Gallbladder selection for histopathological analysis based on a simple method: a prospective comparative study

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ABSTRACT

INTRODUCTION After a cholecystectomy, the current and traditional practice is to send each resected gallbladder to the pathologist for analysis. Some reports have suggested the possibility of selecting only those gallbladders that need to be analysed. The purpose of this study was to show a simple method for selecting which gallbladders should be sent to the pathologist.

METHODS A prospective comparative study was carried out. Two 'tests' were performed in 150 patients to detect or rule out gallbladder cancer. The first test included the patient's variables and a macroscopic gallbladder analysis performed by the surgeon (MGAS). The second test was the analysis performed by the pathologist. The results were compared.

RESULTS Of the 150 patients, 132 were women and 18 men; 130 were under 60 years old. One patient had inflammatory bowel disease, seven had changes on ultrasonography and in four cases intra-operative disturbances were observed. During the MGAS, disturbances were found in 30 patients. Eighty-one cases (54%) had at least one or more risk factors for gallbladder cancer.

CONCLUSIONS In almost half of the gallbladders, it would be safe not to send the specimen to the pathology department, decreasing costs significantly.

KEYWORDS

Gallbladder – Adenocarcinoma – Cholecystectomy – Pathology – Diagnosis

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Gallbladder cancer is a very rare malignancy; the overall incidence has been reported as between 0.5% and 2%.^{1,2} This disease predominantly affects older women over the seventh decade of life. The most common histological type is adenocarcinoma.^{5–6} This is a very aggressive neoplasm that has an extremely poor prognosis; 5-year survival rates of between 5% and 15% have been reported.^{7–9} It is believed that the most significant risk factor for gallbladder cancer is the presence of chronic inflammation in the gallbladder wall, which is related to gallstones in 74–92% of cases.^{10,11}

Despite the advances in radiological imaging, accurate pre-operative diagnosis is rare and most carcinomas are detected late in the course of the cancer. Simple cholecystectomy is reserved for patients in the T1 stage. Most T2b carcinomas will also be treated with hepatic resection and a regional lymphadenectomy.^{12,13} Gallstone disease affects 10–15% of the Western population with an annual incidence of 1 in 200. It is estimated that Mexicans have a prevalence of 6% in men and 20% in women and in countries such as Chile asymptomatic stones are present in up to 50% of the population.^{14,15}

The treatment of choice for gallstone disease is laparoscopic cholecystectomy with histopathological analysis of the specimen. By sending each resected gallbladder to the pathologist, costs generated for the health system are very high. In a large retrospective study published in 1998, Taylor and Huang suggested the possibility of analysing selected gallbladders only.¹⁶ After this report, some authors proposed the same hypothesis.^{17,18} Nevertheless, most surgeons send every specimen for analysis. The aim of the present study was to compare results between two 'tests' for detecting or ruling out gallbladder cancer in order to be able to select gallbladders that should be sent to the pathology department. The first test comprised a simple method to be carried out by the surgeon and the second was histopathological analysis.

Methods

A prospective comparative study was conducted in one institution between August 2010 and February 2011. Patients

with gallstone disease with surgical intervention at our institution were included in the study. Before the procedure, a specific interrogatory, a physical exploration, routine lab tests (complete blood count, liver function test and, in selected cases, blood chemistry) and abdominal ultrasonography were carried out. The procedure performed in all cases was a cholecystectomy, either laparoscopic or open.

Two 'tests' were carried out for each case: one by the main surgeon and, subsequently, one by the pathologist. The results from both tests were compared. This was a double blind study as the surgeon performed his/her test before the specimen was sent to the pathologist and the pathologist was not informed of the surgeon's observations. The protocol was reviewed and approved by the relevant bioethics committee.

First test: clinical examination by surgeon

The initial test was conducted by 20 surgeons or residents without previous training in macroscopic gallbladder analysis. It was divided into two stages: first, the identification of risk factors and, second, the detection or ruling out of cancer.

During the first stage, the positive risk factors for gallbladder cancer were sought. A patient was considered high risk if he or she had one or more of the following risk factors:

- > age ≥ 60 years;
- > history of inflammatory intestinal disease;
- > ultrasonography alterations (performed by a Logiq® 7 [GE Healthcare, Little Chalfont, UK] with a 3.5 MHz transducer) that include disproportionate gallbladder wall thickness, polyps, masses and/or ulcers;
- > pre-operative and/or intra-operative diagnosis of acute or chronic cholecystitis,^{19,20} which was made by the main surgeon and included intractable pain, presence of Murphy's sign, fever, white blood cell elevation, radiological suspicion (ie gallbladder wall thickness > 3 mm, pericholecystic fluid, sonographic Murphy's sign, air in the gallbladder wall or lumen);
- > intra-operative disturbances including anatomy alterations, visible or palpable masses, induration and/or ulcers; and
- > disturbances at macroscopic gallbladder analysis performed by the surgeon (MGAS).

The MGAS was started just after the end of the cholecystectomy, still in the operating room, and before sending the specimen to pathology (Fig 1). During the first step of the MGAS, the serosa of the gallbladder was irrigated with water, observed and palpated on its entire surface (Fig 2). In the second step, the gallbladder was incised longitudinally and the mucosa was irrigated, observed and palpated (Figs 3 and 4). During the serosa and mucosa exploration, the surgeon looked for abnormalities that included masses, indurations, calcifications and/or ulcers. Obtained data were collected in an Excel® spreadsheet (Microsoft, Redmond, WA, US). This procedure was performed in less than four minutes in most cases and the only material required was water. The cutting instrument and the gloves were the same that had been used previously during the surgery.

The second stage of the MGAS consisted of a simple question: 'Based on the patient's history, routine laboratory/image

Figure 1 The extracted gallbladder just before the macroscopic analysis performed by the surgeon appears normal. Arrows point to serosa and mucosa.

Figure 2 Observation and palpation of serosa during irrigation with saline solution

tests, intra-operative findings and the macroscopic analysis, do you think that this gallbladder has a malignant disease?' The answer ('yes' or 'no') was documented and later compared with the final diagnosis reported by the pathologist.

Second test: histopathological analysis

After the MGAS, the gallbladder was placed in formaldehyde and sent to the pathology department. The pathologist had no knowledge of the first test performed by the surgeon. The histopathological analysis was performed according to guidelines.²¹ The gallbladder was observed and palpated searching for lesions, masses or abnormalities on the serosal and mucosal surfaces. If any alteration was found, a sample was taken. After this, three wall samples were taken from the fundus, body and neck. This is because cancer

Figure 3 Longitudinal cut to expose the mucosal surface

Figure 4 Cleaned mucosal surface

lesions originate predominantly in these sites.²² The obtained pieces, either from a lesion or from the three routinely biopsied areas, were observed microscopically. If a malignant lesion was confirmed, a proper report was prepared and information was given to the treating surgeon.

Statistical analysis

Descriptive statistics were used to summarise the overall information. A contingency table was used to compare the tests. Data were analysed using SPSS® version 14.0 (SPSS, Chicago, IL, US). Inter-rater reliability to examine the agreement between the 20 treating surgeons/residents who performed the first test was not assessed in this study.

Results

Data from 150 cases were collected (132 women [88%], 18 men [12%]). The mean patient age was 39 years (standard

Table 1 Cases with risk factors for gallbladder cancer	
Risk factor	Cases (n=150)
Age ≥60 years	20 (13.4%)
History of inflammatory bowel disease	1 (0.6%)
Image alteration	7 (4.6%)
Pre-operative or intra-operative suspicion of acute or chronic cholecystitis	65 (43.4%)
Intra-operative disturbances (anatomy alteration, visible or palpable masses, induration, ulcers)	4 (2.6%)
MGAS disturbances (serosal and/or mucosal)	30 (20%)
Cases with one or more risk factors	81 (54%)

MGAS = macroscopic gallbladder analysis performed by the surgeon

Table 2 Comparison between the two 'tests'	
Pathologist	
Surgeon's test	Positive
	Negative
Positive	3
Negative	0
	147
	3
	147
95% confidence interval	
Sensitivity	100% (49–100%)
Specificity	100% (98.7–100%)
Likelihood ratio (+)	252 (15.3–4,100)
Likelihood ratio (-)	0.14 (0.01–1.8)
Positive predictive value	85.7 (49–100)
Negative predictive value	99.6 (98.7–100)

deviation: 16.05 years). Of the 150 patients, 130 (86.6%) were under 60 years old. The histopathological analysis confirmed three gallbladder adenocarcinomas (2%). One patient (0.6%) had a history of inflammatory bowel disease. Seven (4.6%) had alterations on ultrasonography; two of these had malignant disease confirmed by the pathology analysis. Intra-operative disturbances were observed in four cases (2.6%), with three of these having malignant disease. Acute or chronic cholecystitis was diagnosed pre-operatively and/or intra-operatively in 65 cases (43.4%). Stones were found in 147 cases (98%).

Table 3 Characteristics of patients with gallbladder adenocarcinoma. In all three cases, macroscopic gallbladder analysis was performed by the surgeon but a previous biopsy had already been obtained because of obvious intra-operative malignant features. In the third case, it was difficult to ascertain whether the primary neoplasm originated in the pancreas or the gallbladder.

Age/sex	History	Ultrasonography	Intra-operative findings	Clinical stage	Histopathological diagnosis
35F	Abdominal colic pain	Areas of wall thickening	Gallbladder wall and liver induration, palpable ganglia near choledocus	T3N1M0 (stage IIIB)	Adenocarcinoma with invasion beyond serosa
66F	Jaundice	–	Hartmann's pouch wall and choledocus induration	T3N0M0 (stage IIIA)	Adenocarcinoma with invasion beyond serosa
64F	Jaundice and weight loss	Solid mass in head of pancreas confirmed by computed tomography	Gallbladder wall and liver induration, visible liver metastasis	T4N0M1 (stage IV)	Adenocarcinoma with invasion beyond serosa

Table 4 Comparison between the three cancer cases and the proposed risk factors

	Case 1	Case 2	Case 3
Age ≥60 years		✓	✓
Presence of inflammatory bowel disease			
Image alterations	✓		✓
Suspicion of cholecystitis	✓	✓	✓
Intra-operative disturbances	✓	✓	✓
MGAS disturbances	✓	✓	✓
One or more risk factors	✓	✓	✓

MGAS = macroscopic gallbladder analysis performed by the surgeon

During the MGAS, alterations of the serosa were found in 18 gallbladders (12.0%) and alterations of the mucosa in 22 (14.6%). In 127 cases (84.6%), no serosal or mucosal alterations were found. The risk factors are summarised in Table 1.

The second stage was assessed using the response to the question 'Based on the patient's history, routine laboratory/image tests, intra-operative findings and the macroscopic analysis, do you think that this gallbladder has a malignant disease?' made to the main surgeon; the answer was 'no' in 147 cases (98%) and 'yes' in 3 (2%). Pathology reports were similar to surgeons' reports, confirming the presence of neoplasm in the same 3 patients and ruling it out in the other 147. Atypical lesions of the mucosa not associated with gallbladder cancer were not reported by the pathology department.

Table 2 shows the comparison between the two tests. The specific characteristics and positive risk factors of the three cases with malignant disease are listed in Tables 3 and 4.

Discussion

Gallbladder cancer continues to be a very rare entity. Even though some evidence suggests the possibility of diagnosing this malignant disease before pathology analysis, most centres continue to send every extracted gallbladder to the pathology department for its routine analysis. This is mainly because of the lack of large and prospective studies that can give solid evidence to support the hypothesis that sending every gallbladder to the pathologist is not always necessary.

Nevertheless, we believe that selective gallbladder analysis performed by the pathologist (SGAP) is adequate, offering reassurance to the patient and the physician. Taking into account that the incidence of gallbladder cancer is very low and that it is known that more than 90% of patients are over 50–60 years old,²⁵ then the incidence of gallbladder cancer among people under 60 would be around 0.2–0.3%. Even though the percentage is extraordinarily low, it would be unacceptable to miss a neoplasm in a young patient.

In 2010 Mittal *et al* reported on a 10-year retrospective series in which they detected 13 gallbladder carcinomas.²⁴ Suspicion was raised pre-operatively and/or intra-operatively in every case. Furthermore, the authors of a UK study in 2007 concluded: 'All cases of invasive carcinoma of the gall bladder showed gross macroscopic abnormal appearance either pre- or intra-operatively. A more selective policy to histological examination of the gall bladder specimens would not miss any invasive malignancy.'²⁵

Unfortunately, the few papers that suggest the possibility of SGAP are retrospective. Prospective, better designed studies are necessary to support the hypothesis. Without a doubt, the most important reason to continue with routine gallbladder analysis performed by the pathologist is the possibility of missing a resectable gallbladder adenocarcinoma. This would have a devastating effect on the patient. The increase in lawsuits and surveillance of daily medical practice makes it difficult to change such procedures and SGAP continues to be just an interesting and almost impossible practice.

Given the epidemiological evidence, we believe that the possibility of missing an invasive gallbladder carcinoma pre or intra-operatively is extremely low. What would therefore

happen if we only sent inflamed or abnormal gallbladders for pathologic analysis and those from patients over 60 years of age? Based on the previously mentioned studies, the possibility of missing a neoplasm would be negligible as most gallbladder cancer cases are diagnosed in people over 60 and/or with wall inflammation and macroscopic abnormalities.^{26,27} In spite of the epidemiological evidence, we think it is necessary to perform more tests to be able to provide sufficient certainty to patients while selecting gallbladders for histopathological analysis.

In this study, the main surgeon conducted a macroscopic analysis of the extracted gallbladder. The material cost was close to zero and the time spent was very low. The objective of MGAS is to select cases that for one reason or another should have SGAP. When examining the gallbladder, pathologists use both macroscopic and microscopic analysis. The latter is performed from samples either from macroscopically altered tissue or from areas where carcinomas are most frequently found (fundus, body and neck). Macroscopic examination probably plays the most important role in gallbladder analysis.²¹ It may therefore be possible for MGAS to replace the macroscopic assessment carried out by the pathologist.

Some may argue that a pathologist is more experienced than a surgeon at performing meticulous gallbladder analysis but the frequent exposure to the serosal surface during cholecystectomies could mean that surgeons gain sufficient experience to detect abnormal patterns. However, we believe that a complete mucosal and serosal analysis is necessary to definitely rule out a carcinoma. This being the case, it may be feasible for a surgeon to perform a meticulous gallbladder analysis and send only specific specimens to be analysed microscopically.

In our study, all 150 cases had similar reports when comparing MGAS and pathology analysis: 147 patients with no malignant lesions and 3 cases of cancer. In the three cancer cases, MGAS was performed despite not being required since the neoplasm diagnosis was made intra-operatively and confirmed by trans-operative biopsy. Although we think it is possible to diagnose gallbladder cancer before the pathological analysis, we nevertheless limit ourselves to only proposing that a selection of the specimens with risk factors are sent to the pathologist. In this study, the three cases with malignant disease had more than one risk factor (Table 4). It should be noted that in the proposed algorithm only one risk factor is necessary to send the specimen to the pathologist. In 69 of the 150 cases (46%) there were no risk factors and in 81 cases (54%) there was at least one (Table 1).

It is probable that a method for SGAP can save a lot of human and material resources. This is particularly necessary in low income hospitals where economic improvements in other areas could increase benefits for more patients. On the other hand, there may be a risk of missing a dysplastic lesion or a T1 gallbladder carcinoma due to its small size and less obvious macroscopic features. Based on the epidemiological analysis, the possibility of missing one of these lesions is extremely low, especially if gallbladders from patients with advanced age and inflammation suspicion are sent to the pathology department.²⁸ In addition, the actual

treatment for such lesions is a simple cholecystectomy so the patient would have been treated unintentionally in those cases.

Conclusions

We think that in almost half (46%) of the extracted gallbladders it would be safe not to send the specimen to the pathology unit without compromising patient safety. As a result, the budget can be halved. In this study, we have shown a simple and non-expensive method for selecting gallbladders that must be sent to the pathology department. Nevertheless, we believe that further studies with a greater number of cases are necessary to confirm our hypothesis. If we can decrease human and material resources needed for the traditional method for detecting gallbladder carcinoma, we can use those resources in other areas where they are needed more. This would be especially useful in hospitals with a low income.

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