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Original Article

Impact of different tumor size cut-off points on survival outcome after curative resection of hilar cholangiocarcinoma

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Abstract: Objective: To examine the influence of different tumor size cut-off points on postoperative outcomes following radical resection of hilar cholangiocarcinoma, and to evaluate the possibility of implementing another tumor size cut-off point in new DeOliveira staging system. Methods: A total of 231 patients with hilar cholangiocarcinoma who underwent curative surgery were reviewed retrospectively. Factors associated with survival (especially tumor size) were evaluated by univariate and multivariate analysis. Results: Univariate analysis indicated that survival was inversely proportionate in the case of tumor size 2 cm and 3 cm cut-off (P=0.048 and P < 0.001 respectively), whereas not significant in the case of tumor size 1 cm and 4 cm cut-off (both P > 0.05). Moreover, tumor size 2 cm and 3 cm cut-off was also correlated with other risk factors, with larger size being more likely to be connected with poor differentiation, positive tumor margins and nodal disease. On multivariate analysis, the tumor size 2 cm and 3 cm cut-off was also associated with survival (HR=1.739, 95% CI 1.219-2.482; P=0.002 and HR=2.073, 95% CI 1.410-3.047; P < 0.001 respectively). Conclusion: The tumor size cut-off of 2 cm and 3 cm greatly influenced survival outcome and other prognostic factors than the cut-off of 1 cm and 4 cm. The 2 cm cut-off may become another new potential tumor size cut-off point in addition to the current 1 and 3 cm cut-off in DeOliveira staging system, and may further provide a pivotal reference and guideline for treatment of hilar cholangiocarcinoma.

Keywords: Hilar cholangiocarcinoma, tumor size, DeOliveira staging system, prognosis

Introduction

Hilar cholangiocarcinoma (HCCA), also called Klatskin's tumor, is a rare malignancy [1-3]. Although various chronic inflammations of the biliary duct are known to raise the incidence of bile duct carcinomas, its specific pathogenesis is unknown and most patients present without a peculiar symptom [4-6]. Despite rapid advancement of medical science and surgical technology, the resection of hilar cholangiocarcinoma is still one of the most challenging surgeries performed in hepatobiliary surgery, involving radiological assessment, accompanied by a wide scope of liver resection involving caudate lobectomy, bile duct resection, lymph node dissection, vascular resection, bowel resection, and requiring proper hepato-enteric anastomosis [7, 8]. Thus, involving almost all surgical techniques applied in hepatobiliary surgery.

Hilar cholangiocarcinoma grows in a narrow and pivotal location, surrounded by hepatic blood vessels and bile channels [9, 10]. Tumor located at the confluence of the right and left hepatic bile ducts, has a span less than 5 mm from the portal vein. Combinations of partial hepatectomy or even extensive hepatectomy along with lymphadenectomy and vascular reconstruction is needed to treat HCCA when invasion of portal vein and hepatic artery are present, as hilar cholangiocarcinoma is pathologically characterized by vascular invasion, neural invasion, and early lymph node metastasis [11]. Up to 31%-96% of HCCA cases [12]

presented with caudate lobe invasion and approximately 24%-47% cases [13-15] of lymph node metastasis and about 28%-100% cases [16-19] of perineural invasion. Caudate lobe is one of the most difficult and challenging site for hepatic resection [20]. Furthermore, hilar cholangiocarcinoma has the characteristics of diffusion along the biliary tract mucous membrane [21], which further increases the necessity and difficulty of radical excision. Even so, surgical resection with extended hepatectomy, lymphadenectomy and vascular reconstruction remains the only curative treatment option for cholangiocarcinoma [4, 10, 22-24].

Following radical resection of hilar cholangiocarcinoma, several tumor factors have been identified to impact survival outcome: lymph node status, tumor differentiation, margin status and caudate lobectomy have all been noted to affect survival outcome in several researches [7, 20, 22, 25-28]. Tumor size is also taken in account as an important prognostic factor, as it can directly affect the vascular system and resectability rate. Larger tumor size being more likely to invade the portal vein, the hepatic artery and bile duct system, thus resulting in wider scope of liver resection, with longer perioperative time and higher amount of blood loss. Therefore, as in classification of tumor size in primary liver cancer, the tumor size of HCCA may also act as an important prognostic factor that can affect the classification system and the overall survival outcome. However, current available classification system, the Bismuth-Corlette classification, the AJCC staging system and the Blumgart classification have not included tumor size. DeOliveira staging system has been universally accepted as the classification system for tumor size. However, the DeOliveira staging system is not relatively comprehensive as it has directly regarded tumor size 1 cm and 3 cm as the two cut-off points for explaining prognosis, and not providing specific explanation for survival outcome regarding tumor size 2 cm and 4 cm cut-off points. Furthermore, concerned analysis and literature of the prognosis effect of different tumor size cut-off point on the classification system and survival is inadequate and it still remains controversial.

Thus, the current study aimed to highlight following points: 1) specifically examine the association of different tumor size cut-off point with postoperative outcomes and the survival following radical resection of hilar cholangiocarcinoma, 2) re-evaluate the accuracy and comprehensiveness of tumor size cut-off of 1 cm and 3 cm as suggested by the current DeOliveira staging system, 3) evaluate the likelihood of replenishing another tumor size cut-off point using 2 cm or 4 cm in order to estimate which precise T stage does tumors with 2 cm or 4 cm diameter pertain to, 4) conduct a stratified analysis to evaluate the impact of tumor diameter on resection margin, tumor differentiation and node status so as to provide a pivotal reference for the new classification of T stage in DeOliveira stage system, for guiding the treatment of hilar cholangiocarcinoma and predicting its prognosis following surgery.

Materials and methods

Study patients

231 patients who underwent curative surgery for hilar cholangiocarcinoma from January 1996 to January 2012 at West China Hospital of Sichuan University were the main focus of our study. The overall survival rate of the patients who underwent palliative surgery (e.g. palliative T tube drainage or metallic stent) and the patients who underwent curative surgery were compared. Patients with perioperative mortalities as well as those with carcinoma of the gallbladder, ampulla of the vater, and intrahepatic cholangiocarcinoma were excluded from the study.

Pre-operative work-up

Prior to surgery, patients were evaluated with a detailed medical history and physical exam, imaging studies and serum laboratory tests. Preoperative biliary drainage (PBD) was conducted in patients with cholangitis or obstructive jaundice (bilirubin level > 10 mg/dL) [29]. Biliary drainage was performed until the bilirubin level was reduced, ideally to less than 2 mg/dL. Portal vein embolization (PVE) was mainly carried on patients with compromised liver function, approximately 4-6 weeks prior to the purposed operation date, if the anticipated future remnant volume was less than 30% of the total liver volume. Preoperative biliary drainage and PVE were performed under the guidelines of International Cholangiocarcinoma Association. Resection procedures were selected according to the preoperative and intraoperative assessments. Intraoperative frozen-

Table 1. Patient and operative characteristics

Variable	Patient Characteristics (n=223) Number (%) or median [range]
Age (median [range])	60 [26-82]
Gender	
Male	136 (61.0)
Female	87 (39.0)
Preoperative hospital stay (median [range])	7 [2-44]
Total hospital stay (median [range])	19 [9-113]
Pre-operative CA 19-9 level (U/ml)	348 [0.6-1000]
Pre-operative CA 125 level (U/ml)	19.84 [1.23-257.7]
Pre-operative TB level (mg/dL)	12.25 [0.42-34.3]
Pre-operative ALT level (U/L)	138.3 [10-967]
Preoperative portal vein embolization	32 (14.3%)
Preoperative biliary drainage	110 (49.3%)
Estimated blood loss, median [range]	500 [50-2000]
Postoperative complications	63 (28.2)
Perioperative mortality	8 (3.6)

Percentages exclude missing values. CA-19-9: carbohydrate antigenic determinant 19-9; CA125: carbohydrate antigen 125; TB: total bilirubin; ALT: alanine aminotransferase.

section pathologic examination of bile ducts and liver parenchyma margins was routinely used as a guidance to achieve R0 resection.

Data collection

Standard patients' demographics, clinical data, radiological and histopathologic findings, surgical intervention, operative details, perioperative morbidity and mortality and survival status were obtained retrospectively from hospital's medical database. Tumor diameter was determined on the basis of the information obtained from the pathologic specimen, and was divided into 3 groups: tumor size ≤ 2 cm, tumor size measuring between 2-3 cm and those > 3 cm. Similarly, tumor size ≤ 2 cm and tumor size > 3 cm were further sub-divided into two subgroups: tumor size ≤ 1 cm and tumor size 1-2 cm; tumor size 3-4 cm and tumor size > 4 cm respectively.

Statistical analyses

Statistical calculations were performed using SPSS version16.0. Patient characteristics were reported using frequency and descriptive analysis. Parametric statistical analysis was performed using Student t test, while nonparametric analysis of data was performed using χ^2. Survival (calculated from the time of curative surgery) was computed using Kaplan-Meier methods, and the differences in survival were evaluated using the log-rank test. Prognostic factors in univariate analysis were enrolled in a multivariate Cox proportional hazards model. The hazard ratio (HR) and the 95% confidence intervals (CI) were calculated and $P \leq 0.05$ was considered as significant.

Results

Patients' characteristics

Two hundred and twenty-three of the 231 patients who were operated on with curative surgery had complete histology and survival data available were included in our study. Demographic data and selected characteristics of the 223 patients who underwent curative resection are presented in **Table 1**.

Surgical procedures

Current study includes 32 patients who underwent solely extrahepatic bile duct resection (BDR) and 191 patients underwent hepatectomy with extrahepatic bile duct resection. Hepatic procedures included left hemihepatectomy (n=86), right hemihepatectomy (n=53), left trisegmentectomy (n=21), right trisegmentectomy (n=15), and mesohepatectomy (n=16). Most of the patients that underwent BDR alone were cases that underwent treatment in the first several years of our study or with earlier cases of type I papillary carcinoma. En bloc caudate resection was performed in 190 (85.2%) patients, with a median survival time of 32.1 months following surgery. Most of the caudate lobectomy was performed in the last 8 years of our study. Portal vein resection and reconstruction was performed in 28 (12.6%) patients. Lymphadenectomy was performed in all of these patients.

Postoperative morbidity and mortality

Perioperative morbidity and mortality were deemed as those occurring within 60 days of surgery. The perioperative complication rate was 28.2% (n=63), and the perioperative mortality rate was 3.6% (n=8).

Table 2. Patient, tumor and operative characteristics, stratified by tumor size

Variable	Tumor Size (n=223)				
	≤ 3 cm (n=170)			> 3 cm (n=53)	
	≤ 2 cm (n=91)	2-3 cm (n=79)			
	Number (%)		*P* value†	Number (%)	*P* value‡
Age (median [range])	60 [36-82]	60 [26-75]	0.878	61 [36-73]	0.101
Gender, male	54 (58.2)	49 (62.0)	0.616	33 (63.5)	0.797
Preoperative hospital stay (median [range])	6 [2-33]	7 [3-35]	0.023	8 [2-44]	0.016
Total hospital stay (median [range])	18 [9-56]	19 [11-93]	0.025	22 [10-113]	0.035
Pre-operative CA 19-9 > 100 U/ml					
Yes	68 (74.7)	60 (75.9)	0.854	38 (71.7)	0.857
No	23 (25.3)	19 (24.1)		15 (28.3)	
Bismuth-Corlette classification					
I	36 (39.6)	17 (21.5)	< 0.001	3 (5.7)	< 0.001
II	23 (25.3)	18 (22.8)		13 (24.5)	
IIIa or IIIb	20 (22.0)	26 (32.9)		13 (24.5)	
IV	12 (13.1)	18 (22.8)		24 (45.3)	
Preoperative biliary drainage					
Yes	38 (41.8)	39 (49.4)	0.413	33 (62.3)	0.308
No	53 (58.2)	40 (50.6)		20 (37.7)	
Estimated blood loss, median [range]	509.89 [100-2000]	540.51 [50-2000]	0.082	539.62 [100-1200]	0.190
Receipt > 2 PC during surgery					
Yes	40 (44.0)	41 (51.9)	0.379	27 (50.9)	0.537
No	51 (56.0)	38 (48.1)		26 (49.1)	
Operative time, minutes, median [range]	306.67 [135-720]	322.67 [115-650]	0.458	360.09 [165-605]	0.265
Surgical procedures					
BDR	16 (17.6)	10 (12.6)	0.074	6 (11.3)	0.194
BDR+hepatectomy	75 (82.4)	69 (87.4)		47 (88.7)	
Caudate lobectomy					
Yes	74 (81.3)	71 (89.9)	0.053	45 (85.0)	0.734
No	17 (18.7)	8 (10.1)		8 (15.0)	
Preoperative serum TB > 10 mg/dL					
Yes	37 (46.2)	40 (50.6)	0.421	36 (77.4)	0.501
No	54 (53.8)	39 (49.4)		17 (22.6)	
Differentiation					
Poor	14 (15.4)	19 (24.1)	0.045	25 (47.2)	0.002
Moderate	48 (52.7)	38 (48.1)		17 (32.1)	
Well	29 (31.9)	22 (27.8)		11 (20.7)	
Resection margin					
Positive	6 (6.6)	12 (13.8)	0.037	18 (34.0)	0.011
Negative	85 (93.4)	67 (86.2)		35 (66.0)	
lymph node metastasis					
Yes	32 (36.0)	32 (42.1)	0.048	32 (61.5)	0.012
No	57 (64.0)	44 (57.9)		20 (38.5)	
T stage (AJCC)					
T1/T2	53 (58.2)	31 (39.2)	0.013	26 (49.1)	0.046
T3/T4	38 (41.8)	48 (60.8)		27 (50.9)	
Vascular invasion					
Yes	3 (3.3)	12 (15.2)	0.018	13 (24.5)	0.007
No	88 (96.7)	67 (84.8)		40 (75.5)	
Postoperative complications					
Yes	22 (24.2)	24 (30.4)	0.371	17 (32.1)	0.374
No	69 (75.8)	55 (69.6)		36 (67.9)	

Percentages exclude missing values; †*P* value: comparison between tumor size ≤ 2 cm versus 2-3 cm; ‡*P* value: comparison between tumor size ≤ 2 cm versus 2-3 cm versus > 3 cm; CA-19-9: carbohydrate antigenic determinant 19-9; PC, packed cells; BDR: Bile ducts resection alone; TB: total bilirubin; AJCC: American Joint Committee on Cancer.

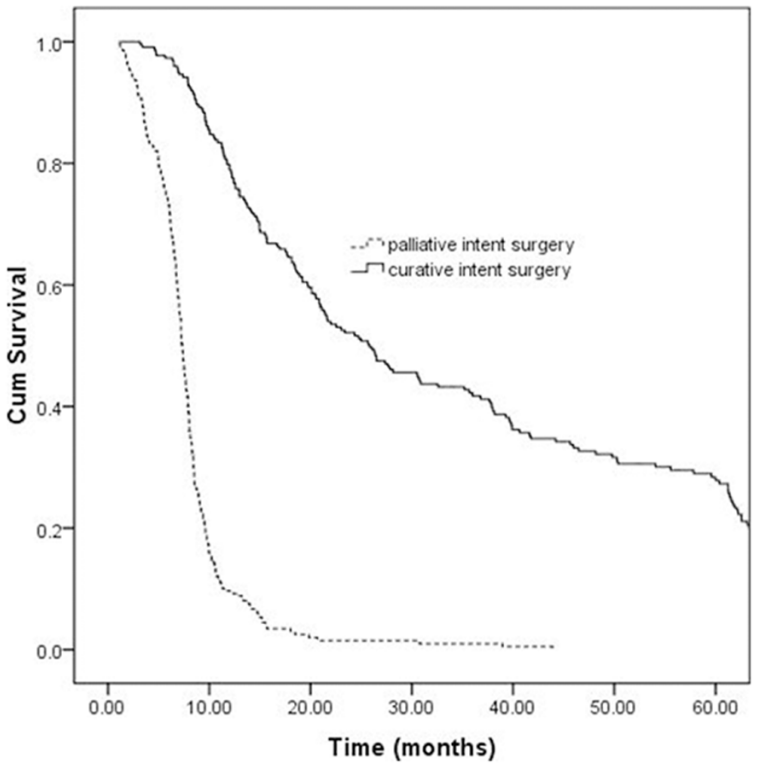

Figure 1. Kaplan-Meier curves comparing survival status between groups based on use of curative surgery and palliative surgery (log-rank test, P < 0.001).

Pathology data

Of these 223 patients treated with curative surgery, pathologic findings were as follows: Overall median tumor diameter was 2.8 cm (range: 0.8-8.0). About 91 (40.8%) patients had a tumor ≤ 2 cm, 79 (35.4%) patients had a tumor 2-3 cm and 53 (23.8%) patients had a tumor > 3 cm (**Table 2**). And in the tumors ≤ 2 cm group, 19 (8.5%) patients had a tumor ≤ 1 cm, while 72 (32.3%) patients had a tumor 1-2 cm. Among tumor diameter > 3 cm, 41 (18.4%) had a tumor 3-4 cm, while 12 (5.4%) had a tumor > 4 cm. As for the tumor grading, almost half of the tumors were moderate-differentiated (n=103, 46.2%). Microscopically negative margins (R0 resection) were obtained in 187 (83.9%) patients and microscopically or macroscopically positive margins (R1 or R2 resection) were seen in 36 (16.1%) patients. With regard to lymph node status, 121 (52.9%) patients did not have lymph node metastasis (N0), while 96 (39.4%) had lymph node metastasis (N1 or N2). Remaining 6 patients could not be evaluated.

Impact of tumor size on operative details and tumor pathological characteristics

Major studies focused on the influence of tumor size on the operative details and pathological findings (**Table 2**). We found that tumor size was directly associated with tumor differentiation, the rate of having well tumor differentiation decreased as the tumor size increased (≤ 2 cm: 31.9%, 2-3 cm: 27.8%, > 3 cm: 20.7% respectively, P= 0.002). Similarly, the rate of lymph node metastasis among patients with tumors ≤ 2 cm was 36% compared with 42.1% among tumors 2-3 cm and 61.5% among tumors > 3 cm (P=0.012). Moreover, surgical margin also correlated with tumor size, the R0 rate among patients with tumors ≤ 2 cm was 93.4% compared with 86.2% among those measuring between 2-3 cm and 66% among those tumors > 3 cm (P=0.011). However there was no statistical difference in tumor grading, node status and tumor margins between tumor size 1-2 cm and ≤ 1 cm (both P > 0.05).

Univariate and multivariate analysis of tumor size on survival

The follow-up for all patients who underwent resection was 96.7%, with a median follow-up period of 21.3 months. For the entire patients who underwent curative surgery for hilar cholangiocarcinoma, the median survival time was 25.8 months, and the 1-, 3-, 5-year survival rate was 79%, 42%, and 28% respectively, while for the patients who were operated on with palliative surgery, the median survival time was 7.3 months (**Figure 1**, P < 0.001).

According to the Kaplan-Meier curves and log-rank test, tumor size was correlated with survival. Specifically, the median survival time and 5-year survival rate of patients with tumors ≤ 2 cm were 39.6 months and 33% respectively, while tumors measuring between 2-3 cm were

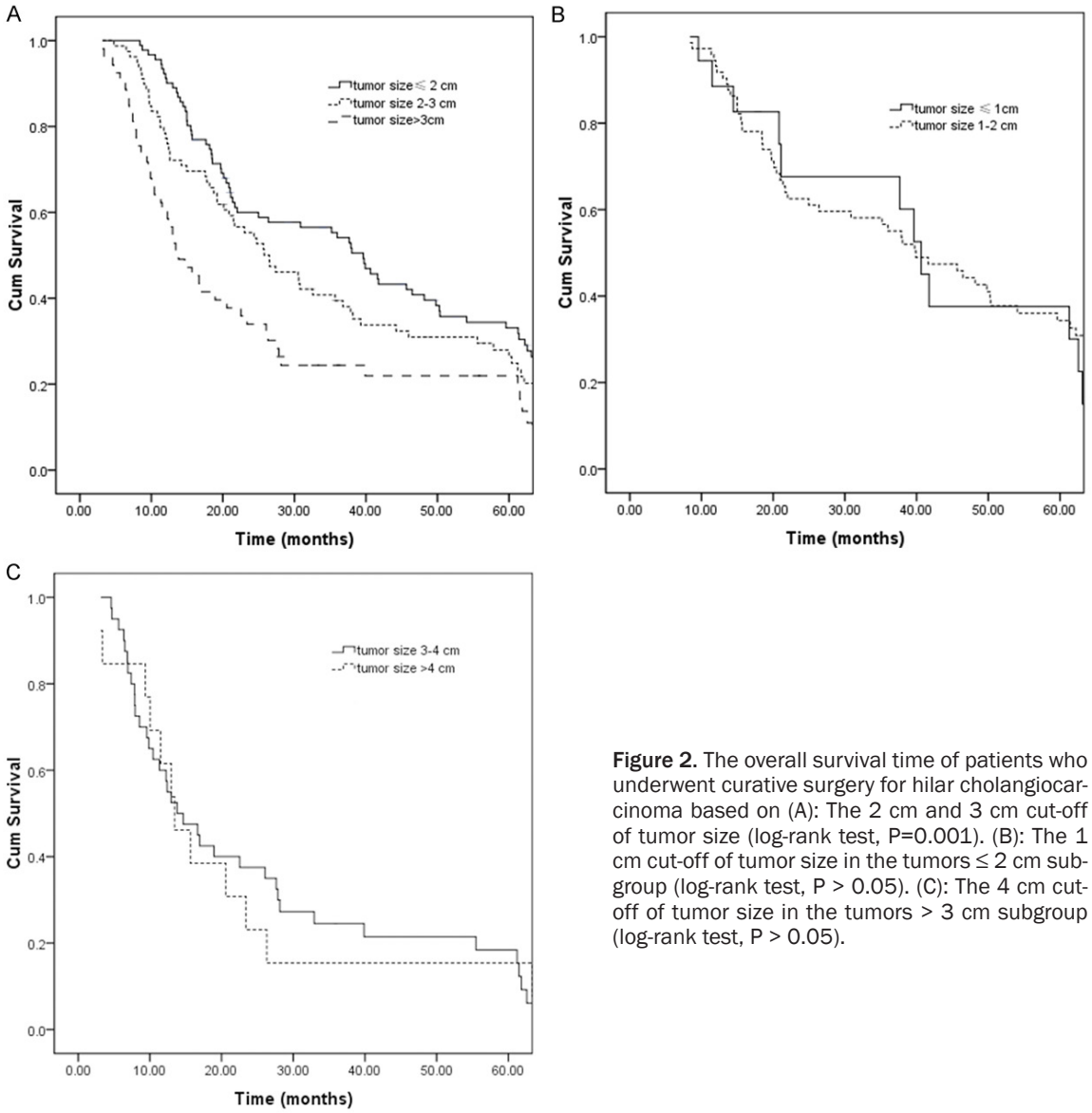

Figure 2. The overall survival time of patients who underwent curative surgery for hilar cholangiocarcinoma based on (A): The 2 cm and 3 cm cut-off of tumor size (log-rank test, P=0.001). (B): The 1 cm cut-off of tumor size in the tumors ≤ 2 cm subgroup (log-rank test, P > 0.05). (C): The 4 cm cut-off of tumor size in the tumors > 3 cm subgroup (log-rank test, P > 0.05).

26.5 months and 28% respectively, and tumors > 3 cm were 13.8 months and 21% respectively (**Figure 2A**, log-rank test, P=0.001). In univariate analysis, when tumor size was investigated as a continuous variable, it correlated with survival (HR=1.424, 95% CI 1.186-1.711; P < 0.001); when censored as a categorical variable, tumor size 2-3 cm and tumor size > 3 cm also correlated with survival (HR=1.357, 95% CI 1.077-1.885; P=0.048 and HR=2.045, 95% CI 1.419-2.949; P < 0.001 respectively, **Table 3**). However, in the subgroup of tumor size ≤ 1 cm and tumor size 1-2 cm, there was no statistically significant difference in survival rate between the two subgroups (HR=0.878, 95% CI 0.445-1.733; P=0.708; **Figure 2B**). In addition, the median survival time and 5-year survival rate was 40.0 months and 33% respectively in patients with tumors ≤ 1 cm compared with 39.1 months and 33% respectively in patients with tumor size 1-2 cm. Similarly, there was also no statistically significant difference in survival rate between the subgroup of tumor size 3-4 cm and tumor size > 4 cm, (HR=1.124, 95% CI 0.812-1.555; P=0.481; **Figure 2C**).

Statistically significant variables in univariate analysis were then considered for multivariate analysis (**Table 4**). In multivariate analysis, the tumor size cut-off of 2 cm and 3 cm independently correlated with survival (HR=1.739, 95% CI 1.219-2.482; P=0.002 and HR=2.073, 95%

Table 3. Univariate analyses of prognostic factors on postoperative survival status

Variables	Hazard ratio	95% CI	*P* value
Age at surgery	1.004	0.991-1.017	0.546
Male gender	0.977	0.729-1.308	0.874
Preoperative hospital stay	1.014	0.985-1.044	0.346
Tumor size			
≤ 2 cm		Reference	
2-3 cm	1.357	1.077-1.885	0.048
> 3 cm	2.045	1.419-2.949	< 0.001
Bismuth-Corlett classification			
I and II		Reference	
III and IV	1.126	0.847-1.495	0.415
CA 19-9 > 100 U/ml	1.212	0.906-1.622	0.195
Surgical procedures			
BDR+hepatectomy		Reference	
BDR	1.034	0.736-1.454	0.847
Positive nodal status	1.885	1.388-2.478	< 0.001
Tumor differentiation			
Well		Reference	
Moderate	1.559	1.103-2.204	0.012
Poor	2.839	1.928-4.180	< 0.001
Positive resection margin	2.053	1.438-2.933	< 0.001
Presence of vascular invasion	1.362	0.991-1.872	0.057
T stage (AJCC)			
T1 and T2		Reference	
T3 and T4	1.182	0.890-1.569	0.248
Caudate lobectomy	1.635	1.008-2.650	0.046
Post-operative complications	1.160	0.860-1.566	0.332

CI: confidence interval; CA-19-9: carbohydrate antigenic determinant 19-9.

Table 4. Multivariate analysis of prognostic factors on postoperative survival status

Variables	Hazard ratio	95% CI	*P* value
Tumor size			
≤ 2 cm		Reference	
2-3 cm	1.739	1.219-2.482	0.002
> 3 cm	2.073	1.410-3.047	< 0.001
Tumor differentiation			
Well		Reference	
Moderate	1.542	1.074-1.821	0.019
Poor	2.732	1.809-4.125	< 0.001
Positive resection margin	2.026	1.354-3.032	0.001
Positive nodal status	1.633	1.205-2.213	0.002

CI: confidence interval.

CI 1.410-3.047; P < 0.001 respectively), showing that tumor size was also an independent factor influencing survival in the multivariate analysis.

Analysis of prognostic factors affecting survival status stratified by tumor size

Poor differentiation, positive resection margin and lymph node metastasis were indicated as independent factors in our current study. Therefore, further stratified analyses were performed focusing on the impact of tumor size on nodal status, excision margin, tumor differentiation and finally on survival.

In our current study, patients with poor differentiation along with tumor size ≤ 2 cm had a median survival time of 21.2 months, while those with poor differentiation accompanied with tumor size 2-3 cm had a median survival time of 13.4 months, simultaneously, those with poor differentiation and tumor size > 3 cm had a median survival time of 10.1 months (**Figure 3A**, P < 0.001). Similarly, in those patients who had positive lymph node metastasis, tumor size ≤ 2 cm conferred to a relatively longer median survival time of 25.0 months, in comparison to those with tumor size 2-3 cm and tumor size > 3 cm with a median survival time of 17.9 months and 9.6 months respectively (**Figure 3B**, P < 0.001). Moreover, the median survival time of those with positive resection margin decreased from 17.6 months to 12.0 months and 7.9 months as we divided tumor size by the 2 cm and 3 cm cut-off (**Figure 3C**, P=0.001). However, in the subgroup of tumor size ≤ 2 cm and tumor size > 3 cm, the cut-off of 1 cm and 4 cm failed to predict statistical difference in the overall survival of patients with poor tumor differentiation/lymph node metastasis/positive resection (both P > 0.05).

In addition, among the 91 patients with tumors ≤ 2 cm, poor tumor differentiation (HR 1.891, 95% CI 1.301-2.746, P=0.001), lymph node metastasis (HR 2.038, 95% CI 1.209-3.437, P=0.008)

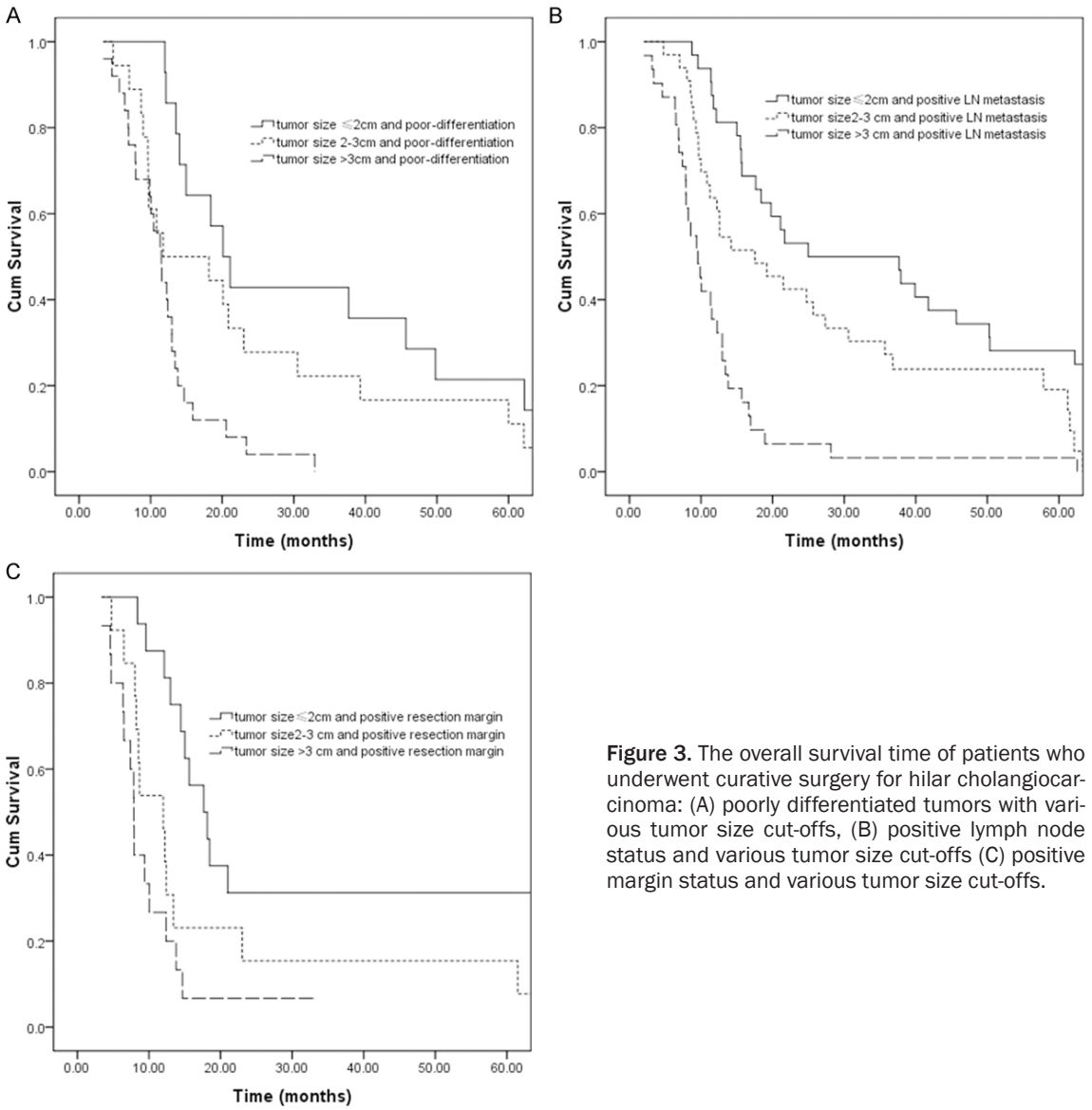

Figure 3. The overall survival time of patients who underwent curative surgery for hilar cholangiocarcinoma: (A) poorly differentiated tumors with various tumor size cut-offs, (B) positive lymph node status and various tumor size cut-offs (C) positive margin status and various tumor size cut-offs.

and positive excision margin (HR 1.876, 95% CI 1.017-3.460, P=0.044) correlated with survival. Among the 79 patients with tumors 2-3 cm, poor tumor differentiation (HR 1.789, 95% CI 1.309-2.444, P < 0.001), lymph node metastasis (HR 2.786, 95% CI 1.634-4.748, P < 0.001) and positive excision margin (HR 8.576, 95% CI 3.978-18.486, P < 0.001) was also associated with survival. And among the 53 patients with tumor size > 3 cm, poor tumor differentiation (HR1.690, 95% CI 1.148-2.490, P=0.008), lymph node metastasis (HR 2.187, 95% CI 1.175-4.071, P=0.014) and positive excision margin (HR 1.969, 95% CI 1.077-3.600, P= 0.028) also predicted statistical relationship with survival. However, in tumor size ≤ 2 cm sub-group, tumor differentiation, resection margin and lymph node metastasis failed to attain statistical difference when we stratified it by tumor size with 1 cm cut-off (both P > 0.05). Similarly, in tumor size > 3 cm sub-group, tumor differentiation, resection margin and lymph node metastasis also failed to attain statistical difference when we stratified it by tumor size with 4 cm cut-off (both P > 0.05).

Discussion

Hilar Cholangiocarcinoma is a technically challenging operation due to its close proximity to major vascular structures and due to arborization of the right and left biliary tree [27, 30].

Previous studies have identified metastasis, positive surgical margins, vascular invasion and poor tumor differentiation as independent negative prognostic factors [7, 22, 31, 32]. Collectively, all these reports emphasized a variety of prognostic factors, but differences in methodology and technology as well as small patient numbers in other studies have produced different and sometimes contradictory results. Kyubo et.al reported tumor size > 2 cm was correlated with inferior distant metastasis-free survival [33]. Moreover, DeOliveira also reported tumor size > 2 cm was associated with survival for hilar cholangiocarcinoma [34]. Thus, the new DeOliveira staging system is not relatively comprehensive as it has directly taken tumor size 1-3 cm as T2, ignoring the role of 2 cm and 4 cm cut-off, and whether the 2 cm and 4 cm cut-off should also be involved into the new staging system has rarely been reported in the literature. So, further researches with larger number of cases are required to precisely discuss the detailed impact of different tumor cut-off on survival outcome and to evaluate the 1 cm and 3 cm cut-off in the DeOliveira staging system.

Aiming to achieve this goal, our study retrospectively reviewed impact of prognostic factors (especially tumor size) on survival outcome. Present study is one of the largest studies focusing on the influence of tumor size on prognosis outcome after successful resection of hilar cholangiocarcinoma in a single institution, aiming to provide a pivotal reference for the new T stage classification in DeOliveira staging system, for better guidance and treatment for hilar cholangiocarcinoma, and thus, accurately predicting its prognosis following surgery. In this study, we detected apparent changes in survival as the primary tumor diameter increased. In univariate analysis, when examined as a categorical variable, comparing with tumor size ≤ 2 cm, tumor size 2-3 cm and tumor size > 3 cm was corresponded with a reduce in median survival time of patients from 39.6 months to 26.5 months and finally to 13.8 months. However in the subgroup of tumor size ≤ 2 cm, the median survival time and 5-year survival rate was 40.0 months and 33% respectively in patients with tumors ≤ 1 cm compared with 39.1 months and 33% respectively in tumor size 1-2 cm and there were no statistical significance between the two groups, showing that compared with 1 cm cut-off, the cut-off of 2 cm and 3 cm was associated with survival and had more clinical value, which was similar to some of the previous results [34, 35]. Our results also indicated that the 2 cm cut-off had greater impact on survival when compared with the 1 cm cut-off, and our result supported that 2 cm cut-off may become another new potential tumor size cut-off point in new DeOliveira staging system in addition to the current 1 cm and 3 cm tumor size cut-off points. After controlling for competing risk factors in multivariate analysis, the 2 cm and 3 cm cut-off of tumor size remained independently associated with survival, concluding that the 2 cm and 3 cm cut-off of tumor size was both a significant and independent factor with survival.

Further analysis of the relationship of tumor size with resection margin, tumor differentiation, node status and the prognosis showed that patients with tumor size ≤ 2 cm along with poor tumor differentiation/lymph node metastasis/positive resection margin manifested better prognosis in comparison to patients with tumor size 2-3 cm and > 3 cm along with poor tumor differentiation/lymph node metastasis/positive resection margin. In the tumor size ≤ 2 cm sub-group, tumor size ≤ 1 cm and 1-2 cm did not show any survival differences in patients who had poor tumor differentiation/lymph node metastasis/positive resection. The tumor size cut-off of 4 cm also did not have significant difference on survival outcome, indicating that compared with the cut-off of 1 cm and 4 cm, the cut-off of 2 cm and 3 cm could affect these prognostic factors and then indirectly affect survival. Hereby, considering the concept that the 2 cm and 3 cm cut-off of tumor size was decidedly associated with these prognostic factors, therefore, explaining the conception that the cut-off of 2 cm and 3 cm would also be a more significant prognostic factor that can indirectly affect the overall survival rate. Thus, according to our analysis and relative reports, 2 cm cut-off may be considered as another new potential tumor size cut-off point in new DeOliveira staging system in addition to the current 1 cm and 3 cm cut-off points.

Currently, there are still debates about selection of the tumor size cut-off in defining the tumor stage of hilar cholangiocarcinoma. The DeOliveira staging system raised up by the

International Cholangiocarcinoma Association have been generally regarded as a full-scale classification system for tumor size, but it is not relatively comprehensive and may be need to be further improvised. In our current study, compared with cut-off of 1 cm, we found that the tumor size 2 cm and 3 cm cut-off was strongly correlated with postoperative survival in both univariate and multivariate analysis. More importantly, the present series confirmed the notion that the new grading system for hilar cholangiocarcinoma proposed by DeOliveira et al. [36, 37] is of great importance, in which tumor size > 3 cm is labelled as T3. And this was similar to some previous studies [36, 38]. However, the current DeOliveira staging system classified tumor size < 1 cm as T1 and tumor size 1-3 cm as T2. On the contrary, in our current study, the 1 cm cut-off failed to predict survival, showing that the 1 cm cut-off as a division point in the new DeOliveira staging system needed to be further investigated and discussed, although one important limitation of our research was that patients with tumor size < 1 cm that underwent curative surgery was relatively few (only 19 cases) and thus, it could not completely prove the rationality of the T1 labeled in the new DeOliveira staging system. More importantly, there were also not adequate clinical materials to support this classification of 1 cm cut-off point, which was associated with a delayed diagnosis and treatment for the eastern hilar cholangiocarcinoma cases. Furthermore, DeOliveira et al. also did not give us the specific reason for choosing tumor size 1 cm as the first dividing point [37]. Therefore, further randomized controlled trials (RCTs) and retrospective studies with 1 cm cut-off are required in future to prove the rationality and reasonability of the 1 cm cut-off. However, our study is still one of the largest studies focusing on the prognostic factor of different tumor cut-off from 1 cm to 4 cm, to investigate survival after successful resection of hilar cholangiocarcinoma in a single institution, including 91, 79, 53 patients with tumor size ≤ 2 cm, 2-3 cm and > 3 cm respectively, and the 2 cm and 3 cm cut-off in our study was simultaneously associated with survival and other adverse factors, indicating that 2 cm cut-off maybe become another new potential tumor size cut-off in the new DeOliveira staging system. In addition, further researches are clearly required to verify the rationality and reasonability of the new classification in respect of tumor diameter.

In conclusion, the tumor size cut-off of 2 cm and 3 cm was more importantly influenced with survival and other prognostic factors than the cut-off of 1 cm and 4 cm. Compared with the 1 cm and 4 cm cut-off, the 2 cm and 3 cm cut-off can strongly correlate with lymph node metastasis, surgical margins, histological differentiation, and then indirectly affect survival outcome. Our results and concerned reports supported that the 2 cm cut-off should be included in the new DeOliveira staging system, and the use of the 1 cm cut-off in the current DeOliveira staging is not immensely suitable for the majority of patients undergoing resection of hilar cholangiocarcinoma. The 1 cm cut-off needs to be verified by more multicenter analysis or larger volume of HCCA cases. The 2 cm and 3 cm as the dividing point is more reasonable than the 1 cm and 3 cm cut-off point in the present DeOliveira staging system. Therefore, 2 cm cut-off should be considered as another new potential tumor size cut-off point in new DeOliveira staging system in addition to the current tumor size cut-off of 1 cm and 3 cm. Nevertheless, further researches and literature are clearly and urgently needed to support the applicability and feasibility of the new staging system about the classification of tumor size to further complete the International bile duct cancer staging system and provide better guidance and treatment of HCCA and thus, accurately predict its prognosis following surgery.

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Disclosure of conflict of interest

None.

Authors' contribution

Hai-Jie Hu contributed to the data acquisition and analysis and drafted the manuscript. Wen-Jie Ma, Jun-Ke Wang and Qin Yang contributed to data acquisition. Anuj Shrestha, Nan-Sheng Cheng and Yong-Qiong Tan were involved in the revision of the manuscript. Fu-Yu Li and Hui Mao contributed to the study design and revision of the manuscript. All authors have read and approved the final version of the manuscript.

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