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Histopathologic Features of 1000 Cholecystectomy Specimens

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Abstract

Background: One of the most common gastrointestinal surgeries is cholecystectomy. Gallstones are the major causes of cholecystectomy and induce various histopathologic changes. Gallbladder carcinoma is rare with poor prognosis. Metaplasic changes in gallbladder epithelium are considered precancerous lesions.
Objectives: This study aimed at revealing the spectrum of histopathological patterns of gallbladder diseases in cholecystectomy specimens.
Methods: A total of 1004 gallbladder H & E stained slides of cholecystectomy specimens over one year were studied. Histopathological evaluation was done by 6 pathologists in terms of variables.
Results: In our study, the male: female ratio was 1:2.1. The age of patients varied from 17 to 96 years with a mean age of 49.1 years. The mean age of patients with gallstones was 47.52 years and was more common in women. Cholesterol, pigmented, and mixed stones were reported in 58.36%, 39.03%, and 2.6% of gallstones, respectively. The most common histological morphology was chronic calculous cholecystitis (61.18%). The incidence of metaplasia and invasive carcinoma was 19.55% and 0.2%, respectively.
Conclusions: In this study, histopathological analysis of gallbladder diseases in the cholecystectomy specimens revealed that chronic calculous cholecystitis was the most common histologic finding. Gallstones were found more commonly in women and cholesterol stone was the predominant type. Pyloric metaplasia was the most frequent type of metaplasia. The rate of invasive carcinoma is compatible with those reported in the literature. Routine histopathological analysis of the cholecystectomy specimens will help to detect incidental carcinoma and precursors of malignancy.

Keywords: Gallbladder diseases, Cholecystectomy, Histopathology

1. Background

The gallbladder is a pyriform sac that is attached to the extrahepatic biliary system, below the right lobe of the liver. It measures up to 10 cm in length and 3 to 4 cm in diameter in normal adults, and its wall thickness is approximately 1 to 2 mm (1).

The wall of the gallbladder is composed of 3 layers, namely mucosa, muscular, and perimuscular layers. Unlike the other parts of the gastrointestinal tract, the gallbladder does not have a muscularis mucosa and submucosa. A few lymphocytes, plasma cells, and mast cells are present in the lamina propria but neutrophils and eosinophils are normally absent in the lamina propria (2).

Cholecystectomy is one of the most common surgical procedures, and gallstones are the main cause of this operation globally. The prevalence of gallstones has been reported 10% to 15% in the European population and 3% to 5% in the Asian population (1). In the last decade, the rising environmental risk factors like obesity and metabolic syndrome have been caused by the increased frequency and complications of gallstones (3-5).

Gallbladder cancer is a rare gastrointestinal malignancy, but it has a poor prognosis with low survival (2). Gallstones were detected in 85% of the cancerous cases, although the incidence of gallbladder cancer in patients with gallstones is 0.5% (6).

According to the literature, in only 30% of the patients, gallbladder cancer was diagnosed preoperatively and other cases were detected incidentally by a surgeon during cholecystectomy or after the surgery by a patholo-

gist. Incidental gallbladder cancer was diagnosed in 0.2% to 3% of all cholecystectomies (7-10).

Among gastrointestinal diseases, gallbladder-related illness is the main cause of hospital admissions (11). In the United States, more than 900 000 cholecystectomies are performed annually.

The cost of Gallbladder disease in the U.S is estimated at $6.2 billion annually (12). Therefore, gallbladder disease is the most common and costly gastrointestinal disease worldwide.

2. Objectives

There is limited information about the incidence of histopathologic findings in patients, who underwent cholecystectomy for gallbladder disease in Iran and the aim of this study was the histopathological assessment of gallbladder after cholecystectomy concerning the prevalence of incidental and unexpected findings.

3. Methods

This descriptive study was performed in hospitals of Shahid Beheshti University of Medical Sciences, Tehran, Iran between March 2016 and March 2017 and enrolled 1004 open and laparoscopic cholecystectomy specimens. The H & E stained slides of the specimens were selected from archives of Pathology Departments of Loghman-e-Hakim, Shohada-Tajrish, and Moddares Hospitals. The ethics committee of Shahid Beheshti University of Medical Sciences approved the study (code: IR.SBMU.RETECH.REC.1397.542).

All histologic slides were reviewed by 6 pathologists in terms of variables. The variables of the study included stromal lymphocyte count/HPF (×400), intraepithelial lymphocyte count per 100 epithelial cells, eosinophil count/HPF (×400), macrophage count/HPF (×400), neutrophil count/HPF (×400), the coexistence of different types of metaplasia, benign tumor, malignant tumor, cholesterolosis, polyps, diverse stones, and wall thickness. Besides, the patient's age, sex, and indications for surgery were retrieved from pathology reports and were recorded independently. The data were analyzed by SPSS 16 software. Also, the data were evaluated as mean, percentage, and minimal and maximal values.

4. Results

We evaluated 1004 gallbladder specimens from March 2016 to March 2017. There were 680 females (67.72%) and 324 (32.27%) males, and the male-to-female ratio was 1:2.1.

The age of 994 out of 1004 patients was recorded ranging between 17 to 96 years with the mean age of 49.1 years. The majority of patients were in the age group between 31 and 40 years (Table 1). The mean stromal lymphocyte count per high-power field (×400) in 992 patients was 38.44/HPF. The majority of them had 11 to 30 stromal lymphocytes/HPF. The presence of intraepithelial lymphocytes in the biliary epithelium was reported in 887 of 1004 patients with a mean value of 8.95/HPF. Most patients (n = 380) had less than 5 IEL/HPF, and only 7 patients had 40 50 IEL/HPF.

Table 1. Comparison of Age Distributions[a]

Age Group	Total	Male	Female
11 - 20	13 (1.3)	3	10
21 - 30	119 (11.97)	25	94
31 - 40	232 (23.34)	66	166
41 - 50	172 (17.3)	49	123
51 - 60	205 (20.62)	71	134
61 - 70	145 (14.58)	55	90
71 - 80	86 (8.85)	41	45
81 - 90	21 (2.11)	11	10
91 - 100	1 (0.1)	0	1

[a]Values are expressed as No. (%).

The mean stromal plasma cell count in 992 patients obtained 15.39/HPF. The majority of patients (n = 624) had less than 10 plasma cells/HPF. Twenty-six patients had more than 51 plasma cells/HPF.

Macrophages in the lamina propria of 992 gallbladder specimens were counted 13.85/HPF on average. Less than 10 macrophages/HPF were seen in most patients (n = 690). The mean neutrophil count in 992 patients was 15.36/HPF. Most specimens (n = 470) had no neutrophil in the lamina propria and 15 patients had over 101 neutrophils/HPF.

Also, we reported the eosinophil number/HPF. The mean count was 7.53 and the majority of specimens (n = 687) had 0 to 5 eosinophils/HPF.

In the present study, the density of inflammatory cells in the gallbladder stroma and IEL were compared between calculous/acalculous, acute, and chronic cholecystitis (Table 2).

We found that the mean age of the patients with gallstones was 47.52 years and the majority of them were women.

Most gallstones were not sent to the pathology department and only 348 out of 607 cases of gallstones were available for assessment, which measured from 1 to 30 mm in diameter. A total of 288 out of 348 patients had multi-

Table 2. Comparison of Inflammatory Cells in the Gallbladder Stroma and IEL in Acute Calculous Cholecystitis (ACC), Acute Acalculous Cholecystitis (AAC), Chronic Calculous Cholecystitis (CCC), and Chronic Acalculous Cholecystitis (CAC)[a]

	ACC	AAC	CCC	CAC
IEL	7.91 (2 - 50)	8.66 (2 - 20)	8.83 (1 - 50)	10.88 (2 - 55)
Stromal Lymphocytes	42.06 (5 - 100)	27.27 (10 - 80)	33.98 (5 - 150)	33.59 (8 - 100)
Stromal Plasma cells	21.97 (2 - 200)	22.27 (5 - 70)	11.36 (0 - 100)	37 (2 - 100)
Stromal Machrophages	15.69 (5 - 200)	31.54 (5 - 100)	31.54 (5 - 100)	21.48 (5 - 100)
Stromal Neutrophils	29.82 (0 - 200)	57 (2 - 100)	57 (2 - 100)	0.027 (0 - 1)
Stromal eosinophils	15.33 (0 - 200)	11.09 (0 - 40)	11.09 (0 - 40)	4.37 (0 - 30)

[a]Values are expressed as median (range).

ple stones and 60 patients had a single stone. In terms of gallstone chemical composition, 3 stone types were reported in 269 patients according to Table 3. The most common type was cholesterol stones with a higher frequency in women.

Table 3. Types of Gallstones[a]

Type of gallstone	Number of Cases	Number in Male	Number in Female
Cholesterol stones	157 (58.36)	42	115
Pigmented stones	105 (39.03)	44	61
Mixed stones	7 (2.6)	2	7

[a]Values are expressed as No. (%).

Indications for surgery in 597 patients were acute (n = 135, 22.42%) and chronic cholecystitis (n = 462, 76.74%) and other causes in 0.83% (n = 5) cases. Histomorphological diagnoses are shown in Table 4.

Histopathological examination showed that chronic calculous cholecystitis was the most common histological finding (61.18%) with a female predominance. Chronic acalculous cholecystitis was seen in 3.72% of the cases.

Other variants of cholecystitis including hyalinizing chronic cholecystitis, chronic active cholecystitis, chronic granulomatous cholecystitis, follicular cholecystitis, eosinophilic cholecystitis, and xanthogranulomatous cholecystitis were found predominantly in women.

Two kinds of gallbladder polyps including cholesterol and adenomatous polyps were seen in 0.1% and 0.5% of the cases, respectively.

Metaplasias (pyloric, intestinal, and pancreatic acinar) were detected in 19.55% of the patients with a mean age of 45.71 years and were most common in women.

Pyloric metaplasia was the predominant type of metaplasia and was observed in 16.43% (n = 163) of the cases with a female predominance. Intestinal metaplasia and pancreatic acinar metaplasia were seen in 2.92% (n = 29) and 0.2% (n = 2) of the cases, respectively.

One case with preinvasive epithelial neoplasia was detected in a 55-year-old man. There was one case with carcinoma in situ (0.1%), one with invasive adenocarcinoma (0.1%), and one showing mucinous adenocarcinoma (0.1%).

Acute calculous cholecystitis and acute acalculous cholecystitis were reported in 15.12% and 1.1% of cases, respectively, with a mean age of 51.01 and 44.81 years in order and female predominance in both. Empyema was identified in two cases.

The gallbladder wall thickness in 939 patients was reported measuring 1 to 45 mm. The mean thickness was 4.26 mm. In most cases (82.64%), the wall thickness was less than 5 mm.

The gallbladder wall thickness in calculous/acalculous chronic and acute cholecystitis are shown in Table 5. Acute acalculous cholecystitis cases had the thickest wall in comparison to other types of cholecystitis. A total of 161 patients with cholecystitis had normal wall thickness (< 3mm).

5. Discussion

Gallbladder disease is a common and costly health issue worldwide. The present study was the largest study of its kind in Iran that evaluated different histopathological patterns of gallbladder disease in cholecystectomy specimens.

In this study, we found that the gallbladder diseases were more common in women than men with a male: female ratio of 1:2.1. These findings are consistent with previous studies (13-15).

The age of patients ranged from 17 to 96 years and the majority of them were in the age group of 31 to 40 years, which is similar to other studies (16-19); however, Kumar et al. (20) reported that more patients were in the age group of 41 to 50 years.

We realized that the prevalence of gallstones was higher in women than in men. Cholesterol gallstone was

Table 4. Histopathological Diagnoses[a]

Histomorphological variants	Number of Cases	Number in Men	Number in Women	Mean Age
Chronic calculous cholecystitis	607 (61.18)	155	452	47.52
Chronic acalculous cholecystitis	37 (3.72)	12	25	46.76
Hyalinizing chronic cholecystitis	2 (0.2)	0	2	65
Chronic active cholecystitis	93 (9.37)	41	52	50.26
Chronic granulomatous cholecystitis	1 (0.1)	0	1	44
Follicular cholecystitis	3 (0.3)	1	2	61.69
Eosinophilic cholecystitis	3 (0.3)	1	2	29.66
Xanthogranulomatous cholecystitis	4 (0.4)	2	2	29.66
Acute calculous cholecystitis	150 (15.12)	63	87	51.01
Acute acalcolous cholecystitis	11 (1.1)	5	6	44.81
Acute gangrenous cholecystitis	95 (9.57)	53	42	54.86
Emphysematous cholecystitis	1 (0.1)	1	0	74
Emyema	2 (0.2)	1	1	73
Cholesterolosis	173 (17.43)	36	137	41.33
Gallbladder hydrops	1 (0.1)	1	0	69
Cholesterol polyp	1 (0.1)	0	1	57
Adenomatous polyp	5 (0.5)	1	4	44.8
Pyloric metaplasia	163 (16.43)	45	118	51.1
Intestinal metaplasia	29 (2.92)	6	23	52.55
Pancreatic acinar metaplasia	2 (0.2)	0	2	33.5
Preinvasive epithelial neoplasia	1 (0.1)	1	0	55
Carcinoma insitu	1 (0.1)	0	1	75
Invasive adenocarcinoma	1 (0.1)	0	1	75
Mucinous carcinoma	1 (0.1)	1	0	71

[a]Values are expressed as No. (%).

Table 5. Comparison of Gallbladder Wall Thickness in Acute Calculous Cholecystitis (ACC), Acute Acalculous Cholecystitis (AAC), Chronic Calculous Cholecystitis (CCC), and Chronic Acalculous Cholecystitis (CAC)

Wall Thickness	ACC	AAC	CCC	CAC
Range	1 - 20	2 - 15	1 - 17	1 - 4
Mean	4.77	6.27	3.61	2.88
Normal (< 3mm)	22	2	109	10

the predominant stone type in our study and was more common in women. These results are in line with previous studies (16, 21); however, Semnani et al. (22) and Tyagi et al. (23) reported the mixed gallstone as the predominant type.

The evaluation of different types of inflammatory cell infiltration in the gallbladder stroma revealed that in acute calculous cholecystitis, the mean stromal lymphocyte and eosinophil counts were higher than acute acalculous cholecystitis and chronic cholecystitis. The average plasma cells count in chronic acalculous cholecystitis was higher than other types of cholecystitis. Stromal neutrophil and macrophage counts were higher in acute acalculous cholecystitis compared to other types of cholecystitis.

Histopathological examination showed that chronic calculous cholecystitis was the most frequent pathology in the gallbladder specimens (61.18%), which is consistent with the results of other studies (15, 17, 19, 24, 25). The mean age of patients with this pathology was 47.52 years and it was more common among women. From other variants of

chronic cholecystitis, hyalinizing chronic cholecystitis was observed in two female patients with the mean age of 65 years, chronic granulomatous cholecystitis in a 44-year-old woman, follicular cholecystitis in 16 cases with female predominance, eosinophilic cholecystitis in 3 cases with the mean age of 29.66 years, and xanthogranulomatous cholecystitis in 4 cases.

Acute cholecystitis (including acute calculous, acute acalculous, and acute gangrenous cholecystitis) was diagnosed in 25.8% of the cases. Acute calculous cholecystitis was the most common form of acute cholecystitis in our study, which is compatible with the literature.

This study showed cholesterolosis in 17.14% of the cholecystectomies with higher incidence compared to other studies such as those conducted by Sharma and Choudhury (15) (4.4%), Kumbhakar (19) (7.5%), and Khan et al. (17) (10%). Cholesterolosis was more common in females, which is similar to the results of previous studies (17, 26).

Gallbladder polyps were detected in 0.6% of the patients with higher frequency in women; although Aldouri et al. (27) reported polyps in 3.3% of the patients and Krishna et al. (cited in Patel et al.) (28) detected polyps in 1.09% of the patients.

In our study, different types of metaplasia were seen in 19.55% of the evaluated specimens. The overall prevalence of metaplasia in studies of Seretis et al. (29), Kumer et al. (cited in Tiwari et al.) (20), and Sharma et al. (30) were 25.6%, 23.75%, and 46.2%, respectively.

In this study, intestinal and pyloric metaplasia were seen in 2.92% and 16.43% of the cases, respectively. Although, Sharma et al. (30) reported intestinal metaplasia in 15.97% and pyloric metaplasia in 42.86% of gallbladders. The prevalence of intestinal and pyloric metaplasia in Segovia Lohse and Cuenca Torres's (31) study was reported 2.1% and 22.6%, respectively.

We noted that pyloric metaplasia was more common than other types of metaplasia, which is comparable to previous studies (30, 32-34). The mean age of the patients with pyloric and intestinal metaplasia was 51.1 and 52.55 years, respectively. These results were in accordance with previous studies (33, 35, 36); intestinal metaplasia and pyloric metaplasia were observed in patients over 50 years old. Sharma et al. (30) illustrated that pyloric and intestinal metaplasia were observed in patients with a mean age of 42.5 and 43 years, respectively.

According to previous studies (35, 37), pyloric and intestinal metaplasia were associated with gallbladder cancer and were considered as precancerous lesions.

Previous studies demonstrated that patients with metaplasia were younger than those with dysplasia and cancer. The incidence of precancerous lesions (intestinal and pyloric metaplasia) might increase by age and will be present in patients over 50 years; these facts were confirmed by our study (35, 37).

Pancreatic acinar metaplasia is a rare condition in the gallbladder that was reported in two female cases (0.2%) in our study.

The mean age of pancreatic acinar metaplasia was 33.5 years, which was presented at a younger age compared to other types of metaplasia.

Two cases of gallbladder carcinoma were recorded in our study (0.2%). One of them was a case of invasive adenocarcinoma in a 75-year-old woman and the other was a case of mucinous carcinoma in a 71-year-old man. In our study, the rate of primary invasive carcinoma was 0.2%, which is compatible with the rate reported in the literature (0.15% - 2.3%).

Two cases of empyema (0.2%) were noted in our study with a similar rate to Khan et al.'s (17) study.

In only 17.14 % of the cases, the gallbladder wall thickness was normal and the rest of the specimens (82.86%) showed a thickened wall of $\geq$ 3mm. Awasthi (38) showed normal wall thickness in 72.8% of the cases and increased wall thickness in 27.2% of the specimens. We found that the gallbladder wall thickness was higher among the acalculous acute cholecystitis group compared to other types of cholecystitis.

5.1. Conclusions

Cholecystectomy is one of the most common surgical procedures globally. This study described different histopathological patterns of gallbladder diseases and their frequency in the cholecystectomy specimens. We concluded that gallbladder diseases were more common in women. Chronic calculous cholecystitis was the most common pathologic change with a female predominance. The most frequent form of acute cholecystitis was acute calculous cholecystitis. Pyloric metaplasia was noted as the most common type of metaplasia. The results of our study showed that the incidence of invasive carcinoma is compatible with those of literature. The majority of the patients with gallstones were women and the cholesterol type was the predominant stone type. Finally, our results emphasize the importance of routine histopathological examination to detect incidental carcinoma and precancerous lesions in cholecystectomy specimens.

Footnotes

Authors' Contribution: Study concept and design: EJ and MA. Acquisition of data: MA, EJ, TM, BK, AS, JK, HH, SA, and MR. Analysis and interpretation of data: MA, EJ, TM, SA,

BK, HH, and MA. Drafting of the manuscript: EJ, MA, and MA. Critical revision of the manuscript for important intellectual content: MA, EJ, and MA.

Conflict of Interests: There is no conflict of interest in this research.

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