

# HISTOPATHOLOGICAL SPECTRUM OF CHOLECYSTECTOMY SPECIMENS: A RETROSPECTIVE CROSS-SECTIONAL STUDY

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## Abstract

**Background:** Cholecystectomy is one of the most frequently performed abdominal surgical procedures worldwide. Although routine Histopathological examination of cholecystectomy specimens is standard practice, its diagnostic utility in identifying incidental pathologies, particularly premalignant and malignant lesions, remains clinically significant.

**Objective:** This study aimed to investigate the histomorphological patterns in 1036 cholecystectomy specimens and emphasize the utility of routine Histopathological examination for precise diagnosis.

**Material and Methods:** A retrospective analysis of 1036 cholecystectomy specimens was conducted from January 2017 to December 2019 at Liaquat National Hospital, Karachi. Standard Histopathological processing and examination techniques were employed. Patient demographics and Histopathological diagnoses were analyzed using descriptive statistics. All consecutive cholecystectomy specimens received during the study period were included irrespective of age or gender. Specimens with inadequate fixation, and incomplete clinic pathological data were excluded. Data were analyzed using SPSS software version 20.0 with frequency distributions and percentages calculated for all variables.

**Results:** Chronic cholecystitis was identified in 421 cases (40.6%), followed by acute cholecystitis in 208 cases (20.1%). These two entities accounted for 60.7% of all specimens examined. Intestinal metaplasia in chronic cholecystitis occurred in 50 cases (4.82%). Dysplasia was detected in 6 cases (0.57%), representing a significant pre-malignant finding. Invasive gallbladder carcinoma was identified in 8 specimens (0.77%). A marked female predominance was observed across major diagnostic categories.

**Conclusion:** Routine histopathological examination of cholecystectomy specimens is essential for detecting diverse gallbladder pathologies and identifying incidental pre-malignant and malignant lesions. Detection of dysplasia in 0.57% and carcinoma in 0.77% of specimens underscores the clinical importance of routine histological evaluation.

**Keywords:** Cholecystectomy; Histopathology; Chronic cholecystitis; Acute cholecystitis; Dysplasia; Gallbladder carcinoma

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## Introduction:

Cholecystectomy represents one of the most frequently performed abdominal surgical procedures globally. During the past decade, the rate of surgical gallbladder removal has increased by approximately 50%, with more than 900,000 procedures performed annually in the United States alone, and over 50,000 cholecystectomies conducted in Pakistan each year. The healthcare burden associated with these procedures is substantial, with annual costs in developed nations ranging from \$6 to \$8 billion.<sup>1-5</sup>

Although the clinical indication for cholecystectomy may appear straightforward, routine histopathological

examination frequently reveals unexpected pathological findings that significantly alter clinical management. These include incidental dysplasia, pre-malignant lesions, and unsuspected carcinomas that would otherwise remain undetected.<sup>6</sup>

Proper specimen handling and processing are fundamental prerequisites for ensuring diagnostic accuracy and clinical utility. Swift fixation of the gallbladder in 10% neutral buffered formalin within 30 minutes of surgical removal is essential to prevent bile-related autolysis and preserve tissue architecture and cellular detail. The standardized approach involves

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systematic opening of the specimen from the fundus to the cystic duct, followed by full longitudinal sectioning and representative sampling. When suspicious lesions are identified, additional tissue blocks are obtained. For specimens with focal epithelial atypia, 2-4 paraffin-embedded blocks are generally considered sufficient; however, higher-risk lesions warrant more comprehensive sampling to ensure accurate diagnosis and appropriate patient management.<sup>7</sup>

Therefore, routine histopathological examination of cholecystectomy specimens remains essential for detecting incidental premalignant and malignant lesions. The aim of this study was to evaluate the histomorphological spectrum of cholecystectomy specimens and determine the frequency of incidental premalignant and malignant lesions identified on routine Histopathological examination.<sup>8</sup>

## Material and Method:

### Study Design and Setting:

This retrospective cross-sectional study was conducted at the Department of Pathology, Liaquat National Hospital, Karachi, Pakistan, from January 2017 to December 2019.

### Patient Population:

The study population comprised patients undergoing cholecystectomy at Liaquat National Hospital, a tertiary care referral center receiving both elective and emergency surgical cases from outpatient clinics, inpatient services, and referred healthcare facilities. A total of 1036 consecutive cholecystectomy specimens were analyzed.

### Inclusion Criteria:

All cholecystectomy specimens received in the Department of Pathology during the study period were included. Both laparoscopic and open cholecystectomy specimens were eligible. No restriction was placed on patient age or gender.

### Exclusion Criteria:

Specimens with inadequate fixation compromising histological assessment were excluded. Cases with incomplete clinical or pathological data and specimens received from outside the study institution were also excluded.

### Specimen Handling and Processing:

All specimens were fixed in 10% neutral buffered formalin shortly after surgical removal to ensure optimal tissue preservation. Following fixation, specimens were opened longitudinally from the fundus to the cystic duct. Multiple representative sections were obtained from the fundus, body, neck, cystic duct, and any grossly abnormal areas.

Tissues were processed into paraffin blocks using standard embedding techniques. Serial sections of 4–5 µm thickness were cut and mounted on glass slides and stained with hematoxylin and eosin (H&E). Special stains, periodic acid–Schiff Alcian Blue (PAS-AB),

immunohistochemistry, and other ancillary techniques were used when indicated.

### Diagnostic Criteria and Classification:

Histopathological diagnoses were rendered according to internationally recognized classification standards.

Acute cholecystitis was defined by acute inflammatory cell infiltration (predominantly neutrophils) with or without ischemic changes, vascular congestion, edema, and epithelial denudation. Chronic cholecystitis was defined by chronic inflammatory infiltration with fibrosis and smooth muscle hyperplasia.

Dysplasia was categorized as low-grade or high-grade based on architectural and cytological atypia. Epithelial metaplasia was classified into gastric, intestinal, squamous, and pancreatic acinar types. Gallbladder carcinoma was classified according to histological type and grade using standard criteria.

### Histopathological Classification for Analysis:

For statistical purposes, each case was assigned a single mutually exclusive primary diagnosis based on the most clinically significant lesion. Although multiple histopathological abnormalities frequently coexisted within the same specimen, associated findings (e.g., cholesterosis and metaplasia) were recorded separately and analyzed descriptively.

### Statistical Analysis

Patient demographic data (age and gender) and Histopathological diagnoses were recorded in a structured database and analyzed using SPSS version 20.0 (IBM Corp., Chicago, IL, USA).

Descriptive statistics were used. Frequencies and percentages were calculated for categorical variables. Age was analyzed as a continuous variable and summarized using mean and range. Gender distribution was calculated within diagnostic categories. Results are presented as frequency counts and percentages.

## Results:

The study cohort comprised 1036 consecutive cholecystectomy specimens. A marked female predominance was observed (73.1%), as shown in Table 1. The overall age of patients ranged from 00 to 88 years as shown in Figure 1. The highest frequency of cholecystectomy specimens was observed in the 35–54 year age group, followed by the 55–64 year group. The mean age for chronic cholecystitis was 52.3 years (range 18–85), while the mean age for acute cholecystitis was 55.1 years (range 22–88). Premalignant epithelial lesions were most frequently observed in middle-aged adults (35–64 years), whereas gallbladder carcinoma predominantly occurred in elderly patients (>63 years) as shown in Table 2, 3 & 4.

| Table 1. Gender distribution (n = 1036) |           |         |
|-----------------------------------------|-----------|---------|
| Gender                                  | Frequency | Percent |
| Male                                    | 279       | 26.9    |
| Female                                  | 757       | 73.1    |
| Total                                   | 1036      | 100.0   |

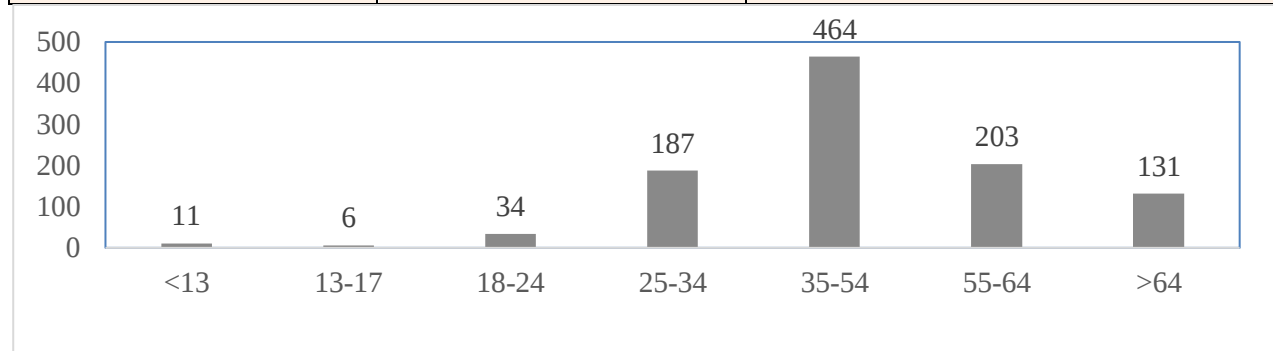


Figure: Frequency of gallbladder lesions in different age groups

| Table 2. Major diagnostic categories in cholecystectomy specimens (n = 1036) |             |              |              |             |
|------------------------------------------------------------------------------|-------------|--------------|--------------|-------------|
| Diagnosis                                                                    | Cases n (%) | AgeRange     | Female n (%) | Male n (%)  |
| Chroniccholecystitis(all variants)                                           | 421 (40.6%) | 52.3 (18–85) | 289 (68.6%)  | 132 (31.4%) |
| Acute cholecystitis(including acute-on-chronic)                              | 208 (20.1%) | 55.1 (22–88) | 145 (69.7%)  | 63 (30.3%)  |
| Intestinal metaplasia (in chronic cholecystitis)                             | 50 (4.82%)  | Mostly 35–64 | 39 (78.0%)   | 11 (22.0%)  |
| Dysplasia (LGD: 4, HGD: 2)                                                   | 6 (0.57%)   | 35–64 yrs    | 5 (83.3%)    | 1 (16.7%)   |
| Gallbladder adenocarcinoma                                                   | 8 (0.77%)   | >63          | 8 (100%)     | 0 (0%)      |
| Total                                                                        | 1036 (100%) |              | 757 (73.1%)  | 279 (26.9%) |

| Table 3. Detailed histomorphological spectrum of gallbladder lesions (n = 1036)                          |      |      |              |           |
|----------------------------------------------------------------------------------------------------------|------|------|--------------|-----------|
| Histomorphological subtype                                                                               | M(n) | F(n) | Age range    | Total (n) |
| Chronic cholecystitis                                                                                    | 101  | 320  | Mostly 25-54 | 421       |
| Acute on chronic cholecystitis                                                                           | 86   | 152  | 25-54        | 238       |
| Acute on chronic cholecystitis with focal intestinal metaplasia, low-grade dysplasia and cholesterol sis | 0    | 1    | 35-54        | 1         |
| Chronic cholecystitis with cholesterol sis                                                               | 31   | 189  | 25-54        | 220       |
| Chronic active cholecystitis                                                                             | 15   | 25   | 35-54        | 40        |
| Chronic cholecystitis with cholesterol sis and low-grade dysplasia                                       | 0    | 1    | >65          | 1         |
| Chronic cholecystitis with focal intestinal metaplasia                                                   | 11   | 39   | 35-54        | 50        |
| Chronic cholecystitis with cholesterolosis and focal intestinal metaplasia                               | 1    | 30   | 35-54        | 31        |
| Acute and chronic cholecystitis with cholesterolosis                                                     | 7    | 13   | 35-54        | 20        |
| Chronic active cholecystitis and focal cholesterolosis with intestinal metaplasia                        | 1    | 2    | 35-54 & >65  | 3         |
| Acute on chronic cholecystitis with focal intestinal metaplasia                                          | 1    | 7    | >65          | 8         |
| Acute necrotizing cholecystitis                                                                          | 13   | 11   | >65          | 24        |
| Follicular cholecystitis                                                                                 | 0    | 2    | >65          | 2         |
| Acute on chronic Xanthogranulomatous cholecystitis                                                       | 4    | 2    | 25-54        | 6         |
| Chronic active cholecystitis and focal cholesterolosis                                                   | 1    | 4    | >65          | 5         |
| Xanthogranulomatous cholecystitis                                                                        | 3    | 5    | 35-54        | 8         |
| Acute hemorrhagic cholecystitis                                                                          | 1    | 0    | >65          | 1         |
| Extensively autolysed tissue                                                                             | 0    | 1    | 35-54        | 1         |
| Acute cholecystitis with perforation                                                                     | 1    | 0    | >65          | 1         |
| Acute cholecystitis                                                                                      | 0    | 1    | >65          | 1         |
| Chronic cholecystitis with focal intestinal metaplasia and low-grade dysplasia                           | 2    | 4    | 35-64        | 6         |
| Chronic active cholecystitis with focal intestinal metaplasia                                            | 0    | 1    | 55-64        | 1         |
| Moderately differentiated infiltrating adenocarcinoma                                                    | 0    | 7    | >65          | 7         |
| Moderately to poorly differentiated infiltrating adenocarcinoma                                          | 0    | 1    | 55-64        | 1         |

| Table 4. Premalignant and malignant epithelial lesions |             |           |              |            |
|--------------------------------------------------------|-------------|-----------|--------------|------------|
| Diagnosis                                              | Cases n (%) | geRange   | Female n (%) | Male n (%) |
| Intestinal metaplasia                                  | 50 (4.82%)  | 35–64 yrs | 39           | 11         |
| Dysplasia                                              | 6 (0.57%)   | 35–64 yrs | 5            | 1          |
| Adenocarcinoma                                         | 8 (0.77%)   | >63 yrs   | 8            | 0          |

The most common finding was chronic cholecystitis (421 cases, 40.6%), followed by acute cholecystitis (208 cases, 20.1%). These two entities collectively accounted for 629 cases (60.7%) of all specimens examined. Intestinal metaplasia within chronic cholecystitis was identified in 50 cases (4.82%). Dysplasia was detected in 6 cases (0.57%), of which 4 were low-grade dysplasia and 2 were high-grade dysplasia. Invasive gallbladder adenocarcinoma was identified in 8 specimens (0.77%), with 6 cases being moderately differentiated and 2 poorly differentiated; 6 of 8 cases showed T2 stage disease. Mean age for chronic cholecystitis was 52.3 years (range: 18–85) and for acute cholecystitis 55.1 years (range: 22–88).

## Discussion:

Our analysis revealed that the highest number of instances occurred within the middle adult age bracket (35–64 years), indicating a significant prevalence of gallbladder disorders in this demographic. Our findings are consistent with international research conducted in various regions, including China<sup>9</sup> and Jammu<sup>10</sup>, as evidenced by similar trends in age distribution and disease prevalence.

Our study found a significant predominance of gallbladder disorders in women, with 757 cases compared to 279 in men, corresponding to a female-to-male ratio of approximately 2.7:1. This trend is consistent with findings from other studies, such as those conducted in northeast India<sup>11</sup> and by Virginian doctors, where 66% of cases were female and 34% male<sup>12</sup>, as well as in Africa (Tunis), which reported a more balanced gender distribution with a sex ratio of 1:2.78.<sup>13</sup>

Regarding histological types, our study highlighted chronic cholecystitis as the predominant type, accounting for 40.6% of cases, followed by acute-on-chronic cholecystitis at 20.1%. Similar findings were reported in studies from Jammu and by Limaïem F et al., with chronic cholecystitis rates of 50% and 67%, respectively.<sup>14</sup> Benkhadoura M et al. observed a slightly higher prevalence of chronic cholecystitis at 70.47%<sup>15</sup>, while a study from Africa mentioned a higher rate of 88.8%. Cholesterosis was frequently

encountered as an associated histopathological finding, most commonly in conjunction with chronic cholecystitis. Previous studies have similarly reported cholesterosis as a common incidental finding, with one series identifying cholesterosis in 62 cases (11%), all associated with chronic cholecystitis.<sup>16</sup> In our study, cholesterosis was predominantly observed alongside chronic cholecystitis and chronic cholecystitis with focal intestinal metaplasia, supporting the concept that cholesterosis commonly coexists with chronic inflammatory gallbladder disease. Acute inflammatory pathology was identified in 208 cases (20.1%), comparable to previous series reporting acute inflammatory changes in 4–30% of cholecystectomy specimens.<sup>17</sup> We documented 24 cases (2.3%) of acute necrotizing/gangrenous cholecystitis, primarily observed in the elderly age group, with a notable slight male predominance (1.18:1). A study from Iran indicated a higher prevalence of 19 cases (37.25%)<sup>18</sup>, with a considerably greater male predilection at approximately 5:1. A study conducted in Korea documented a prevalence of 29.3%<sup>19</sup>. The research from Australia comprised 106 patients (37%) diagnosed with gangrenous cholecystitis<sup>20</sup>, showing similar male predilection and elderly age distribution.

Hyalizing cholecystitis is encountered in about 1% of cholecystectomies, with cancer often detected in cases with minimal or no calcification. Xanthogranulomatous cholecystitis (XGC) was observed in 8 cases (0.8%) in our study. Elevated serum CA19-9 levels have been observed in XGC without identifiable carcinoma, an important clinical consideration. XGC rates were slightly higher in our study compared to 0.30% reported by Limaïem F et al.<sup>14</sup> and 0.10% by Benkhadoura M et al.<sup>15</sup>

Gallbladder cancers contribute to 1.2% of all cancer incidences, typically occurring after the age of 60, ranking as the 20th most common tumor and the 17th deadliest cancer. We identified 8 cases of adenocarcinoma; 6 exhibited T2 stage while only one case was limited to the muscularis propria (T1). Silecchia et al. identified six incidental gallbladder carcinomas in their study.<sup>21</sup> Another study reported 24 cases of incidental gallbladder carcinoma in 4,317 cholecystectomy specimens over 8 years.<sup>22</sup> Comparable series from Siddiqui et al.<sup>23</sup>, Shrestha et al.<sup>24</sup>, Haq et al.<sup>25</sup>, and Ghimire et al.<sup>26</sup> all reported T2 stage predominance among incidentally detected carcinomas, consistent with our findings. Similar patterns of delayed diagnosis and incidental detection of malignancy in routine histopathology have been reported in studies of other organ systems, including

oral cavity and soft tissue tumors. These observations collectively emphasize the critical role of routine histopathological examination in detecting clinically unsuspected malignancies across different surgical specimens.<sup>27,28</sup>

### Study Limitations:

This was a single-center retrospective study conducted at a tertiary care referral hospital; therefore, referral bias may have influenced the spectrum of lesions observed. Only patients undergoing cholecystectomy were included, introducing ascertainment bias and limiting representation of asymptomatic gallbladder disease in the general population. Additionally, the retrospective design limited availability of detailed clinical and follow-up data.

### Conclusion:

This study demonstrates a broad histopathological spectrum in routine cholecystectomy specimens, with chronic and acute cholecystitis representing the majority of cases. Incidental premalignant and malignant lesions were identified in a small but clinically relevant proportion of specimens. These findings support the continued value of routine histopathological evaluation in detecting unsuspected significant pathology.

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