European Journal of Surgical Oncology 45 (2019) 1446–1452

Contents lists available at ScienceDirect

European Journal of Surgical Oncology

journal homepage: www.ejso.com

Lymphatic spread of T2 gallbladder carcinoma: Regional lymphadenectomy is required independent of tumor location

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ARTICLE INFO

Article history:
Received 30 September 2018
Received in revised form
19 March 2019
Accepted 27 March 2019
Available online 30 March 2019

Keywords:
Gallbladder neoplasms
Tumor location
Lymphatic metastasis
Lymph node excision
Lymphatic vessels
Survival rate

ABSTRACT

Background: This study aimed to investigate the incidence and distribution of regional lymph node metastasis according to tumor location, and to clarify whether tumor location could determine the extent of regional lymphadenectomy in patients with pathological T2 (pT2) gallbladder carcinoma.
Methods: In total, 81 patients with pT2 gallbladder carcinoma (25 with pT2a tumors and 56 with pT2b tumors) who underwent radical resection were enrolled. Tumor location was determined histologically in each gallbladder specimen.
Results: Survival after resection was significantly worse in patients with pT2b tumors than those with pT2a tumors (5-year survival, 72% vs. 96%; $p = 0.027$). Tumor location was an independent prognostic factor on multivariate analysis (hazard ratio, 14.162; $p = 0.018$). The incidence of regional lymph node metastasis was significantly higher in patients with pT2b tumors than in those with pT2a tumors (46% vs. 20%; $p = 0.028$). However, the number of positive nodes was similar between the two groups (median, 2 vs. 2; $p = 0.910$). For node-positive patients with pT2b tumors, metastasis was found in every regional node group (12%–63%), whereas even for node-positive patients with pT2a tumors, metastasis was observed in regional node groups outside the hepatoduodenal ligament.
Conclusions: Tumor location in patients with pT2 gallbladder carcinoma can predict the presence or absence of regional lymph node metastasis but not the number and anatomical distribution of positive regional lymph nodes. The extent of regional lymphadenectomy should not be changed even in patients with pT2a tumors, provided that they are fit enough for surgery.

Introduction

Surgical outcomes of gallbladder carcinoma strongly reflect the depth of tumor penetration [1–3]. Patients with advanced gallbladder carcinoma generally have a dismal prognosis. However, cure can be expected for patients with pathological T2 (pT2) gallbladder carcinoma, which is defined as tumor invading the perimuscular (subserosal) connective tissue without extension beyond the serosa or into the liver. For these patients, radical resection with regional lymphadenectomy yields a 5-year survival of 40–83% [2–10]. Because gallbladder carcinoma is known to have a high propensity for lymphatic spread and the pT2 primary tumor itself is limited to the gallbladder wall, its main mode of spread outside the confines of the gallbladder for pT2 gallbladder carcinoma is lymphatic [5,9,11]. In fact, around 40% of patients with pT2 gallbladder carcinoma have lymph node metastasis, indicating that lymphatic spread occurs before the tumor invades into adjacent organs. Therefore, lymphadenectomy is a critical component of oncologic resection for this disease entity [2,6–10].

The tumor-node metastasis (TNM) staging system developed by the American Joint Committee on Cancer (AJCC) eighth edition has subdivided pT2 gallbladder carcinoma into two categories according to the location of the primary tumor: pT2a (tumor on the peritoneal side) and pT2b (tumor on the hepatic side) [12]. It has been reported that survival after resection was significantly worse in patients with pT2b tumors than in those with pT2a tumors [13–15]. A putative cause of this is that the incidence of lymph node metastasis differed between the two groups. Recent studies have shown that patients with pT2b tumors have a higher incidence of

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https://doi.org/10.1016/j.ejso.2019.03.038
0748-7983/

lymph node metastasis than those with pT2a tumors [13,14]. However, it remains unclear whether tumor location predicts the anatomical distribution of lymphatic spread and why patients with pT2b tumors tend to have a higher incidence of lymph node metastasis.

The aims of this study were to investigate the incidence and anatomical distribution of lymph node metastasis according to tumor location in patients with pT2 gallbladder carcinoma, and to clarify whether the extent of lymphadenectomy could be determined from tumor location. Another goal of this study was to investigate whether the wall on the hepatic side of the normal gallbladder contains more lymphatic vessels than that on the peritoneal side for explaining the difference in the incidence of lymph node metastasis between patients with pT2a and pT2b tumors.

Patients and methods

Patient selection

This single-institution study enrolled consecutive patients who underwent radical resection—resection of both the primary tumor and regional lymph nodes—for pT2 gallbladder carcinoma, at the Niigata University Medical and Dental Hospital from May 1982 through June 2017. Patients who underwent radical second resection for incidental pT2 gallbladder carcinoma were included. Patients with an invasive primary malignancy in other organs were excluded. The approval of the Institutional Review Board was obtained for this study.

Resection procedures

The radical resection procedures depended on the extent of tumor spread in each patient. For pT2 gallbladder carcinoma, an extended radical cholecystectomy, in principle, was indicated [5,9]. This procedure consists of cholecystectomy, wedge resection of the gallbladder fossa, resection of the extrahepatic bile duct, and regional lymphadenectomy. Wedge resection, bile duct resection, or both procedures were omitted in some patients with advanced age or comorbid disease [5,16]. Pancreaticoduodenectomy was indicated for tumor with bulky peripancreatic nodal metastases [5,16]. For some patients with pT2 gallbladder carcinoma, major hepatectomy (resection of $\geq$3 Couinaud segments) and/or pancreaticoduodenectomy was performed because pT3 or pT4 tumor was not ruled out before surgery due to severe inflammation from cholecystitis.

In our department, in principle, the cystic duct, pericholedochal, posterior superior pancreaticoduodenal, retroportal, and common and proper hepatic artery lymph node groups were dissected en bloc for pT2 gallbladder carcinoma [5,9,16,17]. In this study, these lymph node groups were treated as regional lymph nodes according to the Japanese TNM staging system [18]. Less extensive lymphadenectomy, in which the pericholedochal and cystic duct node groups were mainly removed, was performed for some patients with advanced age or comorbid disease at the discretion of individual surgeons [16]. In this series, patients who were suspected to have regional lymph node metastases also underwent dissection or sampling of the paraaortic lymph nodes [5,16,17].

Histological examination

The depth of tumor penetration and the location of pT2 tumor were evaluated based on histological examination of multiple sections (median, 29; interquartile range [IQR], 20–37) in each gallbladder specimen. Tumors infiltrating only the free serosal side of the gallbladder wall were classified as being located on the peritoneal side (pT2a tumors; Fig. 1A), whereas at least part of tumors infiltrating the hepatic aspect of the gallbladder wall were classified as being located on the hepatic side (pT2b tumors; Fig. 1B) [12,13]. Histological findings were documented according to the AJCC TNM staging system [12]. Immediately after removal, lymph nodes dissected during surgery were retrieved from the resected specimens and grouped according to the anatomical location by the surgeons. These lymph nodes were assessed histologically for metastasis in a representative section stained with hematoxylin and eosin.

Follow-up after resection

For all patients discharged home, recurrence was regularly monitored at outpatient clinics. Computed tomography or ultrasonography was performed about every 6 months for at least 5 years. Adjuvant chemotherapy was administered mainly for patients with AJCC Stage III or more advanced disease at the discretion of individual surgeons. After around 2007, gemcitabine or tegafur/gimeracil/oteracil potassium (S-1) was used as adjuvant chemotherapy. Before that, oral administration of 5-fluorouracil (5-FU) or its derivatives (other than S-1) and intravenous administration of 5-FU were the main options.

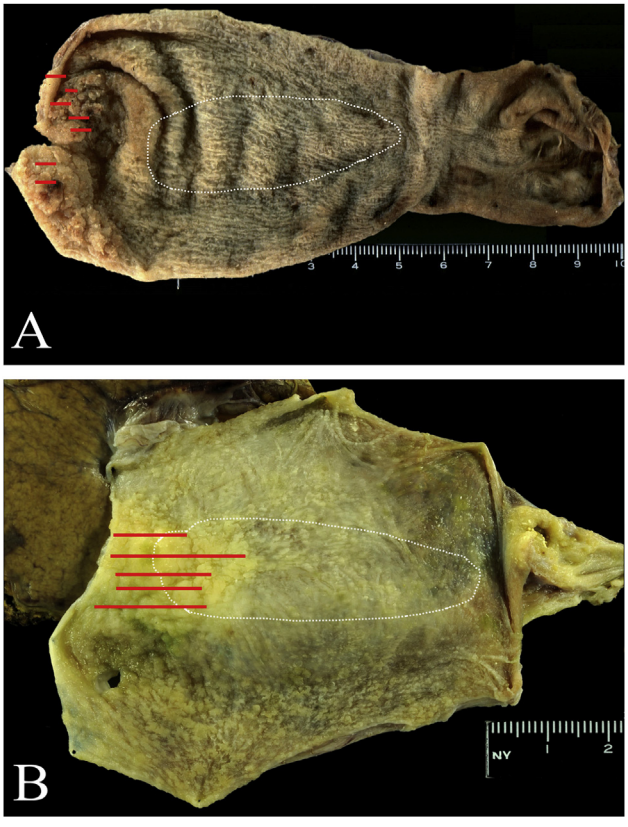

Fig. 1. Classification of pathological T2 (pT2) gallbladder carcinoma according to tumor location [12]. A. pT2a (peritoneal-sided) tumor. Tumor invades only the free serosal side of the gallbladder wall. B. pT2 (hepatic-sided) tumor. At least part of the tumor invades the hepatic side of the gallbladder wall. Red lines indicate the area of carcinoma invasion into perimuscular (subserosal) connective tissue in the gallbladder. A white dotted circle indicates the area of the hepatic side of the gallbladder wall.

Lymphatic vessel density (LVD) according to location in the normal gallbladder

Normal gallbladder specimens were obtained from 10 patients with colorectal carcinoma liver metastasis who underwent partial liver resection combined with cholecystectomy from 2015 to 2017 at our hospital. None of the liver tumors involved the gallbladder. There were neither mucosal lesions nor gallstones in any of the gallbladders. Two longitudinal sections were cut from each gallbladder specimen and embedded in paraffin. One was a section of the hepatic side of the gallbladder, which passed through the center of the area where the gallbladder is attached to the liver. The other was a section of the peritoneal side, which passed through the free peritoneal side of the gallbladder.

The paraffin-embedded blocks of the sections were used for immunohistochemistry. Two serial 3-μm sections were cut from each block: one for routine histologic examination with hematoxylin and eosin staining, the other for immunohistochemical staining with a specific marker for lymphatic vessels (D2-40 monoclonal antibody, Signet Laboratories, Inc., Dedham, MA; 1:200 dilution). LVD was defined as the number of lymphatic vessels per mm^2; a high-power (× 200) field with the highest LVD in an area was referred to as a "hot spot", in accordance with previous studies [19]. The sections stained with the D2-40 antibody were examined for LVD. In each section at low power (× 20), 5 hot spots were identified. To measure LVD, the outlines of individual immunohistochemically stained lymphatic vessels were identified and traced using a computer-aided image analysis system at × 200 magnification in individual hot spots [19].

Statistical analysis

Statistical calculations were performed using the IBM SPSS Statistics 24 software (IBM Japan, Inc., Tokyo, Japan). Variables were compared using the Mann-Whitney *U* test or Fisher's exact test, as appropriate. Disease-specific survival (DSS) was measured from the date of definitive surgery to that of the last follow-up or death from cancer; cases involving deaths from other causes were censored in this analysis. Survival curves were generated using the Kaplan-Meier method and differences between survival curves were assessed with the log rank test. Cox proportional hazards regression model was applied for multivariate analysis. For analysis of LVD, the mean LVD for the 5 hot spots per section was subjected to statistical analyses. Wilcoxon matched-pairs signed-rank test was used to compare LVD between the peritoneal and hepatic sides of the gallbladder wall. All reported p values were two-sided, and $p < 0.05$ was considered statistically significant.

Results

During the study period, 92 consecutive patients with pT2 gallbladder carcinoma underwent radical resection. Of these, 11 with invasive primary malignancy in other organs were excluded. The remaining 81 patients, comprising 29 men and 52 women with a median age of 68 (IQR, 63–73) years, were enrolled in this study.

Of the 81 patients, 71 (88%) underwent extended cholecystectomy and 10 (12%) underwent more extensive resection (Supplemental Table). Regarding the timing of surgery, 25 (31%) underwent radical resection as a second procedure after a prior simple cholecystectomy for presumed benign disease. In this study, 74 (91%) underwent dissection of regional lymph nodes and 7 (9%) underwent less extensive lymphadenectomy; paraaortic lymph nodes were dissected or sampled in 45 (56%) patients.

Resection margin status was judged as no residual tumor in 80 (99%) patients and microscopic residual tumor in 1 (1%). In total, 1,403 lymph nodes were retrieved from the 81 patients. Lymph node metastases was found in 33 (41%) patients. Of these 33 patients, 29 had regional lymph node metastasis alone, 2 had distant lymph node metastasis alone, and 2 had both regional and distant lymph node metastasis. The median number of lymph nodes retrieved was 15 (IQR, 9–23). After surgery, adjuvant chemotherapy was administered in 38 (47%) patients. No patients received adjuvant radiotherapy.

Clinicopathological characteristics according to tumor location

Of the 81 patients with pT2 gallbladder carcinoma, 25 (31%) had pT2a (peritoneal-sided) tumors and 56 (69%) had pT2b (hepatic-sided) tumors. The incidence of undergoing liver resection (93% vs. 72%; $p = 0.030$), bile duct resection (77% vs. 44%; $p = 0.005$), and adjuvant chemotherapy (59% vs. 20%; $p = 0.002$) was significantly higher in patients with pT2b tumors than in those with pT2a tumors (Table 1). The extent of lymphadenectomy was comparable between the two groups ($p = 0.194$). pT2b tumors had more unfavorable pathological characteristics than pT2a tumors (Table 1). The following incidences were significantly higher in patients with pT2b tumors than in those with pT2a tumors: regional lymph node metastasis (46% vs. 20%; $p = 0.028$), lymphatic invasion (63% vs. 20%; $p = 0.001$), and venous invasion (46% vs. 20%; $p = 0.028$).

Number and distribution of positive lymph nodes according to tumor location

Between node-positive patients with pT2a ($n = 6$) and pT2b ($n = 27$) tumors, no significant differences were observed in the number of positive nodes (median, 2 [IQR, 1.25–2] vs. 2 [IQR, 1–3]; $p = 0.910$) or the incidence of 4 or more lymph nodes involved (17% vs. 22%; $p > 0.999$). The anatomical distribution of positive lymph nodes appeared to be comparable between the two groups (Table 2). For node-positive patients with pT2b tumors, metastasis was observed in every regional node group (12%–63%), whereas for node-positive patients with pT2a tumors, metastasis was detected in not only the cystic duct node group (67%) but also in the node groups outside the hepatoduodenal ligament (the common hepatic artery, posterior superior pancreaticoduodenal, and paraaortic node groups, 33%–40%).

Surgical outcomes according to tumor location and prognostic factors

After a median follow-up period of 120 (IQR, 56–189) months, for all 81 patients with pT2 gallbladder carcinoma, the 5-year and 10-year DSS rates after resection were 79% and 77%, respectively. DSS after resection was significantly worse in patients with pT2b tumors than in those with pT2a tumors, with 5-year DSS rates of 72% and 96%, respectively ($p = 0.027$) (Fig. 2). To identify prognostic factors for all 81 patients with pT2 gallbladder carcinoma, univariate and multivariate analyses for DSS were then performed, revealing that pM classification (hazard ratio, 34.646; 95% confidence interval, 7.763–154.610; $p < 0.001$) and tumor location (hazard ratio, 14.162; 95% confidence interval, 1.588–126.291; $p = 0.018$) were significant independent prognostic factors (Table 3). The extent of lymphadenectomy did not affect DSS after resection.

LVD according to location in normal gallbladders

In all 10 gallbladder specimens, D2-40 antibody-positive lymphatic vessels were observed mostly in the perimuscular connective tissue (Fig. 3A and B). Computer-assisted morphometric

Table 1
Clinicopathological characteristics of 81 patients with pT2 gallbladder carcinoma according to tumor location.

Variable	No. of patients		p value
	pT2a (peritoneal-sided) tumor (n = 25)	pT2b (hepatic-sided) tumor (n = 56)	
Age (years)			0.464
≤ 70	13	35	
> 70	12	21	
Sex			0.803
Male	8	21	
Female	17	35	
Gallstone			0.630
Absent	10	27	
Present	15	29	
Timing of radical resection			0.299
Initial radical resection	15	41	
Radical second resection	10	15	
Liver resection			0.030
Absent	7	4	
Present	18	52	
Extent of lymphadenectomy			0.194
Regional lymphadenectomy	21	53	
Less extensive lymphadenectomy	4	3	
Bile duct resection			0.005
Absent	14	13	
Present	11	43	
Adjuvant chemotherapy			0.002
Absent	20	23	
Present	5	33	
pN classification[a]			0.028
pN0	20	30	
pN1 plus pN2[b]	5	26	
pM classification[a]			0.427
pM0	24	50	
pM1	1	6	
Size of the primary tumor (mm)			0.091
≤ 60	16	23	
> 60	9	33	
Histological grade[a]			0.332
G1	12	20	
G2 plus G3 plus G4	13	36	
Histological type[a]			0.366
Adenocarcinoma	22	53	
Other[c]	3	3	
Lymphatic invasion			0.001
Absent	20	21	
Present	5	35	
Venous invasion			0.028
Absent	20	30	
Present	5	26	
Perineural invasion			0.098
Absent	22	39	
Present	3	17	

pT classification, pathological primary tumor classification; pN classification, pathological regional lymph node classification; pM classification, pathological distant metastasis classification; G1, well differentiated; G2, moderately differentiated; G3, poorly differentiated; G4, undifferentiated.

[a] According to the AJCC TNM staging system, eighth edition.[3].

[b] Regional lymph nodes included the cystic duct, pericholedochal, posterior superior pancreaticoduodenal, retroportal, and common and proper hepatic artery lymph node groups.

[c] Other histological types included adenosquamous carcinoma (n = 2), small cell neuroendocrine carcinoma (n = 1), mixed adenoneuroendocrine carcinoma (n = 1), carcinosarcoma (n = 1), and undifferentiated carcinoma (n = 1).

analysis of the lymphatic vessels revealed that LVD was significantly higher on the hepatic side than on the peritoneal side of the wall of the gallbladder (mean ± standard deviation: 22.4 ± 2.4/mm^2 vs. 6.9 ± 1.4/mm^2; p = 0.002), with the highest LVD value found on the hepatic side.

Discussion

Tumor location has become an increasingly recognized prognostic factor in patients with pT2 gallbladder carcinoma [13–15,20]. The new AJCC staging system adopted tumor location for the subclassification of pT2 gallbladder carcinomas, because tumors on the hepatic side (pT2b tumors) carry a worse prognosis than those on the peritoneal side (pT2a tumors) [12]. Two recent studies characterized pT2b tumors as having a higher propensity than pT2a tumors for lymph node metastasis [13,14]. Although lymphatic spread is the main mode of spread for pT2 gallbladder carcinoma [5,9,11], a detailed analysis of the lymphatic spread according to tumor location is lacking. Our findings indicate that tumor location can predict the presence or absence of regional lymph node metastasis but not the number and anatomical distribution of positive regional lymph nodes in patients with pT2 gallbladder carcinoma. Namely, once metastasis develops in regional lymph nodes, the extent of lymphatic spread is similar irrespective of

Table 2
Distribution of positive lymph nodes according to tumor location in 33 patients with pT2 gallbladder carcinoma with lymph node metastasis.

Node group	pT2a patients with nodal disease (n = 6)		pT2b patients with nodal disease (n = 27)	
	No. of patients with node group evaluated	No. of patients with positive nodes (%)[a]	No. of patients with node group evaluated	No. of patients with positive nodes (%)[a]
Cystic duct	6	4 (67)	27	17 (63)
Pericholedochal	6	0 (0)	27	13 (48)
Retroportal	6	0 (0)	25	4 (16)
Proper hepatic artery	6	0 (0)	25	3 (12)
Common hepatic artery	5	2 (40)	25	5 (20)
Posterior superior pancreaticoduodenal	6	2 (33)	26	4 (15)
Paraaortic	3	1 (33)	17	3 (18)

pT classification, pathological primary tumor classification.
[a] Values in parentheses are the incidence of lymph node metastasis for each group calculated by dividing the number of patients with positive lymph nodes from the given group by the number of patients with lymph nodes evaluated in that group.

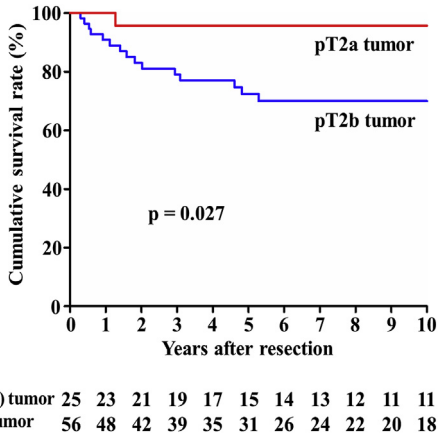

Fig. 2. Disease-specific survival (DSS) of 81 patients who underwent radical resection for pT2 gallbladder carcinoma stratified by tumor location [12]. For patients with pT2a and pT2b tumors, the 5-year DSS rates were 72% and 96%, respectively (p = 0.027).

tumor location. The findings of this study also demonstrated that LVD values were significantly higher in the wall of the normal gallbladder on the hepatic side compared with the peritoneal side, with the highest LVD value on the hepatic side. The presence of more lymphatic vessels on the hepatic side can increase the likelihood that cancer cells will invade the lymphatic vessels. This may partly explain why patients with pT2b tumors are more likely to have regional lymph node metastases than those with pT2a tumors.

The deeper the tumor penetrates the wall of the gallbladder, the greater the likelihood of lymph node metastasis in patients with gallbladder carcinoma [10,21,22]. The incidence of lymph node metastasis is very low for tumors limited to the mucosa or muscle layer (pT1 tumors), but ranges from 28% to 62% for pT2 tumors [6–10,13–17]. Recent studies reported differences in the incidences of metastasis to the regional lymph nodes according to the location of pT2 tumors [13,14]. The reported incidences of regional lymph node metastasis in patients with pT2a and pT2b tumors ranged from 15% to 26%, and from 30% to 39.7%, respectively [13–15]. The present study also confirmed this finding: the incidences of regional lymph node metastasis in patients with pT2a and pT2b tumors were 20% and 46%, respectively. Nevertheless, the number and anatomical distribution of positive lymph nodes was similar between the two groups. For node-positive patients with pT2b tumors, metastasis was found in every regional node group (12%–63%). In comparison, in patients with node-positive pT2a tumors, metastasis was observed even in regional node groups outside the hepatoduodenal ligament (Table 2). These findings indicate that regional lymphadenectomy should be recommended for patients who undergo radical resection for pT2b tumors. Given that approximately one-fifth of patients with pT2a tumors had lymph node metastasis that was observed in node groups not only inside but also outside the hepatoduodenal ligament, regional lymphadenectomy should not be ruled out and the extent of this procedure should not be changed for these patients, provided that they are fit enough for surgery.

Previously, it was reported that perimuscular connective tissue contains more and larger lymphatic vessels than shallower layers in the normal gallbladder based on results of immunohistochemistry using the monoclonal antibody D2-40 [19]. This finding may partially explain the reported high incidence of lymph node metastasis in pT2 or higher advanced gallbladder carcinoma [19]. Using the same method, this study clearly demonstrated that more lymphatic vessels were present on the hepatic side than on the peritoneal side of the wall of the normal gallbladder, partly explaining the higher incidence of lymph node metastasis in patients with pT2b tumors than in those with pT2a tumors. To our knowledge, this is the first study to clarify the mechanism by which tumor location of pT2 gallbladder carcinoma is associated with the incidence of lymph node metastasis.

In this study, tumor location was an independent prognostic factor in patients with pT2 gallbladder carcinoma. The 5-year DSS rates of patients with pT2a and pT2b tumors were 96% and 72%, respectively (p = 0.018). The more aggressive biological behavior (higher incidences of lymph node metastasis and lymphatic and venous invasion) of pT2b tumors in this series may have contributed to this finding. Earlier studies have revealed similar results [13,14]. Together, these results indicate that, clinically, patients with pT2a gallbladder carcinoma would benefit the most from radical resection alone, whereas patients with pT2b gallbladder carcinoma may be good candidates for multimodal treatment including both surgery and adjuvant therapy, because surgical outcomes in these patients appear not to be satisfactory.

Limitations

This study had several limitations. First, this was a retrospective study that included a small number of patients at a single institution over a long period of time. Second, surgical procedures and adjuvant therapy were not performed equally among patients with pT2a and pT2b tumors. A variety of adjuvant chemotherapy regimens were used in this study. Third, normal gallbladder specimens were used in the analysis of LVD according to tumor location, although it has been reported that malignant tumors are often associated with increased LVD in the peritumoral region [23,24]. Therefore, the results of this study should be interpreted with caution. On the other hand, this study is one of the largest single-

Table 3
Prognostic factors influencing disease-specific survival after resection in 81 patients with pT2 gallbladder carcinoma.

Variable	No. of patients	Survival rate (%)		Univariate analysis	Multivariate analysis	
		5-year	10-year	p value	Hazard ratio (95% CI)	p value
Age (years)				0.212		
≤ 70	48	84	82			
> 70	33	72	72			
Sex				0.773		
Male	29	78	78			
Female	52	80	77			
Gallstone				0.058		
Absent	37	67	67			
Present	44	90	87			
Timing of radical resection				0.322		
Initial radical resection	56	76	74			
Radical second resection	25	86	86			
Liver resection				0.553		
Absent	11	73	73			
Present	70	81	79			
Extent of lymphadenectomy				0.855		
Regional lymphadenectomy	74	79	77			
Less extensive lymphadenectomy	7	83	83			
Bile duct resection				0.737		
Absent	27	83	83			
Present	54	79	76			
Adjuvant chemotherapy				0.295		
Absent	43	76	72			
Present	38	83	83			
pN classification[a]				<0.001		
pN0	50	87	87			
pN1[b]	25	80	75			
pN2[b]	6	17	17			
pM classification[a]				<0.001		
pM0	74	84	82		1.000	
pM1	7	29	29		34.646 (7.763–154.610)	<0.001
Size of the primary tumor (mm)				0.677		
≤ 60	39	80	80			
> 60	42	79	75			
Histological grade[a]				0.144		
G1	32	90	86			
G2 plus G3 plus G4	49	72	72			
Histological type[a]				0.261		
Adenocarcinoma	75	80	79			
Other[c]	6	63	63			
Lymphatic invasion				0.769		
Absent	41	82	79			
Present	40	77	77			
Venous invasion				0.257		
Absent	50	82	82			
Present	31	75	70			
Perineural invasion				0.059		
Absent	61	85	83			
Present	20	62	62			
Tumor location[a]				0.027		
T2a (peritoneal side)	25	96	96		1.000	
T2b (hepatic side)	56	72	70		14.162 (1.588–126.291)	0.018

pT classification, pathological primary tumor classification; CI, confidence interval; pN classification, pathological regional lymph node classification; pM classification, pathological distant metastasis classification; G1, well differentiated; G2, moderately differentiated; G3, poorly differentiated; G4, undifferentiated.

[a] According to the AJCC TNM staging system, eighth edition.[3].

[b] Regional lymph nodes included the cystic duct, pericholedochal, posterior superior pancreaticoduodenal, retroportal, and common and proper hepatic artery lymph node groups.

[c] Other histological types included adenosquamous carcinoma (n = 2), small cell neuroendocrine carcinoma (n = 1), mixed adenoneuroendocrine carcinoma (n = 1), carcinosarcoma (n = 1), and undifferentiated carcinoma (n = 1).

institution studies involving patients with pT2 gallbladder carcinoma. In addition, compared with previous studies, this study more clearly evaluated the incidence and anatomical distribution of lymph node metastasis according to tumor location in patients with pT2 gallbladder carcinoma. Furthermore, assuming that the ability of tumor to induce lymphatic vessels in the wall of the gallbladder is comparable irrespective of tumor location, a higher number of pre-existing lymphatic vessels might lead to a higher likelihood of lymph node metastasis in patients with pT2b tumor than in those with pT2a tumor.

Conclusions

The data of this study indicate that tumor location can predict the presence or absence of regional lymph node metastasis but not the number and anatomical distribution of positive regional lymph nodes in patients with pT2 gallbladder carcinoma. Regional lymphadenectomy should be recommended for patients who undergo radical resection for pT2b tumors. Because approximately one-fifth of patients with pT2a tumors will have lymph node metastasis, which were observed in node groups not only inside but also

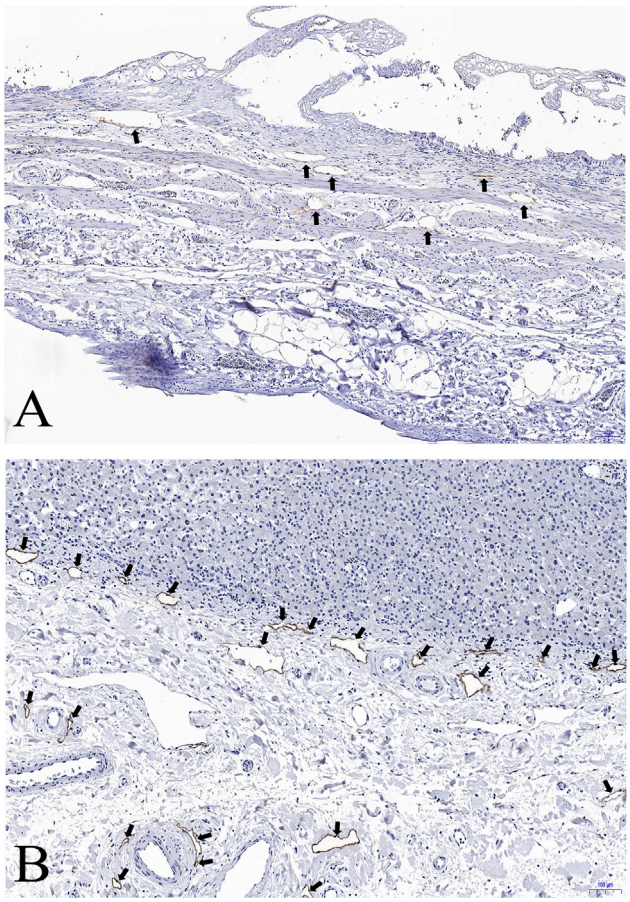

Fig. 3. D2-40 antibody-positive lymphatic vessels in the wall of the normal gallbladder according to location [12]. The gallbladder wall on the hepatic aspect of the normal gallbladder contains more and larger lymphatic vessels than the wall on the peritoneal side. A. The wall of the peritoneal side of the gallbladder. B. The wall of the hepatic side of the gallbladder. Arrows indicate D2-40 antibody-positive lymphatic vessels.

outside the hepatoduodenal ligament, regional lymphadenectomy should not be ruled out and the extent of this procedure should not be changed for these patients, provided that they are fit enough for surgery. This study also demonstrated that the wall on the hepatic side of the normal gallbladder contains more lymphatic vessels than the wall on the peritoneal side, partly explaining why the incidence of lymph node metastasis is higher in patients with pT2b tumors than in those with pT2a tumors.

Conflict of interest statement

No specific funding was received for this study and the authors have no conflicts of interest to disclose.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ejso.2019.03.038.

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