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***Retrospective Cohort Study***

Prognostic value of lymphovascular invasion in Bismuth-Corlette type IV hilar cholangiocarcinoma

Li B *et al*. Association between lvi and prognosis

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**Abstract**

***AIM***

To investigate the prognostic value of lymphovascular invasion (LVI) in Bismuth-Corlette type IV hilar cholangiocarcinoma (HC) patients.

***Methods***

A retrospective analysis of 142 consecutively recruired type IV HC patients undergoing radical resection with at least a 5-year follow-up. Survival analysis by the Kaplan-Meier method and the association between the clinicopathologic variables and survival was evaluated by log-rank test. Multivariate analysis was adopted to identify the independent prognostic risk factors of overall survival (OS) and disease-free survival (DFS). Multiple logistic regression analysis was performed to determine associations between LVI and potential variables.

***Results***

LVI was confirmed histopathologically in 29 (20.4%) patients. Multivariate analysis showed that positive resection margin (HR = 6.255, 95%CI: 3.485-11.229, *P* < 0.001), N1 stage (HR = 2.902, 95%CI: 1.132-7.439, *P* = 0.027), tumor size > 30 mm (HR = 1.942, 95%CI: 1.176-3.209, *P* = 0.010) and LVI positive (HR = 2.799, 95%CI: 1.588-4.935, *P* < 0.001) is an adverse prognostic factor for DFS. The independent risk factors for OS were positive resection margin (HR = 6.776, 95%CI: 3.988-11.479, *P* < 0.001), N1 stage (HR = 2.827, 95%CI: 1.243-6.429, *P* = 0.013), tumor size > 30mm (HR = 1.739, 95%CI: 1.101-2.745, *P* = 0.018) and LVI positive (HR = 2.908, 95%CI: 1.712-4.938, *P* < 0.001) Furthermore, LVI is associated with N1 stage and tumor size > 30 mm. Multiple logistic regression analysis indicated that N1 stage (HR = 3.312, 95%CI: 1.338-8.198, *P* = 0.026) and tumor size > 30 mm (HR= 3.258, 95%CI: 1.288-8.236, *P* = 0.013)were associated with LVI.

***Conclusion***

LVI interacts with N1 stage and a tumor size > 30 mm and adversely influenced DFS and OS in type IV HC patients.

**Key words:** Hilar cholangiocarcinoma; Bismuth-Corlette classification; Lymphovascular invasion; Overall survival; Disease-free survival

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**Core tip:** Previous studies have reported that lymphovascular invasion (LVI) provoked an adverse impact on the long-term survival of several malignances, including breast, gastric, and esophageal carcinoma, among many others. However, the correlation between LVI and hilar cholangiocarcinoma remains unclear. In our study, LVI is an independent risk factor for overall survival and disease-free survival. To our knowledge, this report indicates for the first time that LVI is an adverse predictor of long-term survival in the setting of type IV hilar cholangiocarcinoma.

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**INTRODUCTION**

Hilar cholangiocarcinoma (HC), also known as Klatskin tumor, is a neoplasia arising from the biliary epithelium at the common hepatic duct bifurcation and may extend to intrahepatic biliary tree and liver[1,2]. The only clinical approach that is considered to provide patients with an opportunity for a curative outcome and importantly long-term survival is radical surgical resection[2-6]. The Bismuth-Corlette classification is the most widely used preoperative system of evaluation that can predict resectability of the lesion and can assist in the design of an appropriate surgical approach. A Bismuth type IV lesion is defined as a tumor that can invade the secondary biliary radicals of both hepatic ducts. During the past decades, accompanied liver resection has been increasingly but gradually recognized as the mainstay of surgical approaches for targeting a Bismuth type IV tumor[7-9], however, there are relatively few studies that have reported in any detail the factors that might affect long-term survival of type IV HC patients. The over-arching aim of the current study was to identify prognostic factors for long-term survival of patients following radical surgery for type IV HC patients, and especially in the setting of lymphovascular invasion (LVI).

**MATERIALS AND METHODS**

***Patients selection***

One hundred and forty-two consecutive patients that underwent radical resection for a pathological diagnosis of type IV HC at the West China hospital between January 2000 and February 2012, were enrolled to this study and then reviewed retrospectively. Inclusion criteria included the following: (1) Bismuth type IV HC patients were confirmed by pathological examination; (2) patients that had undergone radical resection (R0 and R1 resection); The exclusion criteria included the following: (1) patients with gallbladder or intrahepatic cholangiocarcinoma extending to the hilum; (2) presence of a recurrent or metastatic tumor; (3) R2 resection.

***Preoperative workup***

Preoperative assessment consisted of acquiring a medical history, physical examination, laboratory test and radiographic analyses. All patients were evaluated by contrast-enhanced ultrasound or contrast-enhanced computed tomography or magnetic resonance cholangiography with magnetic resonance cholangiopancreatography with the intention of determining the Bismuth type, and the location and extent of the tumor. Biliary drainage, including endoscopic retrograde cholangiopancreatography (ENBD) and percutaneous transhepatic cholangiodrainage (PTCD), was applied in the setting of patients presenting with obstructive jaundice that exceeded 85 μmol/L total bilirubin.

***Surgical characteristics of the patients***

Based on the relative location and extent of the tumor, different types of resection were performed that included, extrahepatic bile duct resection and *en bloc* resection of the caudate lobe combined with left hemihepatectomy, right hemihepatectomy and trisectionectomy. In addition, standard regional lymph node dissection should be performed. However, under conditions where tumor metastases to the distant lymph nodes was confirmed during surgery, then the surgical intervention was abandoned. According to American Joint Committee on Cancer (AJCC, 7th edition), the location of regional lymph nodes defined as follow: along the common bile duct, cystic duct, portal vein and proper hepatic artery[10]. Vascular resection and reconstruction was only performed when vessels could not be detached from the tumor.

***Pathological examination***

The pathological evidence of cancer was determined by examination of paraffin sections. All included Bismuth type IV HC were histopathologically confirmed by an experienced pathologist. The presence of tumor emboli within peritumoural endothelial lined spaces was defined as LVI. Resection margin was defined as ductal (*i.e*., proximal and distal ducts) and with evidence of radial margins. The radial margin was defined as the vertical margin between the tumor edge and dissected periductal structures (*e.g*., liver parenchyma, blood vessels and adjacent fat tissues). An R0 resection was defined as the presence of a microscopically tumor-free resection margin. An R1 resection was defined as microscopic evidence of tumor tissue at the resection margin and an R2 resection was defined as macroscopic evidence of tumor tissue at the resection margin. In this study, radical resection was defined as an R0 and R1 resection, a negative resection margin indicated R0 resection and a positive resection margin indicated an R1 resection.

***Follow-up***

Whether or not chemotherapy and radiotherapy can benefit HC patients was controversial. None of the patients received postoperative routine chemotherapy or radiotherapy. All enrolled patients had routine follow-up every 3 moin the first year and every 6 mo subsequently until at least 5 years after the surgery. The tumor markers [serum levels of carbohydrate antigen 19-9 (CA19-9) and carcinoembryonic antigen], liver functions and ultrasonography were conducted. If there was a suspicion of recurrence, contrast-enhanced computed tomography or magnetic resonance imaging was further performed. Tumor recurrence was diagnosed on the basis of the combined findings of typical radiological appearance, quantification CA19-9 levels, and clinical presentation. The date of the first suspicious radiological finding was recorded as the date of initial disease recurrence.

***Statistical analysis***

Patients data were retrospectively collected and statistical analyses were performed by SPSS version 19.0 (SPSS Inc. Chicago, IL, United States). Survival was described using the Kaplan–Meier method and differences between subgroups were reviewed bythe log-rank test. Multivariate analysis for prognostic factors used a Cox proportional hazards model to analyze variables whose P value was less than 0.1 in the univariate analyses. Multiple logistical regression analysis was performed to determine associations between LVI and potential variables. Two-sided *P* values < 0.05 were considered statistically significant.

**RESULTS**

***Patients’ characters and operative outcomes***

Patients’ characters and operative outcomes are shown in the Table 1. Altogether 142 patients had a radical resection for type IV HC that included 75 men and 67 women with a median age of 59 years (range: 23–78). Pre-operative biliary drainage was carried out in 105 of the 123 obstructive jaundice (total bilirubin > 85 μmol/L) patients, wherein 71 patients underwent PCTD and 34 patients underwent ENBD. Preoperative portal vein embolization was performed in 6 patients.

Radical resection included extrahepatic bile duct resection and en bloc resection of the caudate lobe combined with left-hemihepatectomy (*n* = 73, 51.4%), extended left-hemihepatectomy (*n* = 5, 3.5%), left trisectionectomy (*n* = 6, 4.2%), right-hemihepatectomy (*n* = 51, 35.9%), extended right-hemihepatectomy (*n* = 5, 3.5%), right trisectionectomy (*n* = 2, 1.4%). Regional lymph node dissection was conventionally performed. The R0 resection rate was 75.6%.

***Cliniopathological variables influencing DFS and OS***

As shown in Table 2, potential factors that might influence the DFS and OS were analyzed. Univariate analysis demonstrated that age (*P* = 0.039), preoperative ALB (*P* = 0.005), resection margin (*P* < 0.001), histologic grade (*P* = 0.023), T stage (*P* = 0.004), N stage (*P* < 0.001), AJCC stage (*P* < 0.001), LVI (*P* < 0.001), tumor size (*P* < 0.001), portal vein invasion (*P* = 0.003) and hepatic artery invasion (*P* = 0.008) significantly influenced DFS. By contrast,patient gender, preoperative CA19-9, surgical methods, perineural invasion and transfusion did not significantly influence DFS. Preoperative ALB (*P* = 0.009), resection margin (*P* < 0.001), Histologic grade (*P* = 0.026), T stage (*P* = 0.001), N stage (*P* < 0.001), AJCC stage (*P* < 0.001), LVI (*P* < 0.001), tumor size (*P* < 0.001), portal vein invasion (*P* = 0.002) and hepatic artery invasion (*P* = 0.013), but not patient age, gender, preoperative CA19-9, surgical methods, perineural invasion and transfusion significantly influenced the OS. Multivariate analysis indicated that positive resection margin, a higher N stage, a tumor size > 30 mm and LVI were adverse factors that influenced DFS and OS.

***Association between LVI and N stage and tumor size***

LVI was confirmed histologically in 29 (20.4%) of the 142 patients. Table 3 shows the association of LVI with tumor size, N stage and AJCC stage. On univariate analysis, N stage (*P* < 0.001) and tumor size (*P* = 0.001), but not AJCC stage were significantly associated with LVI. Multivariate analysis using a logistic regression model indicated that N1 stage (*P* = 0.026) and tumor size > 30 mm were significant factors that were associated with LVI in Bismuth type IV HC. DFS and OS based on LVI status are shown in Figure 1. The 5-year DFS rate in the LVI negative group was significantly higher than the LVI positive group (22.8% *vs* 0, *P* < 0.001) as was the OS (23.8% *vs* 0, *P* < 0.001). Based on the LVI status, N0 stage patients and tumor size ≤ 30mm patients were divided into two subgroups respectively. In the N0 stage subgroups, the 5-year DFS rate in the LVI negative group was significantly higher than was found in the LVI positive group (31.0% *vs* 0, *P* = 0.002), the 5-year OS rate in the LVI negative group was also significantly higher than was found in the LVI positive group(32.4% *vs* 0, *P* = 0.001). In the tumor size ≤ 30mm subgroups, the 5-year DFS rate in the LVI negative group was significantly higher than the LVI positive group (31.8% *vs* 0, *P* = 0.003), the 5-year OS rate in the LVI negative group was significantly higher than the LVI positive group(34.3% *vs* 0, *P* = 0.006).

**DISCUSSION**

LVI is recognized as a dismal prognostic factor for OS in patients with breast cancer, colorectal cancer, and esophageal cancer[11-14]. However, no studies have yet been published on whether LVI can affect the prognosis of Bismuth type IV HC have been published. Thus, the current study was undertaken to clarify the significance of LVI in type IV HC patients that had a radical resection. LVI is defined as the involvement of arterial vessels, venules and lymphatic channels[11], but it is histologically difficult to distinguish, and the American Joint Committee on Cancer/Union Internationale Contre Cancer staging guidelines use the term lymphovascular to refer to those structures[11]. Furthermore, LVI can be confirmed specifically on HE-stained specimens[13]. In our series, LVI was confirmed histopathologically in 29 patients.

It is well recognized thatresection margin status is the most important factor affecting long-term survival outcomes of HC patients[14,15], and positive resection margin remains a majordismal prognostic factor for type IV HC patients. Furthermore, since type IV HC had been considered unresectable[16], radical resection performed in such patients is technically challenging. Bracingly, in our center the R0 resection rate is 75.4%. Regional lymph node involvement represents another important dismal prognostic factor in HC patients that had undergone radical resection[17,18]. Concordantly, the multivariate analysis showed that lymph node metastases is an adverse factor affecting DFS and OS of type IV HC. Furthermore, DeOliveira *et al*[19] proposed a new staging system, in which tumor size > 30 mm was defined as the T3 stage. The choice of 30 mm as a cutoff value for T3 is based on increasing evidence that the smaller the tumor, the better the prognosis[20,21]. Our results indicated that tumor size > 30 mm had an unfavorable impact on both DFS and OS. More importantly, we found that LVI is a significant adverse prognostic factor influencing DFS and OS in multivariate analysis. To our knowledge, no other reports have shown the correlation between LVI and the prognosis of type IV HC.

Previous studies suggested that LVI may interact with other adverse risk factors, which then have a dismal impact on the OS of esophageal cancer patients[22,23]. Lee *et al*[24] found that the presence of LVI correlated with the presence of lymph node metastasis for patients with gastric carcinoma. For different types of cholangiocarcinoma, the effect of LVI on prognosis was also different. Kim *et al*[25]reported that LVI did not influence the survival of patients with distal cholangiocarcinoma. Fisher *et al*[26] reported that LVI had an adverse influence on survival of patients with intrahepatic cholangiocarcinoma. Both of studies found that LVI was associated with lymph node metastasis[25,26]. In our analysis, N1 stage was a significant factor that was associated with LVI in type IV HC patients. It is generally known that tumor cells and tumor stromal cells (such as macrophages and thrombocytes) can produce pro-lymphangiogenic factors, which increase lymphovascular density in and around the tumors[27,28], mainly peritumoral regions. Lymph node metastases were often occurred in tumors lacking intratumoral functional lymphatics, suggesting that functional lymphatics at the peritumoral regions are the route of lymphatic dissemination. Increased peritumoral lymphovascular density is considered to increase the flow of lymphatic fluid and provided an opportunity for invasive tumor cells access the lymphatic vessels[29]. This may elucidate our results that LVI is closely related to lymph node metastasis. Moreover, we speculated that LVI may be the precursor of lymph node metastasis. Thus, we divided patients without lymph nodes metastases into LVI positive and LVI negative subgroups, the 5-year DFS and OS rates in the LVI negative group were significantly higher than were found in the LVI positive group (Figure 2A and B). The results indicated that LVI is an admirable prognostic predictor for patients with type IV HC when lymph node metastasis is absent.

Additionally, a tumor size > 30 mm was another significant factor correlated with LVI in our series. Tumor size is recognized as a staging basis for many malignant tumors, including thyroid carcinoma, breast carcinoma, and liver carcinoma among others. Gurleyik *et al*[30] reported that LVI correlated with tumor size, and the rate of LVI positive increased with tumor size in patients with breast carcinoma. The significant correlation between LVI and tumor size could be explained through two potential aspects: (1) as the tumor size increases, the peritumoral areas increase. Thus, the tumor is endowed with the potential to make contact with an increasing lymphovasculature, and thus the possibility of LVI increases. And (2) tumor size is proportional to the time of growth: the larger the tumor, the greater is the duration of time that the tumor can continuing developing and growing in size. During a relatively longer growth time, a tumor has increasing opportunities to develop LVI. In our study, patients with a tumor size ≤ 30 mm were also divided into LVI positive and LVI negative subgroups. Here, the 5-year DFS and OS rates in the LVI negative group were significantly higher than those found in the LVI positive group (Figure 3A and B). The results showed that LVI is also an excellent prognostic predictor for a smaller tumor size of type IV HC.

Some limitations of the study should also be taken into account when interpreting the results. First, our study was retrospective with inherent limitations in its design. Thus, some clinical bias was inevitable. Next, LVI was confirmed by a hematoxylin and eosin (HE)-stain alone without application of an immunohistochemical stain with D2-40 antibody, which may improve the detection rate of LVI[31]. Third, N1 stage and a tumor size > 30 mm might serve as potential confounding factors that could affect the assotiation between LVI and the eventual prognosis. Finally, we did not carry out preclinical medical experiments to elaborate the specific molecular mechanism that could play a key mode of action in the capacity of LVI to affect the prognosis of type IV HC patients. Future research should take into account this topic with a greater sample size. Prospective studies, even randomized controlled trials, are also urgently needed. Of course, the specific molecular mechanism responsible for LVI affecting the prognosis of type IV HC patients will be determined by emprical research.

In conclusion, the presence of LVI may be regarded as an indicator of biologically aggressive behavior, of metastatic ability, and of a regional and systemic risk of metastasizing the primary malignancy. LVI can interact with the N1 stage and a tumor size > 30 mm and will impart an adverse influence on OS and DFS in patients that present with type IV HC that received radical resection.

**COMMENTS**

***Background***

Despite advances in surgical techniques, resection rates for Bismuth type IV hilar cholangiocarcinoma (HC) continues to increasing, the prognosis of patients that present with type IV HC remains unsatisfactory. The reasons for this remain unclear and seem to be complex and multifactorial. Lymphovascular invasion (LVI) is associated with a poorer prognosis in patients with various malignances. The study set out to investigate whether LVI could predict type IV HC prognosis.

***Research frontiers***

This study estimates prognostic factors that might be associated with overall survival (OS) and disease-free survival (DFS) after radical resection in type IV HC patients. To our knowledge, this study represents the first clinical insight indcating that LVI is associated with the prognosis of type IV HC.

***Innovations and breakthrough***

Our findings confirmed that R1 resection, N1 stage, presence of LVI and tumor size > 30 mm were adverse prognostic predictors for type IV HC patients after radical resection. Further, LVI was found to interact with an N1 stage and a tumor size > 30 mm and adversely influence OS and DFS.

***Applications***

Observations from the present study might formally indicate novel factors for predicting post-surgical survival in HC. Moreover, LVI might present a potentially novel target for developing of anti-cancer strategies.

***Terminology***

HC is a neoplasia arising from the biliary epithelium at the common hepatic duct bifurcation that might extend to the intrahepatic biliary tree and liver. Bismuth-Corlette classification is the most commonly used HC typing system, which is often used by surgeons to develop preliminary surgical protocols.

***Peer-review***

This is a retrospective study evaluating the effect of LVI on the prognosis of Bismuth-Corlette type IV HC. The authors concluded that LVI had an adverse influence on the prognosis of patients with Bismuth-Corlette type IV HC. this manuscript is very interesting and well written.

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**Table 1 Demographics and operative outcomes**

|  |  |
| --- | --- |
| **Characteristics** | ***n* (%)** |
| Age (yr)1 | 59 (23-78) |
| Gender |  |
| Male | 75 (52.8) |
| Female | 67 (47.2) |
| Albumin level (g/L)2 | 36.74 ± 4.93 |
| CA19-9 |  |
| > 200 | 82 (57.7) |
| < 200 | 60 (42.3) |
| Radiological examination |  |
| contrast-enhanced US | 8 (5.6) |
| contrast-enhanced CT | 43 (30.3) |
| MRI + MRCP | 91 (64.1) |
| Operation time (min)2 | 429.47 ± 134.19 |
| Blood loss (ml)1 | 600 (180-4000) |
| Transfusion | 76 (53.5) |
| R0 resection | 107 (75.4) |
| Differentiation degree |  |
| Well differentiated | 14 (9.9) |
| Moderately differentiated | 90 (63.4) |
| poorly differentiated | 38 (26.8) |
| Perineutral invasion positive | 69 (48.6) |
| N stage |  |
| N0 | 89 (62.7) |
| N1 | 53 (37.3) |
| Tumor diameter (mm) 1 | 30 (12-55 |
| Hospital stay1 | 19 (5-115) |

1Parameters are presented as median and range; 2Parameters are presented as mean ± SD.

**Table 2 The potential prognostic factors for disease-free survival and overall survival after radical resection for type IV hilar cholangiocarcinoma patients excluding operative mortality *n* (%)**

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Variables** | **Disease-free survival** | | | | **Overall survival** | | | | |
| **5-yr survival** | **Univariate analysis**  ***P* value** | **Multivariate analysis** | | **5-yr survival** | **Univariate analysis**  ***P* value** | **Multivariate analysis** | | |
| **HR (95%CI)** | ***P* value** | **HR (95%CI)** | ***P* value** | |
| Gender |  | 0.682 |  |  |  | 0.965 |  |  | |
| Male | 13 (18.8) |  |  |  | 14 (20.3) |  |  |  | |
| Female | 10 (16.4) |  |  |  | 10 (16.4) |  |  |  | |
| Age (yr) |  | 0.039 | 1.291 (0.680-2.453) | 0.435 |  | 0.230 |  |  | |
| < 65 | 19 (19.2) |  |  |  | 20 (20.2) |  |  |  | |
| > 65 | 4 (12.9) |  |  |  | 4 (12.9) |  |  |  | |
| CA19-9 |  | 0.287 |  |  |  | 0.301 |  |  | |
| < 200 | 13 (23.2) |  |  |  | 14 (25) |  |  |  | |
| > 200 | 10 (13.5) |  |  |  | 10 (13.5) |  |  |  | |
| ALB (g/L) |  | 0.005 | 0.669 (0.413-1.085) | 0.103 |  | 0.009 | 0.760 (0.492-1.173) | 0.215 | |
| < 35 | 3 (7.1) |  |  |  | 3 (7.1) |  |  |  | |
| > 35 | 21 (23.9) |  |  |  | 20 (22.7) |  |  |  | |
| Surgical methods |  | 0.847 |  |  |  | 0.684 |  |  | |
| Left-sided hepatectomy | 13 (15.1) |  |  |  | 13 (15.1) |  |  |  | |
| Right-sided hepatectomy | 10 (17.2) |  |  |  | 11 (19.0) |  |  |  | |
| Resection margin |  | < 0.001 | 6.255 (3.485-11.229) | < 0.001 |  | < 0.001 | 6.776 (3.988-11.479) | < 0.001 | |
| Positive | 0 |  |  |  | 0 |  |  |  | |
| Negative | 23 (23.7) |  |  |  | 24 (24.7) |  |  |  | |
| Histologic grade |  | 0.023 | 1.594 (0.994-2.554) | 0.053 |  | 0.026 | 1.294 (0.830-2.017) | 0.256 | |
| Well/moderate | 20 (21.5) |  |  |  | 21 (22.6) |  |  |  | |
| poor |  |  |  |  | 3 (8.1) |  |  |  | |
| Perineural invasion |  | 0.211 |  |  |  | 0.417 |  |  | |
| Present | 14 (22.6) |  |  |  | 15 (24.2) |  |  |  | |
| Absent | 9 (13.2) |  |  |  | 9 (13.2) |  |  |  | |
| T stage |  | 0.004 | 1.582 (0.390-6.415) | 0.521 |  | 0.001 | 2.399 (0.734-7.836) | 0.147 | |
| T 1,2 | 22 (22) |  |  |  | 21 (21) |  |  |  | |
| T3,4 | 2 (6.7) |  |  |  | 2 (6.7) |  |  |  | |
| N stage |  | < 0.001 | 2.902 (1.132-7.439) | 0.027 |  | < 0.001 | 2.827 (1.243-6.429) | 0.013 | |
| N0 | 22 (27.2) |  |  |  | 23 (28.4) |  |  |  | |
| N1 | 1 (2.0) |  |  |  | 1 (2.0) |  |  |  | |
| AJCC stage |  | < 0.001 | 0.673 (0.289-1.567) | 0.358 |  | < 0.001 | 0.844 (0.351-2.028) | 0.704 | |
| Stage Ⅰ,Ⅱ | 20 (32.8) |  |  |  | 21 (34.4) |  |  |  | |
| Stage Ⅲ,Ⅳ | 3 (4.9) |  |  |  | 3 (4.9) |  |  |  | |
| Lymphovascular invasion |  | < 0.001 | 2.799 (1.588-4.935) | <0.001 |  | < 0.001 | 2.908 (1.712-4.938) | < 0.001 | |
| Present | 0 |  |  |  |  | 0 |  |  |
| Absent | 23 (22.8) |  |  |  | 24 (24.8) |  |  |  | |
| Tumor size (mm) |  | < 0.001 | 1.942 (1.176-3.209) | 0.010 |  | < 0.001 | 1.739 (1.101-2.745) | 0.018 | |
| ≤ 30 | 15 (20) |  |  |  | 16 (21.3) |  |  |  | |
| > 30 | 8 (14.5) |  |  |  | 8 (14.5) |  |  |  | |
| Portal vein invasion |  | 0.003 | 1.759 (0.534-5.800) | 0.353 |  | 0.002 | 1.130 (0.408-3.127) | 0.815 | |
| Present | 1 (4.2) |  |  |  | 1 (4.2) |  |  |  | |
| Absent | 22 (20.8) |  |  |  | 23 (21.7) |  |  |  | |
| Hepatic invasion |  | 0.008 | 1.499 (0.612-3.668) | 0.376 |  | 0.013 | 1.196 (0.526-2.719) | 0.669 | |
| Present | 1 (9.1) | 1 (9.1) |  |  | 1 (9.1) |  |  |  | |
| Absent | 22 (18.5) | 22 (18.5) |  |  | 23 (19.3) |  |  |  | |
| Transfusion |  | 0.445 |  |  |  | 0.199 |  |  | |
| Yes | 11 (16.4) |  |  |  | 11 (16.4) |  |  |  | |
| No | 12 (19.0) |  |  |  | 13 (20.6) |  |  |  | |

**Table 3 Logistic regression analyses for factors associated with lymphovascular invasion**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Variable** | **Lymphovascualr invasion (+), *n* = 29** | **Lymphovascualr invasion (-), *n* = 113** | **Univariate analysis (*P* value)** | **OR (95%CI)** | **Multivariate analysis (*P* value)** |
| AJCC stage |  |  | 0.063 | 0.223 (0.026-1.927) | 0.173 |
| I, II | 8 | 27 |  |  |  |
| III, IV | 21 | 86 |  |  |  |
| Tumor diameter (mm) |  |  | 0.001 | 3.258 (1.288-8.236) | 0.013 |
| ≤ 30 | 10 | 77 |  |  |  |
| > 30 | 19 | 36 |  |  |  |
| N stage |  |  | < 0.001 | 3.312 (1.338-8.198) | 0.026 |
| N0 | 10 | 79 |  |  |  |
| N1 | 19 | 34 |  |  |  |





**Figure 1 Disease-free survival and overall survival based on lymphovascular invasion in type IV hilar cholangiocarcinoma patients with radical resection**. A: the 5-yr DFS rate in the LVI negative group were significantly higher than the LVI positive group; B: the 5-yr OS rate in the negative group were significantly higher than the LVI positive group. LVI: lymphovascular invasion; DFS: Disease-free survival; OS: overall survival.





**Figure 2 Disease-free survival and overall survival based on lymphovascular invasion in type IV hilar cholangiocarcinoma patients without lymph nodes metastases.** A: the 5-yr DFS rate in the LVI negative group were significantly higher than the LVI positive group; B: the 5-yr OS rate in the negative group were significantly higher than the LVI positive group. LVI: lymphovascular invasion; DFS: Disease-free survival; OS: overall survival.





**Figure 3 Disease-free survival and overall survival based on lymphovascular invasion in type IV hilar cholangiocarcinoma patients with tumor size ≤ 30 mm.** A. the 5-yr DFS rate in the LVI negative group were significantly higher than the LVI positive group; B: the 5-yr OS rate in the negative group were significantly higher than the LVI positive group. LVI: lymphovascular invasion; DFS: Disease-free survival; OS: overall survival.