899–5276 (print), ISSN 2451–2680 (online) *Adv Clin Exp Med*. 2020;29(7):887–892

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Funding sources
None declared

Conflict of interest
None declared

Received on April 18, 2017
Reviewed on August 16, 2017
Accepted on September 1, 2017

Published online on August 4, 2020

Abstract

Background. Microvascular invasion (MVI) is a significant sign of the invasive property and a strong predictor of poor prognosis in hepatocellular carcinoma (HCC), a life-threatening malignancy. However, recurrence-associated and post-surgical long-term prognosis-associated factors in HCC with MVI remain unknown.

Objectives. To address the abovementioned issues, based on a Chinese patient cohort with HCC after curative hepatic resection.

Material and methods. The patient cohort consisted of 62 consecutive patients with HCC and MVI who underwent curative hepatic resection. The associations between clinicopathologic variables and recurrence, as well as patient overall/disease-free survival, were uni- and multivariately evaluated.

Results. Univariate χ^2 test identified hepatitis B surface antigen (HBsAg) positivity, high Edmondson–Steiner grade and male gender as risk factors of recurrence, whereas Edmondson–Steiner grade and HBsAg positivity were significant or marginally significant in the multivariate stepwise logistic regression analysis. Subsequently, univariate log-rank test showed that Edmondson–Steiner grade, HBsAg positivity and Child–Pugh grade were associated with overall and/or disease-free survival. Among them, the independent prognostic impact of Edmondson–Steiner grade and HBsAg positivity for both overall and disease-free survival were proven in the multivariate Cox regression analysis.

Conclusions. Our data suggested that Edmondson–Steiner grade and HBsAg positivity might serve as useful indicators of recurrence and pessimistic prognosis in HCC with MVI.

Key words: prognostic factor, recurrence, survival, hepatocellular carcinoma, hepatic resection

Cite as
Zhou L, Wang SB, Chen SG, Qu Q, Rui JA. Risk factors of recurrence and poor survival in curatively resected hepatocellular carcinoma with microvascular invasion. *Adv Clin Exp Med*. 2020;29(7):887–892. doi:10.17219/acem/76750

DOI
10.17219/acem/76750

Introduction

Hepatocellular carcinoma (HCC) is well-recognized as a malignant tumor with high incidence and mortality worldwide.[1–3] Therefore, identification of prognostic markers of HCC became a research hotspot. Except for some clinical and pathological variables, such as portal vein tumor thrombosis (PVTT), tumor size, alpha-fetoprotein (AFP), and Child–Pugh grade, that were previously summarized as the most robust predictors of death, microvascular invasion (MVI), one of signs of unfavorable histological features of HCC (especially the invasive property), was also revealed to be of strong predictive potential for poor prognosis in HCC after several treatment methods, such as resection, transplantation and ablation.[4–14] However, it was also reported that some patients who carried HCC with MVI acquired actual ten-year survival.[15] Therefore, there is an urgent need and particular interest for the identification of prognostic determinants in this highly invasive subtype of HCC.

The current investigation aimed to address the issues through identification of factors that were associated with recurrence and long-term survival in this subtype of HCC.

Material and methods

Patients

In total, 62 patients undergoing curative hepatic resection for HCC with MVI were included in the study. Their age ranged from 24 to 76 years (mean ± standard deviation (M ±SD) 56.8 ±12.5 years). Tumor sizes, regarding the largest dimensions, ranged from 2 cm to 26 cm (M ±SD 8.5 ±4.9 cm). Microvascular invasion was diagnosed through the postoperative routine pathological examinations. Portal vein tumor thrombosis was defined as tumor involvement in the main branches of portal vein. Satellite nodules were macroscopic. Histological grading was given based on Edmondson–Steiner criteria.[16] The baseline characteristics of patients are shown in Table 1. The acquisition of clinicopathologic and follow-up data was approved by the institutional Ethics Committee of Peking Union Medical College Hospital, Beijing, China.

Evaluated variables and endpoints

Twelve variables related to general situation of the patient (age and gender), hepatic background (hepatitis B surface antigen (HBsAg), hepatitis C virus (HCV), liver cirrhosis, and Child–Pugh grade) and tumor phenotypes (serum AFP value, tumor size, tumor number, PVTT, satellite nodule, and Edmondson–Steiner grade) were chosen for analyses. Post-surgical recurrence, as well as overall and disease-free survival, served as the endpoints. Disease-free survival was defined as the patient survival interval from surgery to recurrence or distant metastasis occurrence.

Table 1. Baseline characteristics of patients with hepatocellular carcinoma (HCC) and microvascular invasion (MVI)

Variables	n (%)
Age	M ±SD: 56.8 ±12.5 years; range: 24–76 years
Gender	
male	53 (85.5)
female	9 (14.5)
HBsAg	
positive	52 (83.9)
negative	10 (16.1)
HCV	
positive	5 (8.1)
negative	57 (91.9)
Cirrhosis	
present	51 (82.3)
absent	11 (17.7)
Child–Pugh grade	
grade A	58 (93.5)
grade B	4 (6.5)
Tumor size	M ±SD: 8.5 ±4.9 cm; range: 2–26 cm
PVTT	
present	19 (30.6)
absent	43 (69.4)
Satellite nodule	
present	9 (14.5)
absent	53 (85.5)
Tumor number	
solitary	55 (88.7)
multiple	7 (11.3)
AFP level	
>20 ng/mL	49 (79.0)
≤20 ng/mL	13 (21.0)
Edmondson–Steiner grade	
I	3 (4.8)
II	14 (22.6)
III	29 (46.8)
IV	16 (25.8)

M – mean; SD – standard deviation; HBsAg – hepatitis B surface antigen; HCV – hepatitis C virus; PVTT – portal vein tumor thrombosis; AFP – alpha-fetoprotein.

Follow-up

The follow-up for all patients, ranging from 3 to 108 (Me: 14) months, was performed through imaging examinations and serum AFP level detection, as previously reported.[17,18] Follow-up intervals ranged from 1 to 3 months.

Statistical analyses

The uni- and multivariate risk factors of recurrence were identified using χ^2 and stepwise logistic regression tests. Survival analyses were adopted using Kaplan–Meier method and log-rank test. Cox regression (proportional hazard model) was used for multivariate analysis of prognostic factors. Statistical software package SPSS v. 11.5 (SPSS Inc., Chicago, USA) was applied for all the analyses. Statistical significance was indicated when a p-value was less than 0.05.

Results

Risk factors of recurrence in patients with hepatocellular carcinoma and microvascular invasion after curative hepatic resection

Using univariate χ^2 test, gender, HBsAg and Edmondson–Steiner grade were associated with postoperative recurrence ($p < 0.05$; Table 2), but other parameters were not of significance ($p > 0.05$; Table 2). Multivariate stepwise logistic regression showed that Edmondson–Steiner grade was the single independent risk factor of recurrence (hazard ratio (HR) = 6.374, 95% confidence interval (95% CI) = 1.196–33.963, p = 0.030; Table 2), while HBsAg positivity was of marginal significance (HR = 4.933, 95% CI = 0.789–30.833, p = 0.088; Table 2).

Prognostic factors in patients with hepatocellular carcinoma and microvascular invasion after curative hepatic resection

Univariate log-rank test revealed that HBsAg and Edmondson–Steiner grade were significantly associated with overall survival ($p < 0.05$; Fig. 1 and Table 3), whereas Edmondson–Steiner grade, HBsAg and Child–Pugh grade were significant for disease-free survival ($p < 0.05$; Fig. 2 and Table 3). In multivariate Cox regression analyses, Edmondson–Steiner grade and HBsAg were identified as independent prognostic indicators for both overall and disease-free survival (HR = 2.905 and 2.942, 95% CI = 1.263–6.683 and 1.021–8.474, p = 0.012 and 0.046 for overall survival, respectively; HR = 3.407 and 2.944, 95% CI = 1.577–7.360 and 1.124–7.710, p = 0.002 and 0.028 for disease-free survival, respectively; Table 4).

Table 2. Univariate and multivariate factors associated with recurrence in HCC with MVI

Variables	n	Univariate analysis			Multivariate analysis		
		with recurrence	without recurrence	p-value	HR	95% CI	p-value
Age				0.147			
≥65 years	19	14	5		N/A	N/A	N/A
<65 years	43	38	5				
Gender				**0.045#**			0.118
male	53	47	6		4.780	0.671–34.066	
female	9	5	4		1		
HBsAg				**0.001**			0.088
positive	52	47	5		4.933	0.789–30.833	
negative	10	5	5		1		
HCV				1.000#			
positive	5	4	1		N/A	N/A	N/A
negative	57	48	9				
Cirrhosis				0.119#			
present	51	45	6		N/A	N/A	N/A
absent	11	7	4				
Child–Pugh grade				0.838#			
grade A	58	48	10		N/A	N/A	N/A
grade B	4	4	0				
Tumor size				0.962#			
≥5 cm	53	45	8		N/A	N/A	N/A
<5cm	9	7	2				
PVTT				0.672#			
present	19	17	2		N/A	N/A	N/A
absent	43	35	8				
Satellite nodule				1.000#			
present	9	8	1		N/A	N/A	N/A
absent	53	44	9				
Tumor number				0.493#			
solitary	55	45	10		N/A	N/A	N/A
multiple	7	7	0				
AFP level				0.732#			
>20 ng/mL	49	42	7		N/A	N/A	N/A
≤20 ng/mL	13	10	3				
Edmondson–Steiner grade				**0.004#**			**0.030**
I–II	17	10	7		1		
III–IV	45	42	3		6.374	1.196–33.963	

HR – hazard ratio; 95% CI – 95% confidence interval; N/A – not applicable; # χ^2 test with continuity correction; values in bold indicate statistically significant differences.

Table 3. Univariate analysis for overall and disease-free survival of HCC with MVI patients

Variables	n	Overall survival			Disease-free survival		
		M ±SE	95% CI	p-value	M ±SE	95% CI	p-value
Age				0.408			0.250
≥65 years	19	40 ±10	20–60		34 ±10	14–53	
<65 years	43	24 ±3	18–30		16 ±2	12–19	
Gender				0.772			0.213
male	53	28 ±5	19–37		19 ±4	12–27	
female	9	22 ±4	14–30		22 ±5	13–31	
HBsAg				**0.006**			**0.002**
positive	52	22 ±3	16–27		14 ±2	11–17	
negative	10	69 ±15	40–97		59 ±15	29–89	
HCV				0.063			0.167
positive	5	12 ±3	6–18		9 ±2	4–14	
negative	57	30 ±5	20–39		22 ±4	14–30	
Cirrhosis				0.286			0.080
present	51	27 ±5	18–36		18 ±3	11–24	
absent	11	27 ±4	19–36		22 ±4	14–30	
Child–Pugh grade				0.058			**0.017**
grade A	58	30 ±5	20–39		22 ±4	14–31	
grade B	4	11 ±5	1–21		7 ±3	1–12	
Tumor size				0.101			0.147
≥5 cm	53	27 ±5	18–37		21 ±4	12–29	
<5cm	37	74 ±8	58–90		21 ±3	15–27	
PVTT				0.095			0.095
present	19	20 ±5	11–30		13 ±3	7–19	
absent	43	34 ±6	21–46		26 ±5	16–37	
Satellite nodule				0.051			0.142
present	9	16 ±5	7–25		11 ±3	5–18	
absent	53	31 ±5	21–41		23 ±4	12–32	
Tumor number				0.203			0.337
solitary	55	31 ±5	20–41		23 ±4	14–32	
multiple	7	17 ±5	7–28		12 ±4	5–20	
AFP level				0.517			0.260
>20 ng/mL	49	26 ±4	17–34		19 ±4	11–26	
≤20 ng/mL	13	31 ±8	16–46		22 ±5	12–32	
Edmondson–Steiner grade				**0.001**			**<0.001**
I–II	17	56 ±13	31–80		46 ±12	24–69	
III–IV	45	19 ±2	15–23		12 ±1	10–14	

M – mean; SE – standard error; values in bold indicate statistically significant differences.

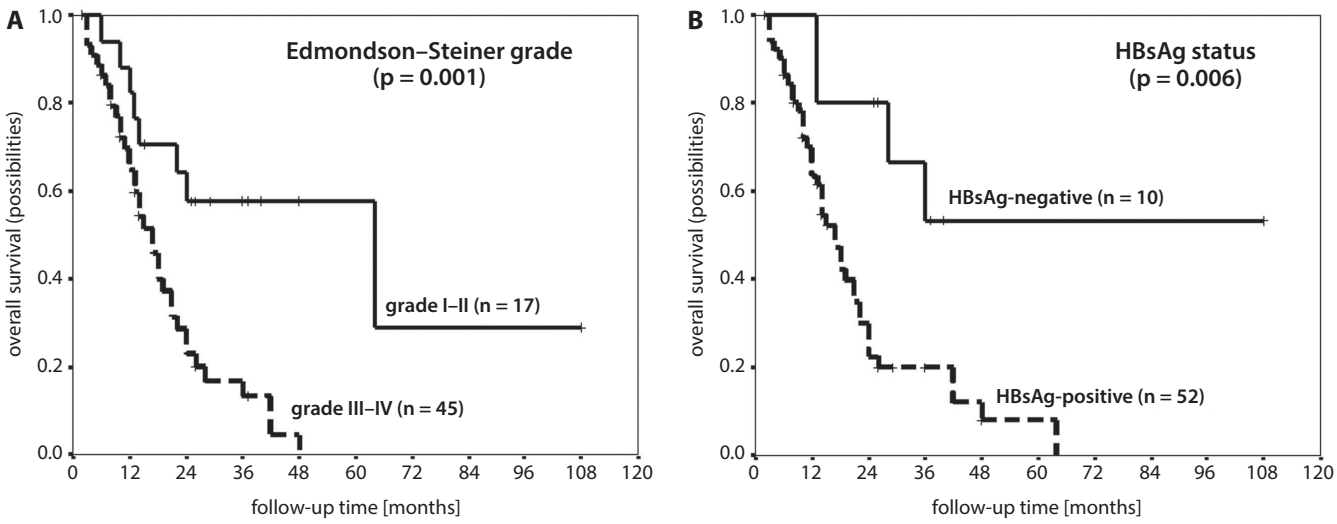

Fig. 1. The significant influence of the variables on overall survival of patients with HCC and MVI after curative hepatic resection. A. Edmondson–Steiner grade (solid line – grade I–II, n = 17; dashed line – grade III–IV, n = 45; p = 0.001); B. HBsAg positivity (solid line – negative, n = 10; dashed line – positive, n = 52; p = 0.006)

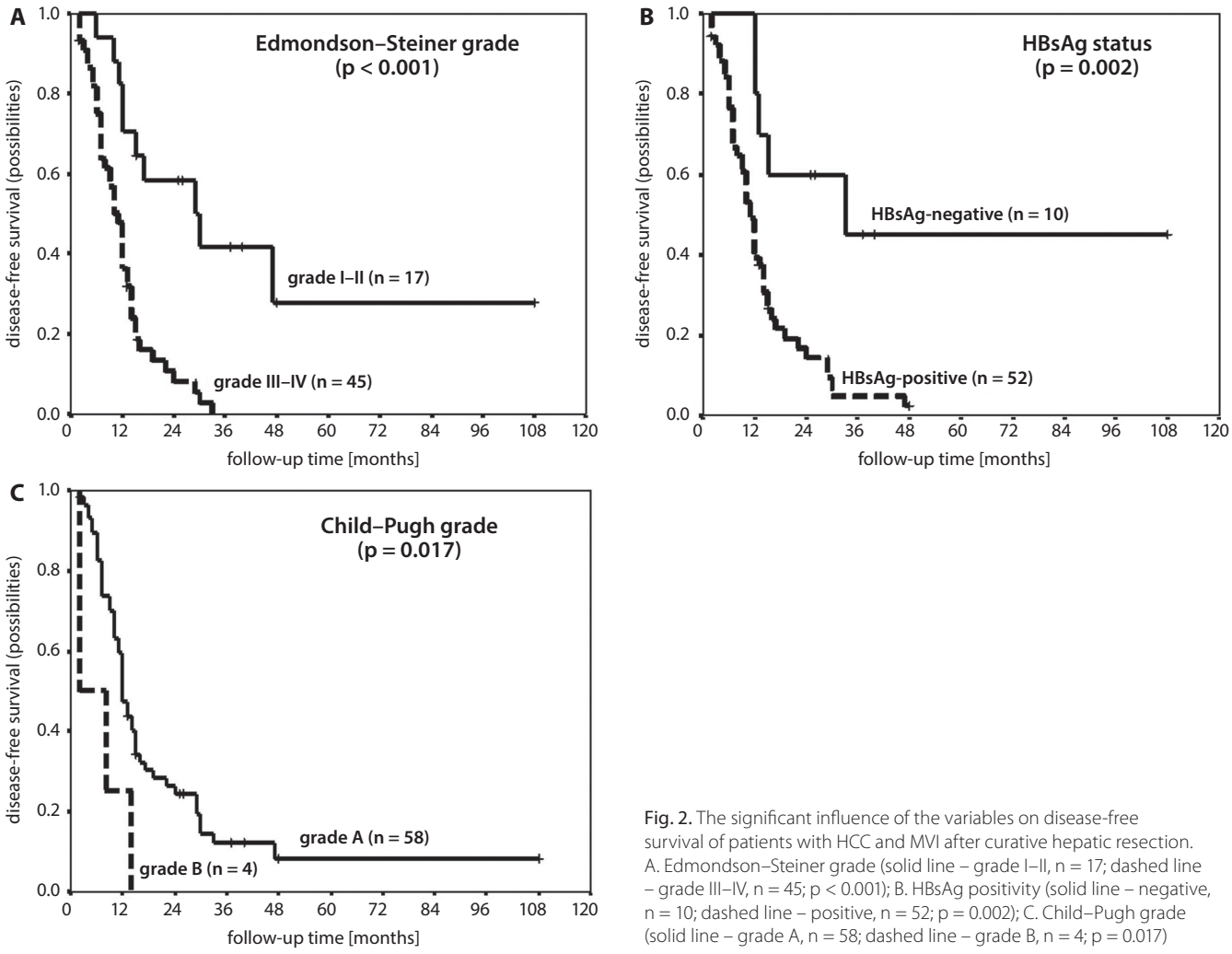

Fig. 2. The significant influence of the variables on disease-free survival of patients with HCC and MVI after curative hepatic resection. A. Edmondson–Steiner grade (solid line – grade I–II, n = 17; dashed line – grade III–IV, n = 45; p < 0.001); B. HBsAg positivity (solid line – negative, n = 10; dashed line – positive, n = 52; p = 0.002); C. Child–Pugh grade (solid line – grade A, n = 58; dashed line – grade B, n = 4; p = 0.017)

Table 4. Multivariate analysis for overall and disease-free survival of HCC with MVI

Variables	Overall survival			Disease-free survival		
	HR	95% CI	p-value	HR	95% CI	p-value
Edmondson–Steiner grade	2.905	1.263–6.683	**0.012**	3.407	1.577–7.360	**0.002**
HBsAg	2.942	1.021–8.474	**0.046**	2.944	1.124–7.710	**0.028**
Child–Pugh grade	N/A	N/A	N/A	2.043	0.718–5.813	0.181

N/A – not applicable; values in bold indicate statistically significant differences.

Discussion

Microvascular invasion, as a marker of the invasive growth, remarkably influences prognosis of HCC in patients who underwent curative treatments, liver resection and transplantation, when recurrence and/or overall as well as disease-free survival served as endpoints.[5–14] However, prognostic factors of HCC with MVI remain to be explored. In the present study, factors associated with tumor recurrence were first identified. It was found that gender, Edmondson–Steiner grade and HBsAg were of statistical significance in univariate χ^2 test, while these 3 variables were significant or marginally significant in multivariate logistic regression, respectively. Previously, gender and Edmondson–Steiner grade, along with MVI, were suggested to be correlated with post-surgical recurrence of HCC.[19,20] This study first showed their roles in HCC with MVI. Moreover, the more important finding of this work is to define HBsAg as a potential risk factor of recurrence in HCC with MVI, based on univariate and multivariate analyses. It was shown that high hepatitis B viral load predicted the recurrence of small HCC after curative resection.[20,21] We provided 2 novel predictors, HBsAg, which is routinely detected, and gender that was not predictive for recurrence in MVI-absent HCC, in addition to Edmondson–Steiner grade.[22] Some authors found

the association between HBsAg positivity and high AFP level, whereas AFP is reported to be involved in many malignant phenotypes of hepatoma cells, even in the activity of dendritic cells, through different mechanisms.[23–25] Furthermore, the finding that knockdown of HBsAg expression inhibits HCC growth provides the direct evidence for the role of HBsAg.[26] Therefore, our results about HBsAg and tumor recurrence might have a molecular basis.

On the other hand, it is well-known that HCC carries unsatisfactory prognosis. Thus far, its prognostic indicators, including MVI, have been identified.[4–14] However, prognostic indicators in HCC with MVI remain unclear and need in-depth exploration, because different survival status was also found in patients who carried HCC with MVI.[15] Our results showed that HBsAg and Edmondson–Steiner grade univariately and multivariately predicted both overall and disease-free survival, in consistence with reports on other types of HCC.[27,28] Considering the fact that these variables were independent or marginally independent risk factors for recurrence, their impact on patient survival of HCC with MVI might be understandable. Previously, the impact of HBsAg positivity on prognosis of HCC after radical or palliative therapies was also suggested.[27] This study first links HBsAg and HCC with MVI. Here, the authors preliminarily speculate on the possible route of its action in this subtype of HCC, as this protein stimulates tumor growth, then facilitates recurrence, and finally causes worse prognosis. Of course, this hypothesis needs to be extensively validated.

Conclusions

Our data suggests that Edmondson–Steiner grade and HBsAg positivity might function as significant predictors for recurrence and poor prognosis in HCC with MVI after curative resection.

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