Original Article

Clinical meaning of the World Health Organization morphologic classification (flat *vs*. tumoral) of gallbladder intraepithelial neoplasm as a prognostic factor in gallbladder cancer

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Background: In the World Health Organization (WHO) classification, gallbladder (GB) intraepithelial lesions are grouped as flat or tumoral, according to their morphological features. The purpose of this study was to investigate the relationship between the morphologies and clinical features of GB cancer (GBC) and to examine the feasibility of using morphologic classification as a prognostic factor.

Methods: From January 2000 to December 2012, the available pathologic slide reviews of 381 patients were analyzed at the Seoul National University Hospital. All pathologic slides were evaluated by two pancreato-biliary tract pathology experts. GBCs were categorized into eight groups (Flat: F1-2, Borderline, Tumoral: Tu1-5), according to the thickness of the mucosal lesion, histologic patterns of the mucosa under microscopy, invasion extent, and patient history of premalignant lesions. According to the morphologic classification, clinical features were compared and survival analysis was performed.

Results: In three groups, flat lesions comprised 179 (46.9%) cases and borderline and tumoral comprised 97 (25.4%) and 105 (27.5%) cases, respectively. More favorable pathologic and clinical results were found within the tumoral group. The borderline group had an intermediate tendency between flat and intraluminal in clinicopathologic parameters. In the curative resected T2 stage group, the borderline group demonstrated an intermediate trend compared to that of the flat and tumoral groups, but this was statistically insignificant (P=0.08).

Conclusions: Flat type GBCs show worse prognosis than tumoral GBCs. The morphological classifications between flat and tumoral on the basis of 1 cm and by papillary feature is feasible. Tumor morphology can be used as a reference while deciding the treatment plan, especially in T2 GBC.

Keywords: Gallbladder neoplasm; morphology; histology; classification; prognosis

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Introduction

To understand the characteristics of malignancy, it is important to understand the carcinogenic signaling pathway. There are two major carcinogenic pathways in gallbladder cancer (GBC) (1,2). First is the dysplasia-carcinoma pathway, which resembles uterine cervix cancer. The other is adenoma-carcinoma sequence, which is similar to that of colorectal cancer. The majority of GBCs manifest via the dysplasia pathway, and the adenoma pathway accounts for approximately 5–10% in GBC (1,3). In these pathways, cancer arises from a premalignant intraepithelial lesion. However, the definition and classification of an intraepithelial lesion in the gallbladder (GB) is ambiguous, and studies on its clinical features not been actively performed.

The World Health Organization (WHO) definition and classification of premalignancy is widely referenced. The WHO classifies the intraepithelial neoplasm of GB into adenoma (tubular, papillary, and tubulopapillay), biliary intraepithelial neoplasia, grade 3 (BilIN-3), intracystic papillary neoplasm (ICPN), and mucinous cystic neoplasm (4). Each subtype is categorized according to morphology and has its own clinical and pathologic features. The morphology of adenoma is typically polypoid, single, and well demarcated. A small portion of adenomas progress to invasive carcinomas (5,6). ICPN is defined as an intracystic papillary neoplasm of the GB and is associated with invasive papillary carcinoma, which has different clinical characteristics than typical GBC (7). Each subtype is distinct, manifesting disease differently; however, they could be grouped instead into flat type or tumoral types, according to their gross and microscopic morphologies, similar to other biliary-pancreas tract neoplasms (*Table 1*) (8).

Adenoma and ICPN are included in the tumoral type. Even though the morphologies of the two subtypes are similar, there are no definitely reliable criteria to differentiate between adenoma and ICPN. It is especially difficult to distinguish between the papillary features of adenoma and ICPN. Therefore, a new definition of intracholecystic papillary tubular neoplasm (ICPTN) has been suggested. ICPTN is exophytic (papillary or polypoid) well-demarcated, with intramucosal GB masses measuring ≥1.0 cm (9). ICPTN could include adenoma and ICPN and exclude exuberant papillary hyperplasia with dysplasia according to the thickness of the mucosa (<1 cm). A determination of ICPTN offers a better prognosis than conventional GBC originating from BilIN-3. A new morphological definition attributed to the thickness of mucosa ≥1.0 cm shows that morphological classification has different clinical features (9).

Another morphological feature that provides a different prognosis is papillary appearance in the lesion. Papillary GB neoplasms show a favorable prognosis. However, the definition of papillary appearance is ambiguous. There is a discrepancy in the categorization of gross and microscopic papillary features. This confusion is more common in GB than other organs due to the histologic features of the normal GB mucosa, where papillary appearance is observed microscopically. Metaplasia and hyperplasia by recurrent inflammation are also observed in the normal GB mucosa (10). Thus, it is difficult to distinguish between a papillary neoplasm or a papillary change from the normal mucosa; and accordingly, papillary appearance needs to be clarified. Therefore, we divided papillary neoplasms according to the thickness of the mucosa into greater than 1 cm or less than 1 cm.

As mentioned above, morphologic features (tumoral or flat, papillary) of the GB lesion are related to their clinical feature. To study the relationship between morphology and the corresponding clinical feature of GB cancer (GBC), the GBC lesions were grouped into flat or tumoral types, according to the thickness of mucosal portion of tumor (1 cm), macroscopic and microscopic morphologies (papillary or not), background premalignant lesions, and histology type of the associated invasive tumor. In the flat type, the borderline group was defined separately. In GBC, the borderline group has papillary features, but its mucosal height is not over 1 cm. Therefore, the purpose of this study was to investigate the relationship between different morphologies (flat *vs.* tumoral) and clinical features of GBC and to examine the feasibility of using morphologic classification as a prognostic factor in GBC. We present the following study in accordance with the STROBE reporting checklist (available at http://dx.doi.org/10.21037/atm-20-432).

Methods

Patients and data collection

From January 2000 to December 2012, 416 consecutive patients underwent surgery for GBC at the Seoul National University Hospital. Among them, 381 patients with pathologic slide reviews available were analyzed. Patient

Table 1 Classification of intraepithelial neoplasms in the biliary-pancreatic tract

Organ	Flat	Tumoral
Bile duct	BilIN	Intraductal papillary neoplasm of bile duct (IPNB)
Gallbladder	BilIN	Adenoma; intracystic papillary neoplasm (ICPN)
Ampulla of Vater	Flat intraepithelial neoplasia high grade	Intestinal type adenoma; non-invasive pancreatobiliary neoplasm
Pancreas	Pancreatic intraepithelial neoplasia (PanIN)	Intraductal papillary mucinous neoplasm (IPMN)

characteristics were reviewed for age, sex, pathologic diagnosis, operation method, adjuvant treatment, and recurrence. The clinical data for these patients were prospectively collected via electronic medical record data. Pathologic data were also reviewed for differentiation, presence of tubular type, invasive component, cell type, lymphatic invasion, vascular invasion, and perineural invasion.

Pathologic classification

All pathologic data, including new classifications, were evaluated by two pancreato-biliary tract specialized pathologists (Kyoung-Bun Lee and Haeryoung Kim). Two pathologists reviewed each of the same slides. When comparing each result with another result, the final classification was decided by discussing the slide again. GBCs were classified according to the morphology of the background intraepithelial lesion and were categorized into eight groups (Flat: F1-3, Tumoral: Tu1-5), and according to the extent of invasion, gross type, and mucosal changes. F1 was defined as minimally invasive carcinoma with flat intraepithelial lesions, which was invisible under gross inspection; had no mucosal hyperplasia and thickening; where the invasive tumor was observed only under microscopic examination (>40× power); or only intraepithelial carcinoma (carcinoma *in situ*) was observed. F2 was defined as a widely invasive carcinoma with and without flat intraepithelial lesions, the presence of nodular sclerotic gross lesions, without mucosal hyperplasia or papillary type dysplasia in the adjacent mucosa both upon macroscopic and microscopic inspection. F3 was defined as possessing papillary features with no dominating presence of polyps; and the mucosa also bore papillary features around the invasive tumor under microscopy, but the mucosal lesion did not have any polypoid lesions greater than 1 cm upon gross inspection. Microscopic features of the mucosa of F3 was micropapillary or short papillary epithelial dysplasia directly on the stroma, just above proper muscle or lamina propria, lacking a thick fibrovascular stalk. The F3 category is comprised of minimally invasive and widely invasive tumors. The borderline group was classified under F3 as it shows papillary features under the microscope, but it is ambiguous macroscopically. Tu1 was defined as demonstrating papillary features with polyps, which results in a polypoid mucosal lesion greater in 1 cm in height. F3 and Tu1 can be distinguished by height: whether the height is below 1 cm or above 1 cm. Tu2 represents noninvasive papillary carcinoma, which has been described as noninvasive papillary carcinoma or biliary papillomatosis. Gross mucosa is replaced entirely by sessile papillary mucosa and is histologically composed of long slender papillary neoplasms with a fibro-vascular core. The major distinguishing factors between Tu1 and Tu2 was merely the observed spreading feature versus a localized polypoid feature. Invasive papillary carcinoma was classified as Tu3, which presents the features of Tu2 but expresses an invasive tumor. Tu4 and Tu5 were defined as minimally invasive adenocarcinomas arising in the adenoma as well as widely invasive carcinomas arising in the adenoma, respectively. The criteria differentiating between minimal and wide invasiveness was the same as for F1 and F2. The morphologic definition of adenoma adheres to the rules of the 2010 WHO classification, in which adenoma is defined as the proliferative epithelial lesion with tubular, tubulopapillary or papillary pattern and grossly identified as an isolated polypoid lesion. Among these patterns, the predominant papillary pattern was classified as Tu1, as mentioned before. These eight subgroups were classified from three groups: flat (F1, 2), borderline (F3) and tumoral (Tu1-5); the key classifying criteria and representative pictures of gross and microscopic features under low and high magnifications are summarized in *Table 2* and *Figure 1*.

Statistical analysis

All statistical analysis was performed using SPSS version 22.0 (IBM, Almank, NY, USA). Nominal variables were

Table 2 Classification according to gross morphologic and histologic patterns

Classification	Height	Papillary	Invasiveness	Spread direction	Premalignant lesion	Carcinogenic pathway	n (%)
F1	<1 cm	–	M	–	Bil IN	Metaplasia/dysplasia	20 (5.2)
F2	<1 cm	–	W	–	Bil IN	Metaplasia/dysplasia	159 (41.7)
Borderline (F3)	<1 cm	Papillary	M or W	–	ICPN	Unknown	97 (25.5)
Tu1	≥1 cm	Papillary	M	Upside	ICPN	Hyperplasia/dysplasia	27 (7.1)
Tu2	≥1 cm	Papillary	M	Laterally	ICPN	Hyperplasia/dysplasia	18 (4.7)
Tu3	≥1 cm	Papillary	W	–	ICPN	Hyperplasia/dysplasia	18 (4.7)
Tu4	≥1 cm	–	M	–	Adenoma	Adenoma-carcinoma	28 (7.3)
Tu5	≥1 cm	–	W	–	Adenoma	Adenoma-carcinoma	14 (3.7)

M, minimally; W, widely; Bil IN, biliary intraepithelial neoplasm; ICPN, intracholecystic papillary neoplasm.

compared using the Chi-squared test or Fisher's exact test, and continuous variables were compared using the Student's *t*-test or analysis of variance (ANOVA). Survival was determined using the Kaplan-Meier method with survival curves compared using the log-rank test. P values of less than 0.05 were considered statistically significant. As a prognostic factor, recurrence was defined through follow up image data such as computerized tomography, magnetic resonance imaging or elevated tumor markers, resulting in the patient undergoing additional chemotherapy. To determine recurrence risk factors the Cox regression test was used.

The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The present study protocol was reviewed and approved by the Institutional Review Board of the Seoul National University Hospital (approval No. 1508-081-695). The process of obtaining informed consent was waived according to the decision of the IRB.

Results

Demographics

The mean age of the 381 patients was 67.4±10.8 years. Two hundred and nine patients were female (54.8%). Classifications of T1a, T1b, T2, T3, and T4 carcinoma were 44 (11.5%), 52 (13.6%), 23 (6.0%), 161 (42.2%), 86 (22.5%), and 15 (3.9%), respectively, as identified via *in situ*. Curative resections were performed on 298 patient samples (78.2%). Cholecystectomies were performed in 191 patients (50.1%) and extended cholecystectomies were performed in 165 patients (43.3%). Hepatectomies; right hemihepatectomies or extended right hemihepatectomies were performed in 12 patients (3.1%). There were eight pancreaticoduodenectomy cases (2.1%) and five case of hepatopancreatic duodenectomies. Gallstones were present in 85 cases (24.5%). The clinicopathological features of 381 patients were shown in *Table 3*.

The most common type of GBCs observed was F2, a widely invasive carcinoma with flat atypia (n=159, 41.7%). The flat tumor types classified under F1 and F2 were combined as one (n=169, 49.9%); while the tumor types classified into Tu1, Tu2, Tu3, Tu4 and Tu5 (n=105, 27.6%) were classified as another. Finally, the F3 flat tumor type (n=97, 25.5%) was separately considered; these were carcinomas with papillary dysplasia, lacking any visible dominant polyps, with either tumors that were either minimally invasive or invasive (*Table 2*).

Clinicopathological features according to morphologic classification

Table 3 shows clinicopathological features according to morphologic classification. There were no observable differences seen with age or sex between the classifications. The maximal diameter of a lesion was seen in the flat group compared to either the borderline or tumoral groups (P=0.06). In the flat group, more patients had advanced T stage disease. In addition, lymph node metastasis and distant metastasis were more common in the flat group than in either the borderline or tumoral groups. Curative resections were more frequently performed in the tumoral group than in the borderline or flat groups. Lastly, most of the patients in the borderline and tumoral groups underwent simple cholecystectomies or extended cholecystectomies.

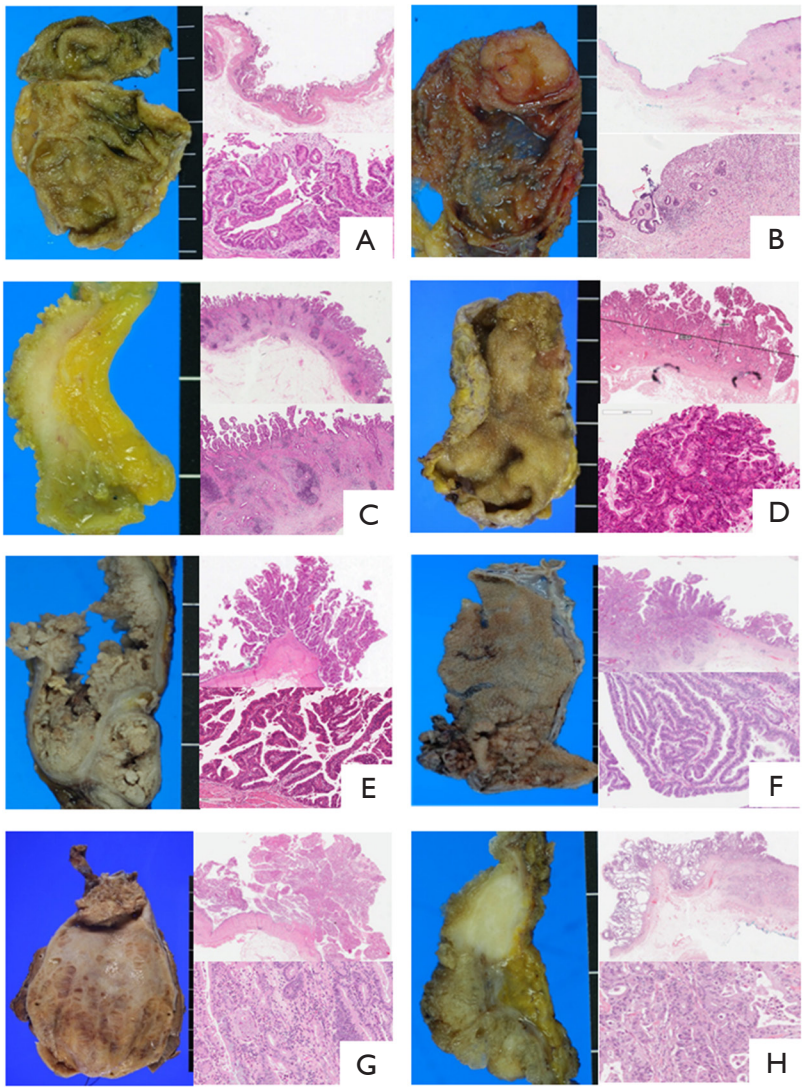

Figure 1 Classification according to gross morphologic and histologic. (A) F1: minimally invasive carcinoma with flat atypia; (B) F2: widely invasive carcinoma with flat atypia; (C) F3: carcinoma with papillary dysplasia but without any dominant polyps; (D) Tu1: carcinoma with papillary dysplasia and dominant polyp; (E) Tu2: noninvasive papillary carcinoma; (F) Tu3: invasive papillary carcinoma; (G) Tu4: minimally invasive adenocarcinoma arising in adenoma; (H) Tu5: widely invasive carcinoma arising in adenoma (left, gross; right upper, hematoxylin & eosin (HE) ×12.5; right lower, HE ×200).

The proportion of well differentiated cancer observed was 27.9%, 46.4% and 66.7% in the flat, borderline and tumoral groups, respectively (P<0.01). More invasive features such as lymphatic invasion, vascular invasion, perineural invasion were found in the flat group. Intestinal cell type of intraepithelial portion was higher in the borderline or tumoral lesions than in the flat lesion (29.9%, 25.75, and 16.8%, P=0.03). Histologic characteristics of dedifferentiation such as signet ring cell features, spindle cells, or solid pattern, were more frequently found in flat lesions than borderline or tumoral lesions (46.4%, 17.5% and 11.4%, P<0.01). The borderline group manifested intermediate histopathologic parameters of the flat and tumoral groups (*Table 4*).

Survival and recurrence analysis

The 5-year survival rates were 58.5% in a total of

Table 3 Demographic and clinicopathological characteristics of 381 patients

Parameters	N=381	Flat (n=179, %)	Borderline (n=97, %)	Tumoral (n=105, %)	P value
Age, years	67.4±10.8	67.9±10.8	67.9±11.9	66.5±9.8	0.921
Sex					0.149
Male	172	90 (50.3)	41 (42.3)	41 (39.0)	
Female	209	89 (49.7)	56 (57.7)	64 (61.0)	
Maximum diameter (cm)	4.38±2.58	4.79±2.88	4.08±2.22	3.96±2.27	0.057
pT					<0.001
Tis	44	12 (6.7)	11 (11.3)	21 (20.0)	
T1a	52	5 (2.8)	17 (17.5)	30 (28.6)	
T1b	23	6 (3.4)	5 (5.1)	12 (11.4)	
T2	161	74 (41.3)	52 (53.6)	35 (33.3)	
T3	86	67 (37.4)	12 (12.4)	7 (6.7)	
T4	15	15 (8.4)	0 (0.0)	0 (0.0)	
pM					0.052
M0	347	157 (87.7)	89 (90.9)	101 (96.2)	
M1	34	22 (12.2)	8 (9.0)	4 (3.8)	
pN					0.002
N0/x	276	119 (60.5)	67 (69.1)	90 (85.7)	
N1	105	60 (33.5)	30 (30.9)	15 (14.3)	
Curative resection					<0.001
Curative	298	120 (67.0)	78 (80.4)	100 (95.2)	
Palliative	83	59 (33.0)	19 (19.6)	5 (4.8)	
Operation					0.002
Simple cholecystectomy	191	75 (41.9)	60 (61.9)	56 (53.3)	
Extended cholecystectomy	165	82 (45.8)	35 (36.1)	48 (45.7)	
Hepatectomy	12	11 (6.1)	1 (1.0)	0 (0.0)	
Pancreaticoduodenectomy	8	6 (3.4)	1 (1.0)	1 (1.0)	
HPD	5	5 (2.8)	0 (0.0)	0 (0.0)	
Presence of gallstone	82	39 (22.7)	16 (16.5)	27 (25.7)	0.362

Hepatectomy, right hemihepatectomy or extended right hemihepatectomy; HPD, hepatopancreaticoduodenectomy.

381 patients and 72.0% in the curative resected group. Moreover, the 5-year disease-free survival rate were 66.2%, 74.9% and 16.8% in the entire, curative and palliative groups, respectively. In the curative resected group, the recurrence rate differed according to the T stage and morphologic classification; both were statistically significant (P<0.01, P<0.01).

Table 5 shows the risk factors of the curative resected GBC patients. Univariate analysis indicates that the following are risk factors: T stage, N stage, morphologic classification, differentiation, vascular invasion, lymphatic invasion, perineural invasion and adjuvant chemo therapy. In the multivariate analysis, the T stage was the only significant recurrence risk factor for curative resected GBC

Table 4 Histopathologic features of GB cancers according to classification

Parameters	N=381	Flat (n=179, %)	Borderline (n=97, %)	Tumoral (n=105, %)	P value
Differentiation of invasive tumor					<0.001
Well	165	50 (27.9)	45 (46.4)	70 (66.7)	
Moderate	131	61 (34.1)	41 (42.3)	29 (27.5)	
Poor	85	68 (38.0)	11 (11.3)	6 (1.6)	
Tubular type adenocarcinoma					0.012
Absent	138	63 (35.2)	26 (26.8)	49 (46.7)	
Present	243	116(64.8)	71 (73.2)	56 (53.3)	
Dedifferentiated component[†]					<0.001
Absent	270	96 (53.6)	80 (82.5)	93 (88.6)	
Present	111	83 (46.4)	17 (17.5)	12 (11.4)	
Cell type of intraepithelial lesion					0.030
Intestinal	86	30 (16.8)	29 (29.9)	27 (25.7)	
Nonintentional	295	149(83.2)	68 (70.1)	78 (74.3)	
Lymphatic invasion					<0.001
Absent	253	97 (54.2)	68 (70.1)	88 (83.8)	
Present	128	82 (45.8)	29 (29.9)	17 (16.1)	
Vascular invasion					<0.001
Absent	323	138(77.1)	83 (85.6)	101(97.1)	
Present	58	41 (22.9)	14 (14.4)	3 (2.9)	
Perineural invasion					<0.001
Absent	267	92 (51.4)	74 (76.3)	101(96.2)	
Present	114	87 (48.6)	23 (23.7)	4 (3.8)	
Pseudoinvasion					<0.001
Absent	258	151(84.4)	53 (54.6)	54 (51.4)	
Present	123	28 (15.6)	44 (45.4)	51 (48.6)	

[†], signet ring cell, spindle cells, solid pattern.

patients (P<0.01).

To determine the effects of morphology, further analysis according to the T stage was performed. Within the T1 group, there was no recurrence within 5 years. There was no difference within the distinct morphologic classifications of the T3/4 advanced GBC group. However, in the T2 group the five-year disease-free survival rates were 79.2%, 67.4% and 59.6% in the tumoral, borderline and flat groups, respectively. The DFS rate tended to be lower according to the morphologic type, but there was no statistical significance (P=0.08) (*Figure 2*).

Discussion

There are many prognostic factors in GBC; one of them is macroscopic or microscopic morphology (11-13). This study was designed to deal with two questions. The first question was about papillary feature. The cancer which shows papillary morphologic feature offers excellent prognosis (14). However, the definition of papillary

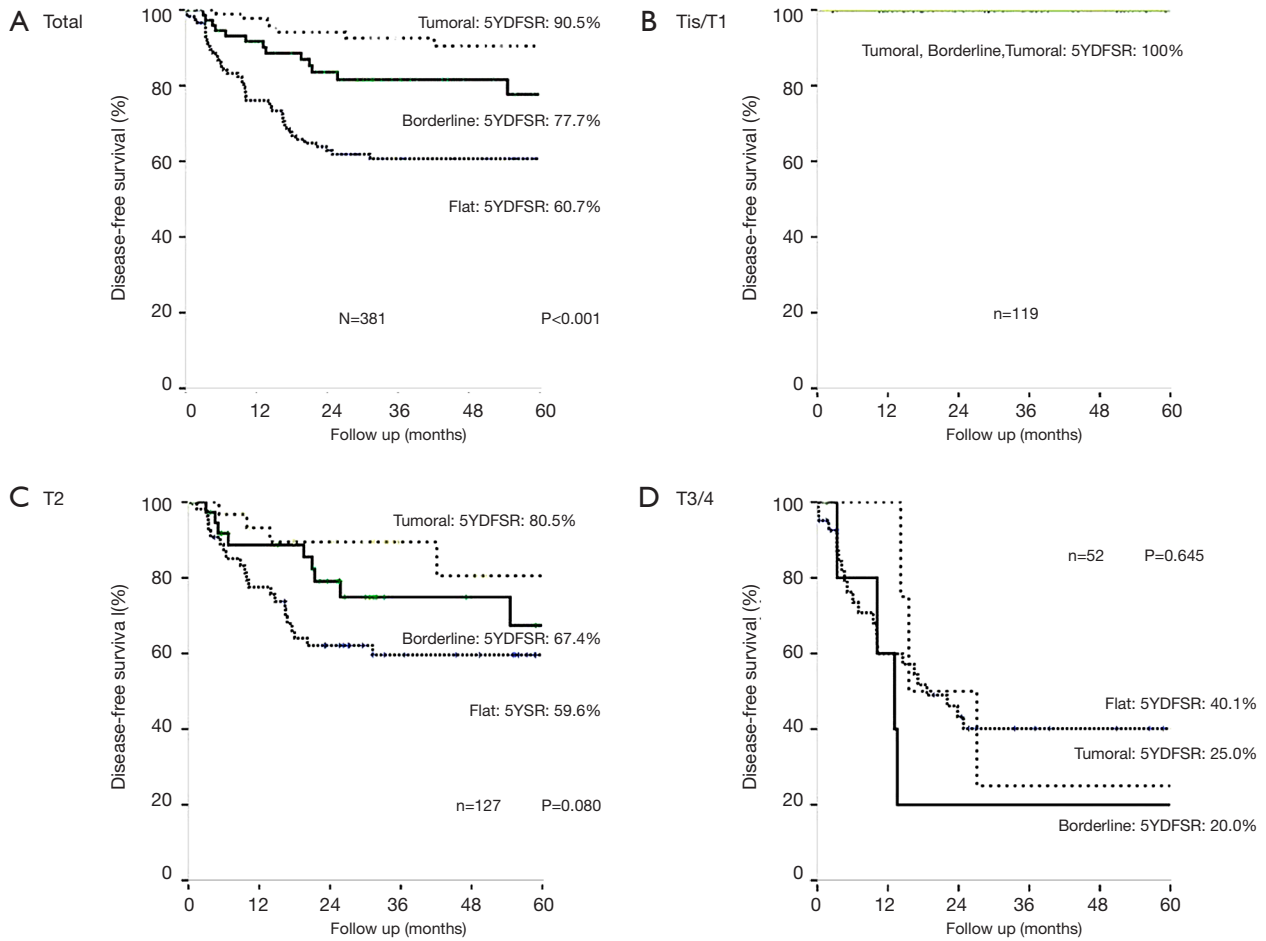

Figure 2 Disease-free survival (DFS) rates according to T classification subgroups. (A) DFS of total patients (n=381); (B) no event in Tis/T1 stage (n=119); (C) DFS in pT2 stage (n=127); (D) DFS in pT3/4 stages (n=52).

is ambiguous, 'What is the papillary? Microscopically or macroscopically?' Normal GB mucosa has papillary folding, and this mucosal folding is accentuated when there is recurrent inflammation (10). This increased papillary folding is referred to as papillary hyperplasia; and when the covered epithelial cells have signs of dysplasia such as stratification, increased nuclear atypia and pleomorphism, increased mitosis, etc., there is no standard for labeling this lesion as dysplasia in papillary hyperplasia of a papillary neoplasm. Accordingly, this makes it difficult to define a papillary lesion. The first aim of this study was to clarify papillary GBC. The second question was to determine whether there is any difference of prognosis between flat and tumoral types in GBC, and the feasibility of height (i.e., 1 cm) as a determinant of polypoid lesion. To reach these objectives, we defined flat and tumoral types according to a height of 1 cm as the cutoff, referring to the WHO classification and Aday's definition of ICPTN (4,9). Next, we looked to identify the disease subgroup (borderline), which manifested characteristics that fell in between tumoral (Tu1) and flat (F2) subgroups.

This study was a large-scale single-institute study. GBC was subdivided into 3 groups. In the flat type GBC, the lesions were commonly found in the advanced stage and curative resection rates were lower than in the tumoral type. Tumor morphology as well as the size of the lesion, when the GB mass is discovered, needs to be consideration factors for subsequent treatment. We define borderline as something too difficult to be included within ICPN, but which demonstrates papillary features with microscopic findings. The prognosis of the borderline zone is intermediate (between that of flat and tumoral). Of note,

Table 5 Recurrence risk factors in GBC patients following curative resection

Variables	Univariate analysis			Multivariate analysis		
	HR	95% CI	P value	HR	95% CI	P value
Sex (male)	0.811	0.401–1.324	0.685			
Age	0.996	0.794–1.017	0.685			
T						
1	–	–	<0.001	–	–	0.002
2	38.754	5.309–282.923	<0.001	20.656	2.706–157.624	0.002
3, 4	113.62	15.462–834.96	<0.001	39.190	4.704–326.435	0.001
N	4.478	2.726–7.356	<0.001	1.570	0.896–2.749	0.115
Classification (tumoral)[†]			<0.001			0.407
Borderline	5.789	2.604–12.873	<0.001	1.758	0.731–4.227	0.208
Flat	2.649	1.068–6.568	0.035	1.363	0.524–3.544	0.525
Differentiation (WD)			<0.001			0.251
MD	5.590	2.842–10.994	<0.001	1.762	0.853–3.683	0.126
PD	8.122	4.007–16.461	<0.001	1.801	0.833–3.895	0.135
Vascular invasion	3.152	1.711–5.807	<0.001	0.994	0.565–1.750	0.984
Lymphatic invasion	3.491	2.218–5.729	<0.001	0.917	0.417–1.786	0.798
Perineural invasion	5.432	3.303–8.934	<0.001	1.417	0.784–2.559	0.248
Adjuvant chemo therapy	2.957	1.807–4.841	<0.001	0.977	0.577–1.712	0.934

[†], dominant polyp: ≥1 cm in height.

the borderline group is more similar to flat than to tumoral lesions. The T2 stage group demonstrated a recurrence-free survival rate similar to that of the borderline group, falling in the middle of the flat and tumoral types. Although the lesion bore papillary-like characteristics, its length fell below 1 cm; and therefore, the group exhibited different characteristics from general papillary lesions.

In 2010, the WHO classified the premalignant lesion of GB into adenoma (tubular, papillary, tubulopapillay), biliary intraepithelial neoplasia, grade 3 (BilIN-3), intracystic papillary neoplasm, and mucinous cystic neoplasm (4). These lesions could also be grouped as flat to reflect the following: BilIN-3 or luminal lesions; adenoma, intracystic papillary neoplasm, and mucinous cystic neoplasm. However, there is no reliable criteria between adenoma and intracystic papillary neoplasm. Therefore, Adsay *et al.* (9) have suggested a new definition (ICPTN), which encompasses adenoma and intracystic papillary neoplasm that is characterized by an exophytic well-demarcated mass-forming tumor greater than 1 cm. The makeup of ICPTN is papillary (43%), tubulopapillary (31%) and tubular (26%). The predominant cell lineage patterns of morphology, supported by specific immunohistochemical markers, were biliary in 50%, gastric foveolar in 16%, gastric pyloric in 20%, intestinal in 8%, and oncocytic in 6%. The factors associated with invasiveness were the extent of high-grade dysplasia, cell type, and papilla formation. Invasive ICPTN has a significantly better overall prognosis than others.

In our study, we tried to incorporate various criteria and terminologies of papillary lesions that have been argued by pathologists over time (Tu1-5). Accordingly, we compared these entities with flat lesions (F1-2) as well as an ambiguous entity that falls in the middle of flat and tumoral lesions (borderline, F3). Interestingly, the F3 group exhibited intermediate clinicopathologic features resembling something in between that of flat and tumoral lesions. In this group, the stage and tumor progression resembled more closely to those of a flat lesion, but histological features were more similar to those of a tumoral lesion, even though the invasive portion was similar to a flat lesion.

A possible mechanism could perhaps be that dysplasia in the flat type lesion occurs via a hyperplastic process, although further studies are needed to confirm this hypothesis.

"Papillary" is a broadly used terminology in anatomic pathology. The general concept of papillary is that of a leaf-like structure composed of epithelial cells with or without the presence of a fibro-vascular core. Depending on the size of papillae, papillary lesions have opposite clinicopathological meanings. In the papillary lesion that is only visible by microscopy at high magnification, fibrovascular cores are scant or absent, and epithelial cells are directly budded on underlying the stromal tissue. This type is usually labeled as "micropapillary," and has been reported as a histologic indication of poor prognosis in several invasive carcinomas (15-18). In contrast, papillary lesions described at the macroscopic level or at a low microscopic magnification have a thick fibro-vascular core or form sessile or stalked polypoid lesions. This papillary lesion is similarly classified as papillary neoplasm in the pancreatobiliary organ and exhibits indolent biologic behavior and has better prognosis than a non-papillary neoplasm. The use of this mixed terminology may be one of the reasons for the few reports on the papillary neoplasm. Therefore, the morphologic criterion defining a papillary neoplasm need to be refined.

A similar spectrum of lesions exists in another biliary-pancreas tract. Pancreatic intraepithelial neoplasia (PanIN) and intraductal papillary mucinous neoplasm (IPMN) are premalignant lesion in the pancreas. Recently, a similar finding was observed in the ampulla of Vater: intraepithelial neoplasms intra-ampullary papillary-tubular neoplasia (IAPNs) (8). These lesions have been grouped into flat and tumoral types (*Table 1*). The characteristics of flat lesions include grossly unidentifiable and non-mucinous tumor types. As the change occurs, shifting to malignancy, the lesion usually shows a tubular structure and leads to a poor prognosis. In contrast, tumoral types are generally identifiable and mucinous and have a better prognosis than do flat lesions.

Cell lineage also differs between the two groups: non-intestinal type (pancreatobiliary, gastric and oncocytic) in the flat, and intestinal type in the tumoral type. Subsequent immunohistochemical analysis yielded distinctive results based on tumor type. MUC1 is usually positive in pancreatobiliary differentiation (pancreatic IPMNs, ampullary IAPNs) (19-21). MUC2 and CDX2 are positive in intestinal differentiation (8,21), MUC6 is pyloric marker and MUC5AC is foveolar mucin marker (22). Flat and tumoral types appear to different in the carcinogenesis pathway. The flat premalignant lesions follow a metaplasia-carcinoma sequence while the tumoral type follows an adenoma-carcinoma sequence (23,24). Thus, the flat and tumoral types are unique and should be considered as separate entities.

The treatment strategy for GBCs, including the feasibility of laparoscopic surgery, extent of liver resection, adjuvant therapy, and regimen of chemo therapy, differs according to the T stage. For surgery, in T1 GBCs, a simple cholecystectomy may be undertaken, or even laparoscopic cholecystectomy is feasible, according to some reports (25). However, in T2 cancer, there is still some controversy as to the extent of surgery and adjuvant treatment plans that should be followed (26). Therefore, studies on T2 GBC have been actively undertaken (27-29). One of them examines tumor location ("hepatic side" or "peritoneal side"). Hepatic side T2 GBC offers a poor prognosis compared to the peritoneal side (30,31). Thus, we believe tumor morphology could be another influencing factor like tumor location and tumor stage.

Conclusions

In conclusion, flat type GBCs are associated with a worse prognosis than tumoral GBCs. Thus, unusual thickening of the GB wall, which may lead to malignancy, needs to be assessed with care. Morphologic classifications of flat or tumoral on the basis of length (1 cm) and papillary features are feasible. Additionally, the borderline group possesses papillary features as well as a length of 1 cm; yet, overall, the group reflects the characteristics of the disease manifested by the flat type GBC. The most powerful prognostic factor is the T stage in GBC. Thus, tumor morphology can be utilized as a reference when deciding on the treatment plan, especially in T2 GBC.

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Footnote

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Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The present study protocol was reviewed and approved by the Institutional Review Board of the Seoul National University Hospital (approval No. 1508-081-695). The process of obtaining informed consent was waived according to the decision of the IRB.

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