rates indicated that the presence of non-hypervascular hypointense nodules detected during the hepatobiliary phase of Gd-EOB-DTPA-enhanced MRI is a risk factor for recurrence of HCC after hepatectomy. Although we did not find differences in the rate of intrahepatic metastasis recurrence according to non-hypervascular hypointense hepatic nodule status, we found a significantly higher rate of multicentric recurrence in patients with preoperative concurrent non-hypervascular hypointense hepatic nodules. In addition, the majority of multicentric recurrences involved the hypervascularization of non-hypervascular hypointense hepatic nodules observed preoperatively with Gd-EOB-DTPA-enhanced MRI. It is controversial whether all non-hypervascular hypointense nodules detected during the hepatobiliary phase of Gd-EOB-DTPA-enhanced MRI have the potential to progress to typical, hypervascular HCC. However, 26.5% of non-hypervascular hypointense nodules showed hypervascular spots with a long-term follow-up in our previous study [28]. In addition to the likelihood of non-hypervascular hypointense nodules progressing to HCC, the results of the present study suggest that the presence of non-hypervascular hypointense nodules detected during the hepatobiliary

phase of preoperative Gd-EOB-DTPA-enhanced MRI may indicate a high risk of multicentric recurrence of HCC after hepatectomy. Interestingly, multicentric recurrence was observed in all patients with multiple preoperative non-hypervascular hypointense nodules. Whereas intrahepatic metastasis recurrence is considered as a occurrence of metastasis of the HCC that had been resected, multicentric recurrence is considered as new development of HCC that is not related to the resected HCC. Therefore, the presence of non-hypervascular hypointense nodules, especially multiple nodules, may indicate enhanced hepatocarcinogenesis even when the nodule itself does not progress to HCC.

There are several limitations to this study. The sample size was not large and the observation period was relatively short because Gd-EOB-DTPA has been in clinical use since February 2008 in Japan. In addition, the impact of the presence of non-hypervascular hypointense hepatic nodules on survival after hepatectomy was not analyzed because there were no patient deaths during the study period. However, we believe that our data should be shared with clinicians because of the markedly high rates of recurrence after hepatectomy in patients with preoperative

non-hypervascular hypointense hepatic nodules. Further studies with more patients and a longer observation period are needed to confirm this observation. Furthermore, measures to suppress multicentric recurrence in patients with preoperative concurrent non-hypervascular hypointense hepatic nodules should be investigated in the future.

In conclusion, patients with preoperative concurrent non-hypervascular hypointense hepatic nodules on the hepatobiliary phase of Gd-EOB-DTPA-enhanced MRI are at higher risk of HCC recurrence after hepatectomy. Clinicians should take this into consideration when determining of treatment modalities.

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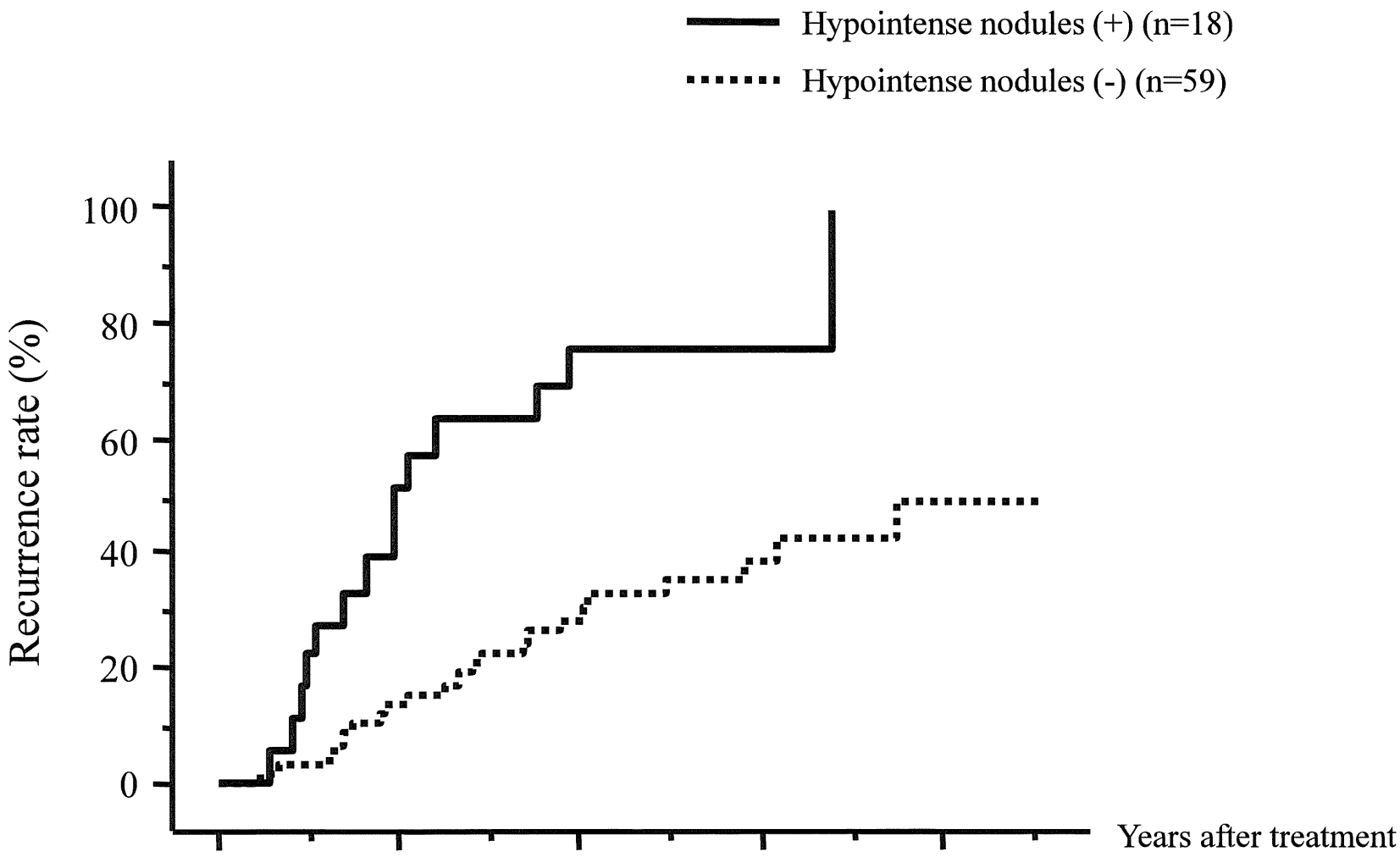
Figure legends

Figure 1. Overall recurrence rate after hepatectomy in patients with or without concurrent non-hypervascular hypointense hepatic nodules detected during the hepatobiliary phase of preoperative gadolinium-ethoxybenzyl-diethylenetriamine pentaacetic acid-enhanced magnetic resonance imaging.

Figure 2. (A) Rates of intrahepatic metastasis recurrence after hepatectomy in patients with or without concurrent non-hypervascular hypointense hepatic nodules detected during the hepatobiliary phase of preoperative gadolinium-ethoxybenzyl-diethylenetriamine pentaacetic acid-enhanced magnetic resonance imaging. (B) Rates of multicentric recurrence after hepatectomy in patients with or without concurrent non-hypervascular hypointense hepatic nodules detected during the hepatobiliary phase of preoperative gadolinium-ethoxybenzyl-diethylenetriamine pentaacetic acid-enhanced magnetic resonance imaging, among 59 patients excluding 16 patients with intrahepatic metastasis recurrence.

Figure 3. (A) Hepatobiliary phase of gadolinium-ethoxybenzyl-diethylenetriamine pentaacetic acid-enhanced magnetic resonance imaging (Gd-EOB-DTPA-enhanced MRI, left panel) and computed tomography during hepatic arteriography (CTHA, right panel) before hepatectomy for hepatocellular carcinoma (HCC). In addition to the typical HCC located in segment VIII, hypointense hepatic nodule was detected in segment VI during the heratobiliary phase of Gd-EOB-DTPA-enhanced MRI (arrow). No hypervascular nodule was detected at this site by CTHA (arrow). (B) Hepatobiliary phase of gadolinium-ethoxybenzyl-diethylenetriamine pentaacetic acid-enhanced magnetic resonance imaging (Gd-EOB-DTPA-enhanced MRI, left panel) and computed tomography during hepatic arteriography (CTHA, right panel) 10 months after hepatectomy for hepatocellular carcinoma (HCC). The nodule detected during the heratobiliary phase of Gd-EOB-DTPA-enhanced MRI showed minute growth in size with clearer margin comparing preoperative image (arrow). The hypervascularity of this nodule was identified by CTHA (arrow). This nodule was detected by re-hepatectomy and was diagnosed as HCC pathologically.

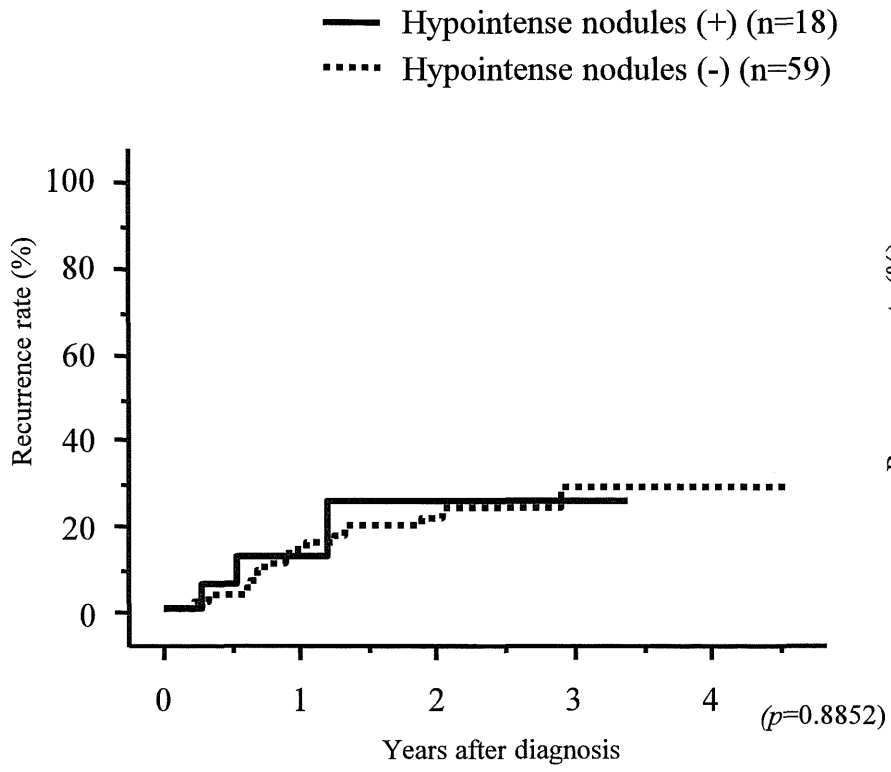


Patients at risk	0	1	2	3	4
Hypointense nodules (+)	18	17	14	7	2
Hypointense nodules (-)	59	58	47	29	16

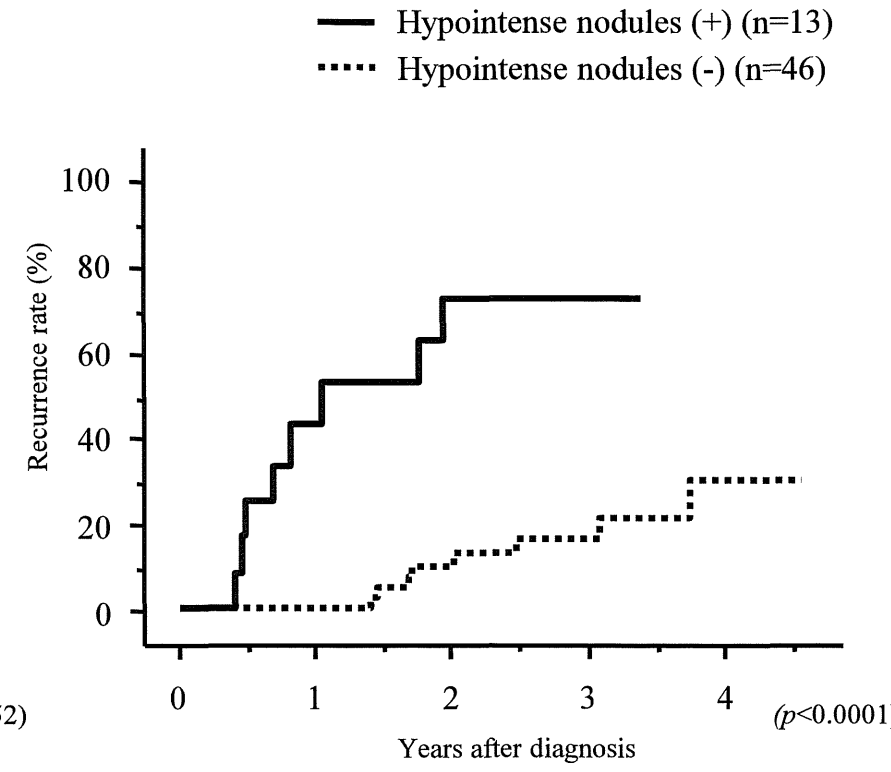
(p<0.0001)

Figure 1

(A)



(B)



Patients at risk

Hypointense nodules (+)	18	17	14	7	2	13	12	10	5	1
Hypointense nodules (-)	59	58	47	29	16	46	44	35	19	9

Figure 2

(A)

(B)

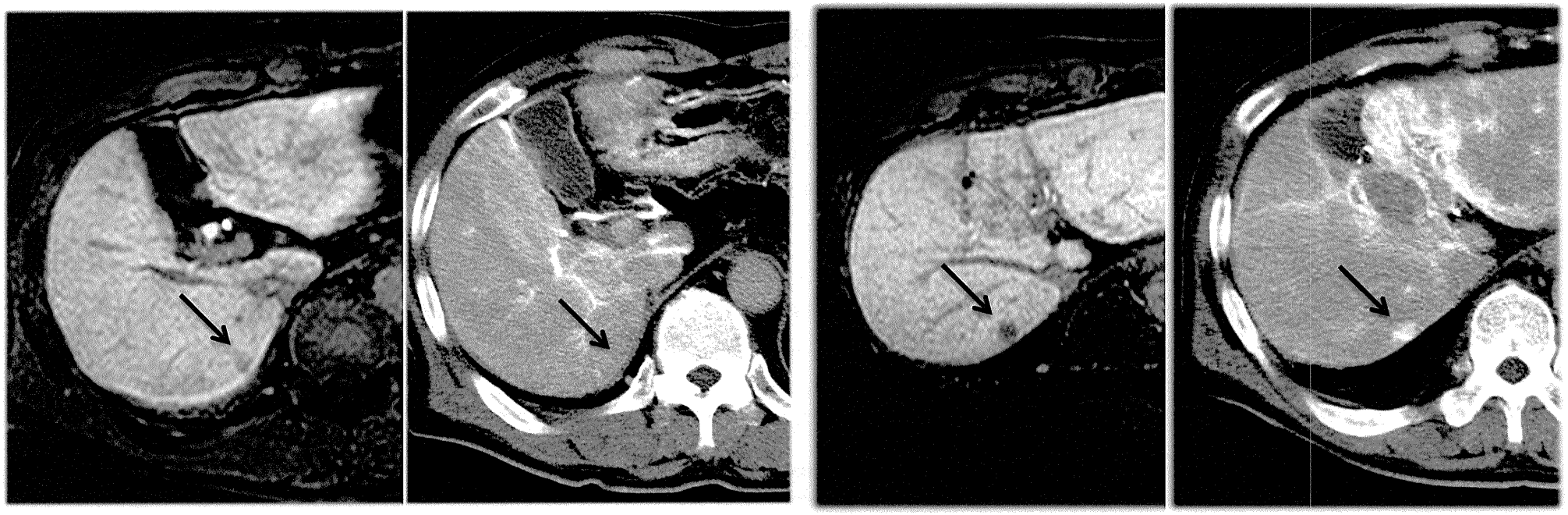


Figure 3

Table 1. Comparison of clinical characteristics of study patients based on the presence of non-hypervascular hypointense nodules detected during the hepatobiliary phase of gadolinium-ethoxybenzyl-diethylenetriamine pentaacetic acid-enhanced magnetic resonance imaging (n = 77)

	Non-hypervascular hypointense nodule (+) (n=18)	Non-hypervascular hypointense nodule (-) (n=59)	*p* value
Age (mean ± SD, years) (range)	65.8 ± 9.0 (46-76)	69.1 ± 7.0 (53-82)	0.2727
Sex (female/male)	3 (16.7)/ 15 (83.3)	18 (30.5)/ 41 (69.5)	0.3921
Etiology (HBV/ HCV/ non-HBV, non-HCV)	2 (11.1)/ 11 (61.1)/ 5 (27.8)	9 (15.3)/ 39 (66.1)/ 11 (18.6)	0.6796
Child-Pugh class (A/B)*	17 (94.4)/ 1 (5.6)	58 (98.3)/ 1 (1.7)	0.9474
Albumin (mean ± SD, g/dL)	3.91 ± 0.51	4.08 ± 0.32	0.1664
Total bilirubin (mean ± SD, mg/dL)	0.88 ± 0.36	0.84 ± 0.33	0.7296
15-minute ICG retention rate (%)	18.1 ± 5.4	16.0 ± 6.7	0.2405
Prothrombin (%)	95.3 ± 15.6	95.1 ± 11.2	0.9105
Platelet count (x1000/mL)	132 ± 47	152 ± 66	0.5433
Tumor size (mean ± SD, cm) (range)	2.52 ± 0.99 (1.3-4.7)	2.84 ± 1.54 (1.0-8.6)	0.6600
Number of tumors (single/multiple)	15 (83.3)/ 3 (16.7)	53 (89.8)/ 6 (10.2)	0.7358
Portal vein invasion (absent/present)**	17 (94.4)/ 1 (5.6)	50 (84.7)/ 9 (15.3)	0.4989
Differentiation (well-/moderately or poorly)**	7 (38.9)/ 11 (61.1)	21 (35.6)/ 38 (64.4)	0.9999
Growth pattern (expansive/ infiltrative)**	14 (77.8)/ 4 (22.2)	52 (88.1)/ 7 (11.9)	0.4718
Follow-up period (months) (median, range)	31.3 (9.4-53.9)	34.9 (8.5-55.4)	0.4200

Percentages were in parentheses. HBV, hepatitis B virus; HCV, hepatitis C virus; ICG, indocyanine green test.

*Child-Pugh class A includes patients without cirrhosis.

**Evaluated by pathologic examination based of resected specimens.

Table 2. Univariate and multivariate analyses of factors associated with post-operative recurrence in HCC patients (n=77)

Factor		Univariate analysis		Multivariate analysis	
		Risk ratio (95% C.I.)	*p* value	Risk ratio (95% C.I.)	*p* value
Age		0.9943 (0.9535-1.0396)	0.7974	-----	
Sex	Male	1			
	Female	1.0068 (0.6818-1.4290)	0.9711	-----	
Child-Pugh class*	A	1			
	B	0.0428 (0.0198-1.5669)	0.2068	-----	
Tumor size		0.9376 (0.7179-1.1700)	0.5935	-----	
Number of tumors	Single	1			
	Multiple	1.0419 (0.5669-1.6643)	0.8792	-----	
Differentiation**	Well-	1		1	
	Moderately/poorly	1.5871 (1.0958-2.4354)	0.0134	1.6536 (1.1381-2.5445)	0.0073
Growth pattern**	Expansive	1			
	Infiltrative	1.1101 (0.6798-1.6625)	0.6487	-----	
Portal vein invasion**	Absent	1		1	
	Present	1.5659 (1.0161-2.2813)	0.0428	1.7818 (1.1388-2.6597)	0.0134
Non-hypervascular	Absent	1		1	
hypointense nodules	Present	1.9396 (1.3615-2.7222)	0.0004	2.1767 (1.5089-3.1105)	0.0001

C.I., conidence interval.

*Child-Pugh class A includes patients without cirrhosis.

**Evaluated by pathologic examination of resected specimens.

Table 3. Univariate and multivariate analyses of factors associated with post-operative intrahepatic metastasis recurrence in HCC patients (n=77)

Factor		Univariate analysis		Multivariate analysis	
		Risk ratio (95% C.I.)	*p* value	Risk ratio (95% C.I.)	*p* value
Age		0.9825 (0.9265-1.0470)	0.5743	-----	
Sex	Male	1			
	Female	0.9022 (0.4784-1.5192)	0.7148	-----	
Child-Pugh class*	A	1			
	B	0.0242 (0.0059-2.1819)	0.3573	-----	
Tumor size		1.0051 (0.6929-1.3406)	0.9755	-----	
Number of tumors	Single	1			
	Multiple	0.7038 (0.1655-1.5643)	0.4504	-----	
Differentiation**	Well-	1		1	
	Moderately/poorly	1.7843 (1.0185-3.7176)	0.0424	1.6742 (0.9520-3.4993)	0.0769
Growth pattern**	Expansive	1			
	Infiltrative	0.9266 (0.3678-1.7453)	0.8365	-----	
Portal vein invasion**	Absent	1		1	
	Present	2.1224 (1.2405-3.4608)	0.0079	2.0041 (1.1672-3.2828)	0.0138
Non-hypervascular	Absent	1			
hypointense nodules	Present	1.0474 (0.5012-1.8442)	0.8864	-----	

C.I., conidence interval.

*Child-Pugh class A includes patients without cirrhosis.

**Evaluated by pathologic examination of resected specimens.

Table 4. Univariate and multivariate analyses of factors associated with post-operative multicentric recurrence in HCC patients (n=59)

Factor		Univariate analysis		Multivariate analysis	
		Risk ratio (95% C.I.)	*p* value	Risk ratio (95% C.I.)	*p* value
Age		1.0047 (0.9359-1.0823)	0.8985	-----	
Sex	Male	1			
	Female	1.0701 (0.5999-1.7781)	0.8038	-----	
Child-Pugh class*	A	1			
	B	0.0664 (0.0176-5.7947)	0.7029	-----	
Tumor size		0.9517 (0.6300-1.2943)	0.7801	-----	
Number of tumors	Single	1			
	Multiple	1.1331 (0.4469-2.1714)	0.7510	-----	
Differentiation**	Well-	1			
	Moderately/poorly	1.5198 (0.8959-2.8769)	0.1249	-----	
Growth pattern**	Expansive	1			
	Infiltrative	1.3486 (0.7124-2.2884)	0.3270	-----	
Portal vein invasion**	Absent	1			
	Present	1.2908 (0.5077-2.4730)	0.5312	-----	
Non-hypervascular	Absent	1		1	
hypointense nodules	Present	2.8436 (1.6900-4.8407)	0.0002	2.8436 (1.6900-4.8407)	0.0002