Ann Surg Oncol (2021) 28:797–805
https://doi.org/10.1245/s10434-020-09390-w

Annals of
SURGICAL ONCOLOGY
OFFICIAL JOURNAL OF THE SOCIETY OF SURGICAL ONCOLOGY

ORIGINAL ARTICLE – HEPATOBILIARY TUMORS

Tumor Necrosis Impacts Prognosis of Patients Undergoing Curative-Intent Hepatocellular Carcinoma

Tao Wei, MD, PhD[1,2], Xu-Feng Zhang, MD[1,3], Fabio Bagante, MD[3,4], Francesca Ratti, MD[5], Hugo P. Marques, MD[6], Silvia Silva, MD[6], Olivier Soubrane, MD[7], Vincent Lam, MD[8], George A. Poultsides, MD[9], Irinel Popescu, MD[10], Razvan Grigorie, MD, PhD[10], Sorin Alexandrescu, MD[10], Guillaume Martel, MD[11], Aklile Workneh, MD[11], Alfredo Guglielmi, MD[4], Tom Hugh, MD[12], Luca Aldrighetti, MD[5], Itaru Endo, MD, PhD[13], and Timothy M. Pawlik, MD, MPH, PhD, FACS, FRACS (Hon.)[3]

[1]Department of Hepatobiliary Surgery and Institute of Advanced Surgical Technology and Engineering, The First Affiliated Hospital of Xi'an Jiaotong University, Xi'an, China; [2]Department of Hepatobiliary and Pancreatic Surgery, The First Affiliated Hospital, Zhejiang University School of Medicine, Hangzhou, China; [3]Department of Surgery, Division of Surgical Oncology, Professor of Surgery, Oncology, Health Services Management and Policy, The Urban Meyer III and Shelley Meyer Chair for Cancer Research, The Ohio State University Wexner Medical Center and James Comprehensive Cancer Center, Columbus, OH; [4]Department of Surgery, University of Verona, Verona, Italy; [5]Department of Surgery, Ospedale San Raffaele, Milan, Italy; [6]Department of Surgery, Curry Cabral Hospital, Lisbon, Portugal; [7]Department of Hepatobiliopancreatic Surgery, APHP, Beaujon Hospital, Clichy, France; [8]Department of Surgery, Westmead Hospital, Sydney, Australia; [9]Department of Surgery, Stanford University, Stanford, CA; [10]Department of Surgery, Fundeni Clinical Institute, Bucharest, Romania; [11]Department of Surgery, University of Ottawa, Ottawa, Canada; [12]Department of Surgery, School of Medicine, The University of Sydney, Sydney, Australia; [13]Yokohama City University School of Medicine, Yokohama, Japan

ABSTRACT

Background. The impact of tumor necrosis relative to prognosis among patients undergoing curative-intent resection for hepatocellular carcinoma (HCC) remains ill-defined.

Methods. Patients who underwent curative-intent resection for HCC without any prior treatment between 2000 and 2017 were identified from an international multi-institutional database. Tumor necrosis was graded as absent, moderate (< 50% area), or extensive (≥ 50% area) on histological examination. The relationship between tumor necrosis, clinicopathologic characteristics, and long-term survival were analyzed.

Results. Among 919 patients who underwent curative-intent resection for HCC, the median tumor size was 5.0 cm (IQR, 3.0–8.5). Tumor necrosis was present in 367 (39.9%) patients (no necrosis: $n = 552$, 60.1% vs < 50% necrosis: $n = 256$, 27.9% vs ≥ 50% necrosis: $n = 111$, 12.1%). Extent of tumor necrosis was also associated with more advanced tumor characteristics. HCC necrosis was associated with OS (median OS: no necrosis, 84.0 months vs < 50% necrosis, 73.6 months vs ≥ 50% necrosis: 59.3 months; $p < 0.001$) and RFS (median RFS: no necrosis, 49.6 months vs < 50% necrosis, 38.3 months vs ≥ 50% necrosis: 26.5 months; $p < 0.05$). Patients with T1 tumors with extensive ≥ 50% necrosis had an OS comparable to patients with T2 tumors (median OS, 62.9 vs 61.8 months; $p = 0.645$). In addition, patients with T2 disease with necrosis had long-term outcomes comparable to patients with T3 disease (median OS, 61.8 vs 62.4 months; $p = 0.713$).

Tao Wei and Xu-Feng Zhang contributed equally to this work.

Electronic supplementary material The online version of this article (https://doi.org/10.1245/s10434-020-09390-w) contains supplementary material, which is available to authorized users.

First Received: 12 July 2020
Accepted: 5 October 2020;
Published Online: 28 November 2020

T. M. Pawlik, MD, MPH, PhD, FACS, FRACS (Hon.)
e-mail: Tim.Pawlik@osumc.edu

Conclusion. Tumor necrosis was associated with worse OS and RFS, as well as T-category upstaging of patients. A modified AJCC T classification that incorporates tumor necrosis should be considered in prognostic stratification of HCC patients.

Hepatocellular carcinoma (HCC) is the second leading cause of cancer-related death worldwide.[1] Surgical resection remains a common curative-intent treatment option for patients with HCC. Survival following hepatectomy for HCC is poor due to a high incidence of postoperative recurrence in both the short and long term.[2,3] In turn, considerable efforts have been made to identify prognostic factors to guide disease surveillance strategies, possible adjuvant therapy, as well as inform discussions related to prognosis.[4,5] The American Joint Committee on Cancer (AJCC) TNM staging system incorporates several pathological features, such as the tumor size and number, as well as presence of vascular invasion, into the prognostic schema of HCC.[6] However, patients within the same pathologic stage often have divergent long-term outcomes, suggesting that other factors may be important in the prognostic stratification of patients with HCC.[7]

Tumor necrosis is frequently observed on histopathological examination of solid tumors. The clinical significance of tumor necrosis has been studied in several malignancies including renal clear cell carcinoma,[8,9] breast cancer,[10] lung cancer,[11] and colorectal cancer.[12,13] Previous data have suggested that the presence of tumor necrosis may be associated with worse survival.[7] Other studies have noted that necrosis may impact prognosis among patients with primary liver malignancies including intrahepatic and hilar cholangiocarcinoma.[14,15] Cholangiocarcinoma is typically a poorly vascularized tumor and tumor necrosis has been attributed to relative hypoperfusion of the tissue.[14,15] In contrast, HCC is traditionally a highly vascularized cancer. In turn, HCC associated tumor necrosis has been postulated to be due to aggressive tumor growth, impaired oxygen delivery, as well as increased inflammation in the tumor microenvironment.[16–18] The relevance of tumor necrosis in HCC among patients undergoing hepatic resection has not been well defined. As such, the objective of the current study was to correlate HCC tumor necrosis with other tumor-specific characteristics, as well as define the prognostic impact of tumor necrosis among patients undergoing curative-intent resection of HCC.

METHODS

Study Population

Patients who underwent surgical resection with curative intent for HCC between 2000 and 2017 were identified from an international multi-institutional database.[3] Patients were followed and outcomes were recorded in a pre-determined multi-institutional database. The study was approved by the institutional review board of each participating institution. Patients who died within 30 days after surgery were excluded.

Clinicopathological Variables

Clinicopathological factors, including age, gender, α-fetoprotein (AFP), Child–Pugh classification, Barcelona Clinic Liver Cancer (BCLC) staging, maximum tumor size, tumor number and location, tumor differentiation/grade, presence of cirrhosis and microvascular invasion, liver capsule involvement, and width of resection margin, were recorded. Presence and extent of tumor necrosis was prospectively evaluated for all patients at each center. Specifically, tumor necrosis was histologically classified into three groups: absence of necrosis, moderate necrosis ($< 50\%$), and extensive necrosis ($\geq 50\%$). For patients with multiple lesions, tumor necrosis status was defined according to the lesion with the most severe necrosis. Tumor staging was classified according to the eighth AJCC TNM staging manual.[19]

Long-term outcomes including overall survival (OS) and recurrence-free survival (RFS) were calculated from the date of surgery. Recurrence patterns, including timing of recurrence [early (within 12 months) or late recurrence (beyond 12 months)], recurrence site (intrahepatic, extrahepatic or both), and tumor number associated with the recurrence, as well as treatment approach for recurrent lesions, were recorded. Local recurrence was defined as recurrence close to the resection margin of the primary tumor.

Statistical Analysis

Clinicopathological variables were summarized using frequencies and percentages for categorical variables, as well as medians and interquartile range (IQR) for continuous covariates. Categorical covariates were compared using the Chi square test or Fisher's exact test, and continuous variables with the Mann–Whitney *U* test. OS and RFS were calculated by the Kaplan–Meier method and differences were compared using the log-rank test. Univariate analysis was performed to screen potential risk factors for OS; factors with *P* values less than 0.1 were

included to identify independent risk factors using a multivariate Cox regression model. The Akaike information criterion (AIC) provided an objective way to determine which model (8th AJCC T categories vs modified T classification plus extensive necrosis) was better at predicting long-term survival of patients. Specifically, the better prognostic model had a lower AIC value. All statistical analyses were conducted using SPSS version 23.0 (IBM SPSS, Chicago, IL, USA). A 2-tailed P value of < 0.05 was considered statistically significant.

RESULTS

Baseline Characteristics

A total of 919 patients underwent curative-intent resection for HCC and were included in the analytic cohort. Median patient age was 62 years (IQR, 53–71); the majority of patients were male (n = 734, 80.0%) and had an ASA class ≤ 2 (n = 552, 59.9%). Most patients had unifocal disease (n = 802, 87.3%) with a median tumor size of 5.0 cm (IQR, 3.0–8.5); most tumors were categorized as BCLC stage 0 or A (n = 775, 84.7%). Median AFP was 15.8 ng/ml (IQR, 3.6–187.0) and the overwhelming majority of patients presented with well-compensated Child–Pugh class A liver function (n = 687, 93.9%). On histopathological examination, most patients (n = 672, 76.2%) had well-differentiated to moderately differentiated tumors and a subset had microvascular invasion (n = 280, 33.1%) (Table 1).

On pathological examination, tumor necrosis was present in 367 (39.9%) patients (no necrosis: n = 552, 60.1% vs $< 50\%$ necrosis: n = 256, 27.9% vs $\geq 50\%$ necrosis: n = 111, 12.1%). Of note, the presence of tumor necrosis correlated with several adverse clinicopathological characteristics. Specifically, patients with necrotic HCC tumors were more likely to be older, have a larger tumor size, as well as have poor tumor differentiation and vascular invasion compared with patients who had no tumor necrosis (all $p < 0.001$) (Table 1). Of note, extent of tumor necrosis was also associated with clinicopathological factors, as patients who had resection of HCC $\geq 50\%$ necrosis were more likely to have higher AFP (> 400 ng/ml: $< 50\%$ necrosis: n = 40, 18.1% vs $\geq 50\%$ necrosis: n = 32, 34.1%), larger tumors (> 5 cm: $< 50\%$ necrosis: n = 147, 57.4% vs $\geq 50\%$ necrosis: n = 80, 72.1%), multifocal disease (multifocal: $< 50\%$ necrosis: n = 28, 10.9% vs $\geq 50\%$ necrosis: n = 21, 18.9%), vascular invasion (present: $< 50\%$ necrosis: n = 16, 6.3% vs $\geq 50\%$ necrosis: n = 14, 12.7%), as well as advanced BCLC stage (BCLC stage B/C: $< 50\%$ necrosis: n = 35, 13.7% vs $\geq 50\%$ necrosis: n = 30, 27.3%) (all $p < 0.05$) (Table 1).

Impact of Tumor Necrosis on Overall Survival and Recurrence

Median follow-up of the entire cohort was 28.8 months (IQR, 14.7–49.9). Median OS, 1-, 3-, and 5-year OS among the entire cohort was 72.1 months, 84.0%, 69.1%, and 55.4%, respectively; median RFS, 1-, 3-, and 5-year RFS was 45.7 months, 61.0%, 48.9%, and 38.7%, respectively. HCC tumor necrosis was associated with both OS (median OS: no necrosis, 84.0 months vs $< 50\%$ necrosis, 73.6 months vs $\geq 50\%$ necrosis: 59.3 months; $p < 0.001$) (Fig. 1a) and RFS (median RFS: no necrosis, 49.6 months vs $< 50\%$ necrosis, 38.3 months vs $\geq 50\%$ necrosis: 26.5 months; $p < 0.05$) (Fig. 1b).

At last follow-up, 443 (44.1%) patients had experienced a recurrence. The majority of patients (n = 248, 74.3%) had recurred solely at an intrahepatic site, whereas a subset of individuals had extrahepatic metastasis (n = 59, 17.7%) or simultaneous intra- and extrahepatic recurrence (n = 27, 8.1%). Of note, the presence of HCC tumor necrosis was associated with a higher likelihood of extrahepatic $\pm$ intrahepatic recurrence (no tumor necrosis, 15.9% vs tumor necrosis, 37.7%; $p < 0.001$) (Table 2). In addition, patients with HCC necrosis were more likely to experience an early recurrence within 12 months after hepatic resection (early recurrence: no necrosis 73.2% versus necrosis 83.6%, p = 0.014) (Table 2).

Correlation of Tumor Necrosis with Tumor Size and AJCC T Category

The incidence of tumor necrosis incrementally increased with tumor size (≤ 5 cm, 29.8% vs > 5 cm, 50.6%; $p < 0.01$). Among patients with tumors ≤ 5 cm (median OS: no necrosis, 84.7 months vs moderate necrosis, 74.4 months vs extensive necrosis, 50.6 months; $p < 0.001$), as well as patients with HCC > 5 cm (median OS: no necrosis, 74.5 months vs moderate necrosis, 61.8 months vs extensive necrosis, 61.2 months; $p < 0.001$), the risk of death was associated with extent of tumor necrosis (Fig. 2a, b).

Additional stratified analyses were performed relative to AJCC T category. Of note, among patients with T1 tumors (n = 576) (solitary tumor ≤ 2 cm, or > 2 cm without vascular invasion), patients with HCC tumors that had extensive necrosis had a worse OS compared with patients who had a tumor with moderate necrosis (median OS, 62.9 vs not attained; p = 0.040) or no necrosis (median OS, 62.9 vs 89.7 months; p = 0.001)(Fig. 3a). Interestingly, patients with T1 tumors that had extensive $\geq 50\%$ necrosis had an OS comparable to patients with T2 tumors (solitary tumor > 2 cm with vascular invasion, or multiple tumors, none > 5 cm) (median OS, 62.9 vs 61.8 months;

TABLE 1 Clinicopathological variables of patients stratified by tumor necrosis

Variables	Total (n = 919)	Presence of necrosis			Severity of necrosis		
		Absence (n = 552)	Presence (n = 367)	P	Moderate (n = 256)	Extensive (n = 111)	P
Age, years				0.008			0.276
≤ 60	432 (47.0%)	279 (50.5%)	153 (41.7%)		102 (39.8%)	51 (45.9%)	
> 60	487 (53.0%)	273 (49.5%)	214 (58.3%)		154 (60.2%)	60 (54.1%)	
Gender				0.395			0.116
Male	734 (80.0%)	436 (79.1%)	298 (81.4%)		213 (83.5%)	85 (76.6%)	
Female	183 (20.0%)	115 (20.9%)	68 (18.6%)		42 (16.5%)	26 (23.4%)	
AFP, ng/ml				0.273			0.008
≤ 400	616 (79.6%)	365 (80.9%)	251 (77.7%)		181 (81.9%)	70 (68.6%)	
> 400	158 (20.4%)	86 (19.1%)	72 (22.3%)		40 (18.1%)	32 (31.4%)	
Neutrophil-to-lymphocyte ratio	3.0 ± 2.7	2.8 ± 2.4	3.4 ± 3.2	< 0.001	3.3 ± 2.7	3.5 ± 4.2	0.871
Child–Pugh				0.860			0.399
A	687 (93.9%)	412 (94.1%)	285 (93.8%)		199 (93.0%)	86 (95.6%)	
B	45 (6.1%)	26 (5.9%)	19 (6.3%)		15 (7.0%)	4 (4.4%)	
Tumor size, cm				< 0.001			0.008
≤ 5	470 (51.1%)	330 (59.8%)	140 (38.1%)		109 (42.6%)	31 (27.9%)	
> 5	449 (48.9%)	222 (40.2%)	227 (61.9%)		147 (57.4%)	80 (72.1%)	
Tumor number				0.646			0.039
Single	802 (87.3%)	484 (87.7%)	318 (86.6%)		228 (89.1%)	90 (81.1%)	
Multiple	117 (12.7%)	68 (12.3%)	49 (13.4%)		28 (10.9%)	21 (18.9%)	
Macrovascular invasion	54 (5.9%)	24 (4.4%)	30 (8.2%)	0.015	16 (6.3%)	14 (12.7%)	0.039
Tumor location				0.364			0.148
Unilobar	846 (92.5%)	513 (93.1%)	333 (91.5%)		235 (92.9%)	98 (88.3%)	
Bilobar	69 (7.5%)	38 (6.9%)	31 (8.5%)		18 (7.1%)	13 (11.7%)	
BCLC staging				0.086			0.002
0/A	775 (84.7%)	475 (86.4%)	300 (80.2%)		220 (86.3%)	80 (72.7%)	
B/C	140 (15.3%)	75 (13.6%)	65 (17.8%)		35 (13.7%)	30 (27.3%)	
Cirrhosis	398 (43.4%)	262 (47.6%)	136 (37.1%)	0.002	94 (36.7%)	42 (37.8%)	0.838
Grade				< 0.001			0.790
Well/moderate	672 (76.2%)	434 (81.3%)	238 (68.4%)		172 (68.0%)	66 (69.5%)	
Poor	210 (23.8%)	100 (18.7%)	110 (31.6%)		81 (32.0%)	29 (30.5%)	
Microvascular invasion	280 (33.1%)	135 (27.0%)	145 (41.8%)	< 0.001	98 (39.7%)	47 (47.0%)	0.210
Capsule involvement	277 (37.5%)	166 (38.0%)	111 (36.8%)	0.734	74 (33.6%)	37 (45.1%)	0.066
AJCC 8th T categories				0.002			0.364
T1	576 (65.6%)	381 (70.4%)	195 (57.9%)		139 (59.1%)	56 (54.9%)	
T2	193 (22.0%)	103 (19.0%)	90 (26.7%)		65 (27.7%)	25 (24.5%)	
T3	60 (6.8%)	32 (5.9%)	28 (8.3%)		16 (6.8%)	12 (11.8%)	
T4	49 (5.6%)	25 (4.6%)	24 (7.1%)		15 (6.4%)	9 (8.8%)	

AFP, α-fetoprotein; BCLC, Barcelona Clinic Liver Cancer; AJCC, American Joint Committee on Cancer

p = 0.645) (Fig. 3b). In addition, patients with T2 disease with extensive necrosis had long-term outcomes comparable to patients with T3 disease (multifocal tumors at least one of which > 5 cm) (median OS, 61.8 months vs 62.4 months; p = 0.713). Of note, on multivariable analysis, after controlling for competing risk factors, while patients with T3 tumors (referent, T1: HR 1.9, 95% CI 1.1–3.2, p = 0.017) had a worse long-term survival compared with individuals who had T1 HCC, there was no survival difference comparing T2 versus T1 tumors

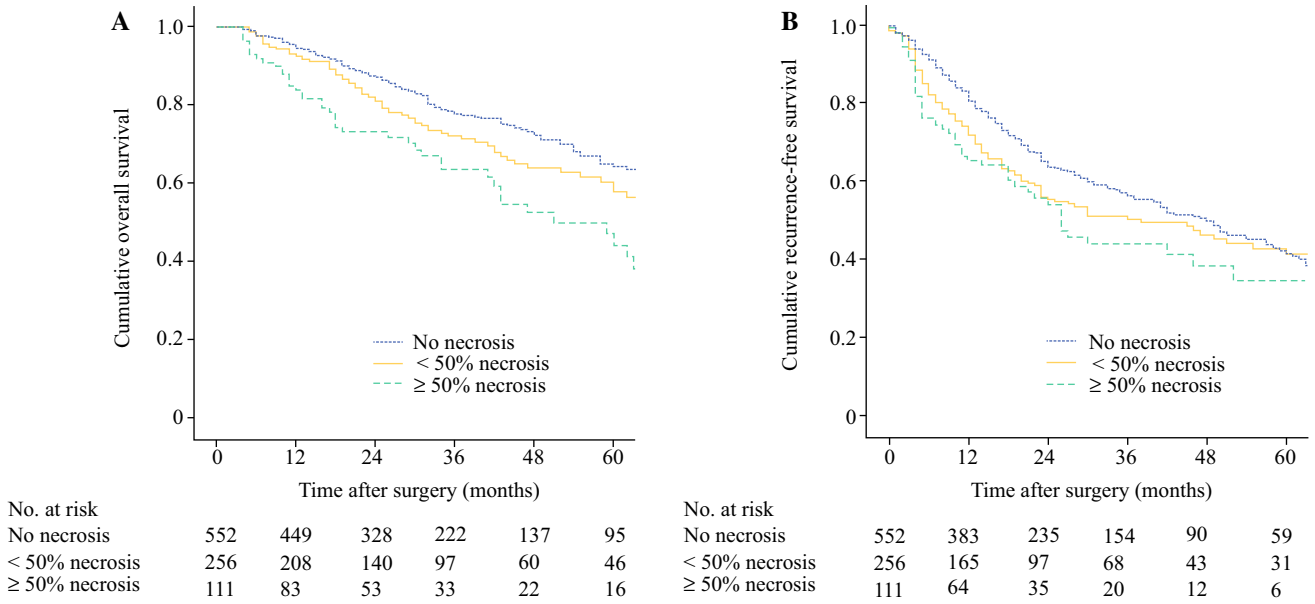

FIG. 1 Overall survival (**a**) and recurrence-free survival (**b**) of patients with HCC after curative-intent resection stratified by extent of necrosis

TABLE 2 Recurrence patterns of patients stratified by tumor necrosis

Variables	Presence of necrosis			Severity of necrosis		
	Absence (n = 552)	Presence (n = 367)	P	<50% (n = 256)	≥ 50% (n = 111)	P
Timing of recurrence	n = 224	n = 171		n = 117	n = 54	
Early	164 (73.2%)	143 (83.6%)	0.014	98 (83.8%)	45 (83.3%)	0.944
Late	60 (26.8%)	28 (16.4%)		19 (16.2%)	9 (16.7%)	
Local recurrence	n = 183	n = 149		n = 104	n = 45	
Yes	22 (12.0%)	17 (11.4%)	0.863	14 (13.5%)	3 (6.7%)	0.231
No (away from margin)	161 (88.0%)	132 (88.6%)		90 (86.5%)	42 (93.3%)	
Recurrence site	n = 183	n = 151		n = 105	n = 46	
Intrahepatic	154 (84.2%)	94 (62.3%)	< 0.001	63 (60.0%)	31 (67.4%)	0.662
Extrahepatic	23 (12.6%)	36 (23.8%)		27 (25.7%)	9 (19.6%)	
Both	6 (3.3%)	21 (13.9%)		15 (14.3%)	6 (13.0%)	
Number of recurrent lesions	n = 139	n = 102		n = 69	n = 33	
Single	84 (60.4%)	61 (59.8%)	0.922	43 (62.3%)	18 (54.5%)	0.454
Multiple	55 (39.6%)	41 (40.2%)		26 (37.7%)	15 (45.5%)	

(referent, T1: HR 1.2, 95% CI 0.8–1.8, p = 0.495) (Table 3) (Supplemental Fig. 1a). In contrast, after incorporating necrosis into the T categories (i.e., Tn), patients with T1n versus T2n versus T3n HCC had an incrementally worse long-term prognosis (referent T1n: T2n, HR 1.4, 95% CI 1.0–2.1, p = 0.034; T3n, HR 1.9, 95% CI 1.2–3.1, p = 0.007) (Table 3) (Supplemental Fig. 1b). The proposed T categories that incorporated necrosis demonstrated a lower AIC, and thus better performance to stratify patient prognosis versus the current AJCC T categories (AIC: 2549.5 vs 2763.1).

DISCUSSION

Tumor necrosis is generally attributed to rapid tumor growth, inadequate tissue vascularization, as well as sustained tissue hypoxia and associated local inflammation.[15,17,20] While tumor necrosis has been observed on pathological examination of different solid tumor types, the impact of tumor necrosis on outcomes after curative-intent resection of HCC has not been well defined. The current study was therefore important as we specifically examined the effect of HCC tumor necrosis on

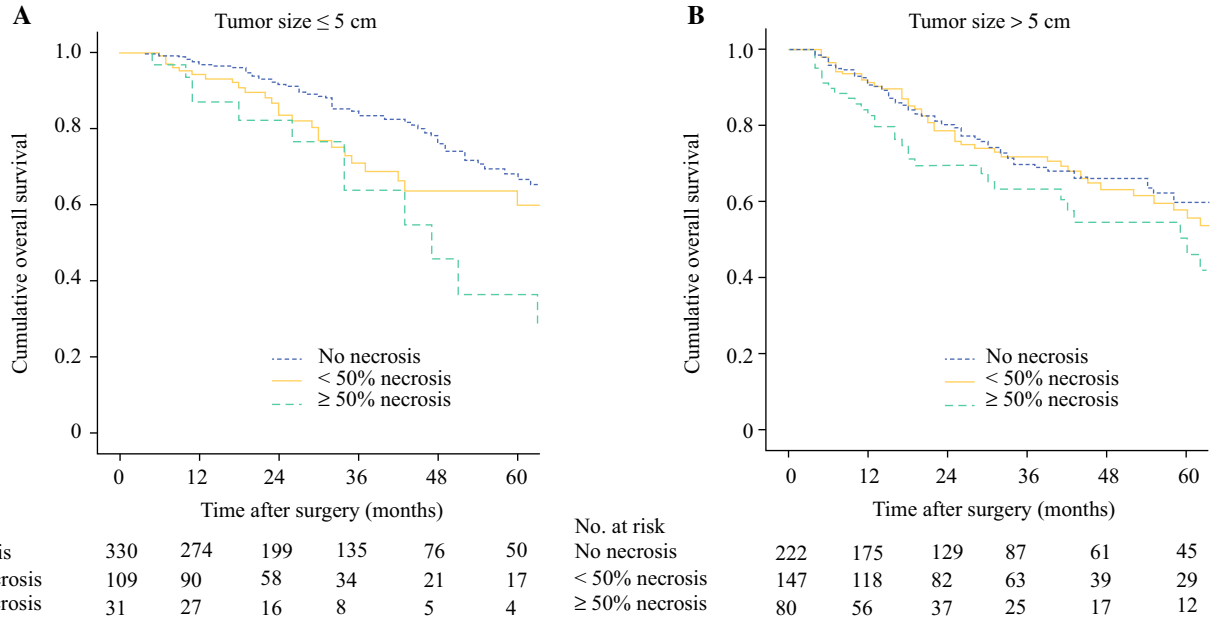

FIG. 2 Overall survival of patients with HCC after curative-intent resection according to extent of necrosis stratified by tumor size ≤ 5 cm (**a**), and > 5 cm (**b**)

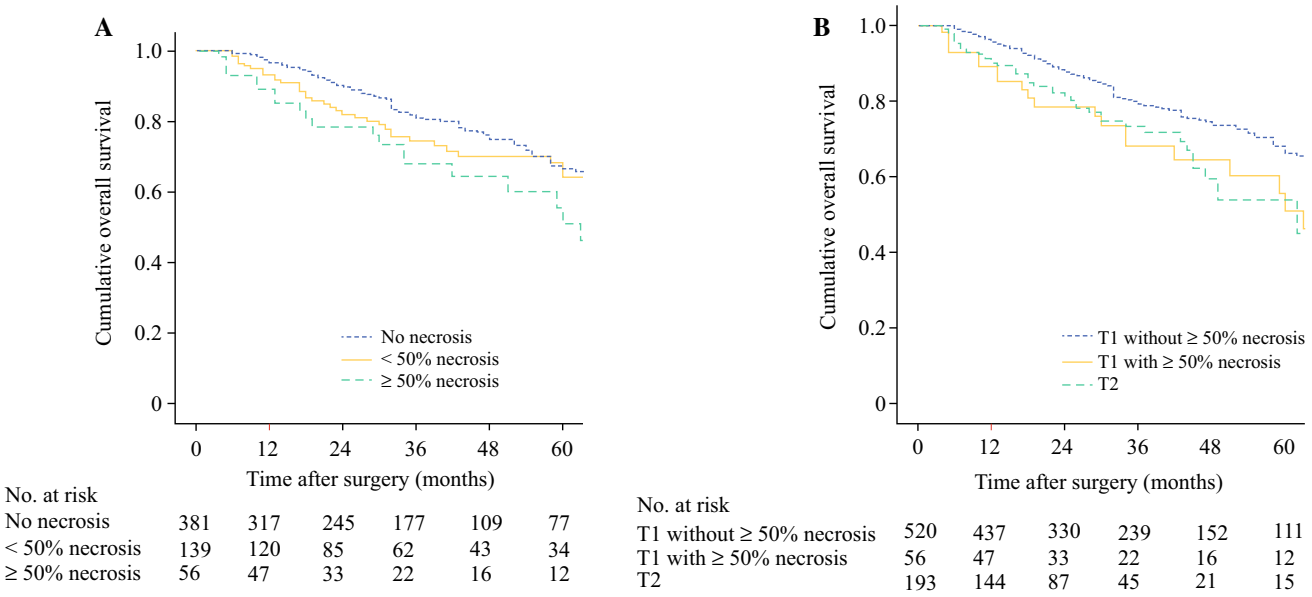

FIG. 3 **a** Overall survival of patients with AJCC pathological T1 tumors after curative-intent resection stratified by extent of necrosis. **b** "Up-staging" of AJCC pathological T1 and T2 tumors due to extensive tumor necrosis (≥ 50% necrosis)

long-term outcomes among a large cohort of patients derived from a multi-institutional international database. In particular, the data demonstrated that tumor necrosis was a tumor-specific factor that negatively impacted both OS and RFS, as well as the pattern of recurrence following surgery. Although the incidence of tumor necrosis increased incrementally with tumor size, necrosis was associated with a worse OS regardless of tumor size. In addition, among patients with T1 tumors, degree of tumor necrosis (no vs moderate vs extensive) was incrementally associated with worsening OS. In fact, tumor necrosis "up-staged" patients with HCC. Specifically, patients with T1 tumor and extensive necrosis had a comparable OS to patients with T2 tumors, while T2 patients with necrosis had an OS the same as T3 patients.

A handful of small studies have reported necrosis incidence of 40–60% among patients with resected intrahepatic cholangiocarcinoma or pancreatic adenocarcinoma.[15,21]

TABLE 3 Multivariable Cox regression analysis of risk factors for overall survival of patients who underwent curative resection for hepatocellular carcinoma

Variables	Model 1*		Model 2*	
	HR (95% CI)	*P*	HR (95% CI)	*P*
Age (years), > 65 vs ≤ 65	1.2 (0.9–1.7)	0.115	1.3 (1.0–1.7)	0.092
Cirrhosis, Yes vs No	1.1 (0.8–1.5)	0.385	1.1 (0.8–1.5)	0.437
Resection margin, R1 vs R0	1.0 (0.6–1.8)	0.914	1.0 (0.6–1.7)	0.984
Macrovascular invasion, Yes vs No	1.6 (0.9–2.9)	0.135	1.8 (1.0–3.4)	0.056
Microvascular invasion, Yes vs No	1.7 (1.2–2.4)	0.007	1.5 (1.0–2.1)	0.031
Tumor grade, poor vs well/moderate	1.5 (1.1–2.0)	0.011	1.6 (1.2–2.2)	0.003
Tumor necrosis				
no necrosis	Reference			
< 50% necrosis	1.2 (0.9–1.7)	0.251		
≥ 50% necrosis	1.7 (1.1–2.6)	0.016		
AJCC 8th T categories				
T1	Reference			
T2	1.2 (0.8–1.8)	0.495		
T3	1.9 (1.1–3.2)	0.017		
New AJCC 8th T categories				
T1n (T1 with no extensive necrosis)			Reference	
T2n (T1 with extensive necrosis + T2 with no extensive necrosis)			1.4 (1.0–2.1)	0.034
T3n (T2 with extensive necrosis + T3)			1.9 (1.2–3.1)	0.007

*In Model 1, the AJCC 8th T categories, as well as extent of tumor necrosis, were included in multivariable analysis; whereas in Model 2, the new T categories incorporating the tumor necrosis (T1n, T2n, T3n), but not AJCC 8th T categories or tumor necrosis, were included in multivariable analysis

Several small studies reported that tumor necrosis was identified among 25–50% of HCC patients.[17,22,23] In the current study, the incidence of tumor necrosis was 39.9%, with roughly 1 in 8 patients having over 50% necrosis. The cause of tumor necrosis may have been due to inadequate blood supply and aberrant tumor angiogenesis, as well as a hypoxic tumor microenvironment.[7] In support of this hypothesis, large tumors were more prone to exhibit necrosis. Several authors have noted that as tumors increase in size, the partial pressure of oxygenation of tissue can decrease, leading to tumor necrosis.[24,25] In fact, tumor necrosis was not only associated with tumor size, but also other traditional aggressive tumor characteristics such as tumor grade, vascular invasion, as well as advanced BCLC stage. In fact, the presence of necrosis was strongly associated with unfavorable OS and RFS. Specifically, patients with moderate and extensive necrosis had an OS and RFS that was incrementally worse with increasing amount of necrosis, providing biologic plausibility associated with a "dose effect" related to necrosis. A recent single-center study similarly reported a negative correlation of tumor necrosis with long-term outcomes among patients who underwent curative resection for small single HCC ≤ 3 cm.[22] While this study only included patients with small single HCC from a single center, and did not evaluate necrotic extent on patient outcomes,[22] the current study analyzed a much large number of patients and examined a broader spectrum of T categories/tumor necrosis using an international multi-institutional database. We also performed additional stratified analyses based on tumor size and the results demonstrated that, even after controlling for tumor size with stratification, extensive tumor necrosis remained strongly associated with adverse outcomes. Specifically, in patients with small tumors ≤ 5 cm, median OS was 60% shorter among patients with extensive necrosis compared with patients who had resection of HCC with no necrosis; similar worse outcomes were noted relative to RFS. In addition, several previous studies that focused on endometrial, breast, and colon cancer have also proposed tumor necrosis as a "new" prognostic factor.[13,26,27] Collectively, the data suggest that tumor necrosis should be considered in the stratification of patients relative to long-term prognosis.

The mechanism underlying the relationship between necrosis and prognosis has been a topic of recent investigation.[7,14,27] Rapidly growing malignant tumors are subject to hypoxia and nutrient deprivation, which can result in necrotic cell death in the core region of the tumor. In turn, necrotic cells release cellular cytoplasmic contents into the extracellular space, including high mobility group box 1

(HMGB1), which is a non-histone nuclear protein that acts as a proinflammatory and tumor-promoting cytokine.[27,28] These molecules subsequently recruit immune and inflammatory cells, which exert tumor-promoting activity by inducing angiogenesis, proliferation, and invasion.[28] Other studies have noted that cell debris produced during necrosis plays an important role in triggering inflammation and reshaping the phenotype of immune cells,[18,29] which further accelerates tumor growth, and remodels the tumor microenvironment to promote immune evasion.[20] A few other studies have reported increased risk of metastasis among patients with tumors characterized by necrosis due to the abnormal structure and function of the neo-angiogenetic vasculature.[30] The aberrant leaky vessels theoretically provide routes for tumor cells to disseminate and metastasize. In addition, the hypoxia in the necrotic microenvironment may enhance metastasis by upregulation of hypoxia-inducible factor 1-α expression, as well as induction of epithelial-mesenchymal transition.[31] Interestingly, patients in the current study who had HCC tumors with extensive necrosis were not only at risk of worse RFS, but were also more likely to recur with an extrahepatic metastatic site of disease as a component of their recurrence. Patients with tumor necrosis also had a higher risk of early recurrence after surgical resection of HCC, as more patients with extensive tumor necrosis recurred within the first year following surgery.

The AJCC TNM staging of HCC incorporates several factors including tumor size, tumor number, and vascular invasion to dictate prognosis.[19] In the current study, we noted that the presence of necrosis on pathology essentially "up-staged" a patient relative to long-term prognosis. Specifically, patients with T1 tumor and extensive necrosis had a comparable OS to patients with T2 tumors, while T2 patients with extensive necrosis had an OS the same as T3 patients. Based on these findings, a modified T classification that incorporated the absence/presence of extensive necrosis was proposed (Table 3). Indeed, necrosis has been incorporated into prediction models for patients for other cancers such as renal clear cell carcinoma in the tumor stage, size, grade, and necrosis (SSIGN) score.[8] In turn, findings from the current study highlight that tumor necrosis should be integrated into the pathological staging of HCC.

Several limitations should be considered when interpreting the results. While the international, multi-institutional-based cohort increased sample size and generalizability, patient selection, surgical procedures, as well as follow-up strategies may have been variable at each center. However, only high-volume academic centers were included in the collaboration—all of which have experienced hepatopathologists who followed standard pathologic assessment guidelines. Given the retrospective nature of the study, there may have been residual collinearity in assessing the prognostic importance of necrosis relative to other factors such as tumor size, vascularity, and grade. We did, however, perform both multivariable as well as stratified analyses by tumor size that demonstrated a strong persistent association of necrosis with prognosis.

In conclusion, tumor necrosis was noted in up to 1 in 5 who underwent curative resection for HCC. Tumor necrosis was associated with worse OS and RFS, as well as increased risk of extrahepatic recurrence. Extensive tumor necrosis represented an independent prognostic predictor of long-term survival. The effect of extensive tumor necrosis persisted on stratified analyses that accounted for tumor size. Importantly, extensive tumor necrosis essentially upstaged patients. A modified AJCC T classification that incorporates tumor necrosis should be considered in the prognostic stratification of patients undergoing resection of HCC.

ACKNOWLEDGEMENT Tao Wei and Xu-Feng Zhang were supported by the Clinical Research Award of the First Affiliated Hospital of Xi'an Jiaotong University of China (No. XJTU1AF-CRF-2017-004).

FUNDING There was no financial support for this study

DISCLOSURE The authors declared no conflict of interest.

REFERENCES

1. Forner A, Reig M, Bruix J. Hepatocellular carcinoma. *Lancet.* 2018;391:1301–14.
2. Tabrizian P, Jibara G, Shrager B, Schwartz M, Roayaie S. Recurrence of hepatocellular cancer after resection: patterns, treatments, and prognosis. *Ann Surg.* 2015;261:947–55.
3. Wei T, Zhang XF, Bagante F, et al. Early versus late recurrence of hepatocellular carcinoma after surgical resection based on post-recurrence survival: an international multi-institutional analysis. *J Gastrointest Surg.* 2020. https://doi.org/10.1007/s11605-020-04553-2.
4. Tsilimigras DI, Bagante F, Moris D, et al. Recurrence patterns and outcomes after resection of hepatocellular carcinoma within and beyond the barcelona clinic liver cancer criteria. *Ann Surg Oncol.* 2020;27:2321–31.
5. Tsilimigras DI, Mehta R, Guglielmi A, et al. Recurrence beyond the Milan criteria after curative-intent resection of hepatocellular carcinoma: a novel tumor-burden based prediction model. *J Surg Oncol.* 2020;122:955–963. https://doi.org/10.1002/jso.26091.
6. Shindoh J, Andreou A, Aloia TA, et al. Microvascular invasion does not predict long-term survival in hepatocellular carcinoma up to 2 cm: reappraisal of the staging system for solitary tumors. *Ann Surg Oncol.* 2013;20:1223–9.
7. Richards CH, Mohammed Z, Qayyum T, Horgan PG, McMillan DC. The prognostic value of histological tumor necrosis in solid organ malignant disease: a systematic review. *Future Oncol.* 2011;7:1223–35.

8. Frank I, Blute ML, Cheville JC, Lohse CM, Weaver AL, Zincke H. An outcome prediction model for patients with clear cell renal cell carcinoma treated with radical nephrectomy based on tumor stage, size, grade and necrosis: the SSIGN score. *J Urol.* 2002;168:2395–400.
9. Brinker DA, Amin MB, de Peralta-Venturina M, Reuter V, Chan DY, Epstein JI. Extensively necrotic cystic renal cell carcinoma: a clinicopathologic study with comparison to other cystic and necrotic renal cancers. *Am J Surg Pathol.* 2000;24:988–95.
10. Gilchrist KW, Gray R, Fowble B, Tormey DC, Taylor SGt. Tumor necrosis is a prognostic predictor for early recurrence and death in lymph node-positive breast cancer: a 10-year follow-up study of 728 Eastern Cooperative Oncology Group patients. *J Clin Oncol.* 1993;11:1929–35.
11. Swinson DE, Jones JL, Richardson D, Cox G, Edwards JG, O'Byrne KJ. Tumour necrosis is an independent prognostic marker in non-small cell lung cancer: correlation with biological variables. *Lung Cancer.* 2002;37:235–40.
12. Richards CH, Roxburgh CS, Anderson JH, et al. Prognostic value of tumour necrosis and host inflammatory responses in colorectal cancer. *Br J Surg.* 2012;99:287–94.
13. Pollheimer MJ, Kornprat P, Lindtner RA, et al. Tumor necrosis is a new promising prognostic factor in colorectal cancer. *Hum Pathol.* 2010;41:1749–57.
14. Atanasov G, Schierle K, Hau HM, et al. Prognostic significance of tumor necrosis in hilar cholangiocarcinoma. *Ann Surg Oncol.* 2017;24:518–25.
15. Atanasov G, Dietel C, Feldbrügge L, et al. Tumor necrosis and infiltrating macrophages predict survival after curative resection for cholangiocarcinoma. *Oncoimmunology.* 2017;6:e1331806.
16. Leek RD, Landers RJ, Harris AL, Lewis CE. Necrosis correlates with high vascular density and focal macrophage infiltration in invasive carcinoma of the breast. *Br J Cancer.* 1999;79:991–5.
17. Atanasov G, Dino K, Schierle K, et al. Angiogenic inflammation and formation of necrosis in the tumor microenvironment influence patient survival after radical surgery for de novo hepatocellular carcinoma in non-cirrhosis. *World J Surg Oncol.* 2019;17:217.
18. Zhang J, Zhang Q, Lou Y, et al. Hypoxia-inducible factor-1α/interleukin-1β signaling enhances hepatoma epithelial-mesenchymal transition through macrophages in a hypoxic-inflammatory microenvironment. *Hepatology.* 2018;67:1872–89.
19. Amin MB. *American Joint Committee on Cancer: AJCC Cancer Staging Manual 8th Edition.* Springer, New York; 2017.
20. Crusz SM, Balkwill FR. Inflammation and cancer: advances and new agents. *Nat Rev Clin Oncol.* 2015;12:584–96.
21. Hiraoka N, Ino Y, Sekine S, et al. Tumour necrosis is a postoperative prognostic marker for pancreatic cancer patients with a high interobserver reproducibility in histological evaluation. *Br J Cancer.* 2010;103:1057–65.
22. Ling YH, Chen JW, Wen SH, et al. Tumor necrosis as a poor prognostic predictor on postoperative survival of patients with solitary small hepatocellular carcinoma. *BMC Cancer.* 2020;20:607.
23. Ha SY, Choi S, Park S, et al. Prognostic effect of preoperative neutrophil-lymphocyte ratio is related with tumor necrosis and tumor-infiltrating lymphocytes in hepatocellular carcinoma. *Virchows Arch.* 2020. https://doi.org/10.1007/s00428-020-02841-5.
24. De Jaeger K, Merlo FM, Kavanagh MC, Fyles AW, Hedley D, Hill RP. Heterogeneity of tumor oxygenation: relationship to tumor necrosis, tumor size, and metastasis. *Int J Radiat Oncol Biol Phys.* 1998;42:717–21.
25. Milross CG, Tucker SL, Mason KA, Hunter NR, Peters LJ, Milas L. The effect of tumor size on necrosis and polarographically measured pO2. *Acta Oncol.* 1997;36:183–9.
26. Maiorano E, Regan MM, Viale G, et al. Prognostic and predictive impact of central necrosis and fibrosis in early breast cancer: results from two International Breast Cancer Study Group randomized trials of chemoendocrine adjuvant therapy. *Breast Cancer Res Treat.* 2010;121:211–8.
27. Bredholt G, Mannelqvist M, Stefansson IM, et al. Tumor necrosis is an important hallmark of aggressive endometrial cancer and associates with hypoxia, angiogenesis and inflammation responses. *Oncotarget.* 2015;6:39676–91.
28. Lee SY, Ju MK, Jeon HM, et al. Regulation of tumor progression by programmed necrosis. *Oxid Med Cell Longev.* 2018;2018:3537471.
29. Chen Q, Wang J, Zhang Q, et al. Tumour cell-derived debris and IgG synergistically promote metastasis of pancreatic cancer by inducing inflammation via tumour-associated macrophages. *Br J Cancer.* 2019;121:786–95.
30. Martin JD, Seano G, Jain RK. Normalizing function of tumor vessels: progress, opportunities, and challenges. *Annu Rev Physiol.* 2019;81:505–34.
31. Jain RK. Antiangiogenesis strategies revisited: from starving tumors to alleviating hypoxia. *Cancer Cell.* 2014;26:605–22.