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Mucosal Dysplasia of Gallbladder: Isolated and Adjacent Lesions to Carcinoma

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Isolated dysplasia was found in 46 (54 lesions) of 2,027 gallbladders, which were histologically studied by serial step sectioning. Average age of patients was 63.8 with a male-to-female ratio of 1 to 3. These lesions, excluding polypoid lesions of adenoma, were located most frequently in the fundus and body, showing flat granular or irregular appearance. As to the size, 68.6% were smaller than 1 cm in diameter (89% were smaller than 2 cm). Most mild dysplasias were found in small lesions less than 0.5 cm in diameter and moderate ones in lesions of less than 1.5 cm, though severe dysplasias were found in various sizes. Gallstones were found in 86.9% of the cases with dysplasia, and intestinal metaplasia and pseudopyloric gland metaplasia were found in 80.4% and 100%. Dysplasia adjacent to carcinoma was found in 46 of 110 cases (41.8%), and this change was frequently found in lesions at the early stage and well-differentiated carcinoma. It seems that mucosal damage caused by a stone and subsequent inflammation may be important in the histogenesis of intestinal metaplasia and dysplasia.

Key words: Gallbladder — Intestinal metaplasia — Dysplasia — Carcinoma

Mucosal dysplasia of the gallbladder can occur after surgical resection, as determined by histological examination, and reported incidences range from 0.4%[1] to 33.8%.[2] Such a large discrepancy may be mainly explained by differences in the definitions of dysplasia, because the lesions of dysplasia occasionally show similar appearance to regenerative atypy and/or carcinoma *in situ*.

In the present paper, mucosal dysplasia of the gallbladder was investigated by examination of resected specimens.

MATERIALS AND METHODS

The materials were 2,027 gallbladders resected following the diagnosis of cholecystitis, stone or polyp between 1981 and 1987 in Mie University and related hospitals. The male-to-female ratio was 1 to 1.5 with an average age of 61.4. The resected gallbladders were opened along the liver-bed side and fixed in 10% formalin on the board. For histological examination, step sections, 5 mm apart, were taken and stained by the hematoxylin and eosin (HE), periodic acid-Schiff (PAS), PAS-alcian blue (PAS-AB), and high iron-diamine (HID-AB) methods. The peroxidase-antiperoxidase (PAP) technique was applied for demonstrating carcinoembryonic antigen (CEA) on formalin-fixed paraffin-embedded sections. Antiserum to CEA was purchased from Dakopatts A/C (Denmark).

Forty-six cases (54 lesions) with isolated dysplasia were found among the 2,027 cases described above. The dysplasia did not form a remarkably elevated lesion, but showed mild irregular and granular appearance. Mild dysplasias showed a crowded appearance, with stratification and hyperchromatism. The nuclei were elongated or ovoid in shape with small nuclei and dense nucleoplasm. Severe dysplasia had nuclei which were stratified and extended into the luminal halves of the cells. The N/C ratio was increased and the nuclei, which had central or eccentric prominant nucleoli, were more rounded with a thick nuclear membrane showing indentation and irregular clearing of chromatin (Figs. 1–3).

On the other hand, 110 cases of gallbladder carcinoma resected between 1978 and 1987 were also investigated by the same methods. Dysplasia adjacent to carcinoma tissue was found in 46 cases, 41.8%.

For these cases with isolated or adjacent dysplasia, age distribution, location, size, incidence of intestinal metaplasia, and incidence of pseudopyloric gland metaplasia were investigated. Mucosal change with goblet cells was considered to be intestinal metaplasia (incomplete) in the present investigation, because of the rarity of complete metaplasia with Paneth' cells and enterochromaffin cells. Similar glands to pyloric gland of gastric mucosa were considered to be pseudopyloric gland metaplasia. Lesions of carcinoma were classified from the gross findings into papillary, nodular and flat (granular in early, and ulcerative and/or infiltrative in advanced stages), and they were divided into early cases (cancer invasion within the muscle layer) and advanced ones (cancer invasion beyond muscle layer). Histological types were classified into well, moderately, and poorly differentiated.

RESULTS

Isolated dysplasias were frequently found in the 6th decade (32.6%) and 8th decade (26.1%) with an average age of 63.8. The male-to-female ratio was 1 to 3. The

Fig. 1. Intestinal metaplasia with goblet cells. The nuclei are elongated and confined to the basal halves of the cells. HE staining, ×320; inset, 60.

Fig. 2. Moderate dysplasia with irregular arrangement of nuclei and basophilic cytoplasm. HE staining, ×60.

frequency of location was 10.8% in the neck, 26.1% in the body, 50% in the fundus, and 13.1% in other areas (Table I). As to the size, 51.9% of 54 lesions were smaller than 0.5 cm in diameter, 16.7% from 0.5 to 1.0 cm, 20.4% from 1.0 to 1.9 cm, 3.7% from 2.0 to 2.9 cm, 7.4% from 3.0 to 3.9 cm, and 1.9% larger than 4.0 cm. Gross findings usually showed granular and irregular appearance with loss of the normal reticular pattern. As to the relation of histological atypia of dysplasia to size, lesions exhibiting mild dysplasia amounted to 50% and they were usually smaller than 0.5 cm in size (77%). Lesions exhibiting moderate dysplasia were found in 40%, and most of them were smaller than 1.5 cm in diameter. Severe dysplasias of various sizes which were

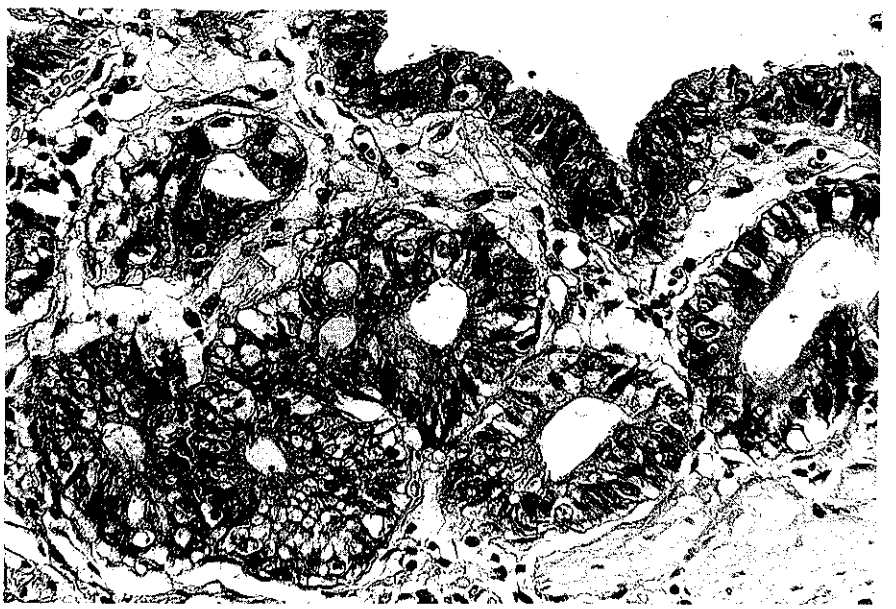

Fig. 3. Severe dysplasia with irregular rounded nuclei extending into the luminal halves of the cells and irregular clearing of chromatin. HE staining, ×280.

Table I. Location of Isolated Dysplasia and Carcinoma with Adjacent Dysplasia

Location	Number and frequency (%) of cases with isolated dysplasia	Number and frequency (%) of cancer cases with adjacent dysplasia
Neck	5 (10.8)	7 (15.2)
Body	12 (26.1)	14 (30.4)
Fundus	23 (50.0)	20 (43.5)
Different areas	6 (13.1)	5 (10.9)
Total	46	46

occasionally difficult to differentiate from carcinoma *in situ*, were found in 9.3% (Table II).

Surrounding the isolated dysplasia, intestinal metaplasia and pseudopyloric gland metaplasia were found in 80.4% and 100% of cases. The frequencies of intestinal metaplasia and pseudopyloric gland metaplasia were 20% and 73.3% in the cases without both dysplasia and stone, and 42% and 89.2% in the cases with stone. On the other hand, the frequencies of these metaplastic changes were 68% and 84% in the cases of carcinoma without stone, and 74.4% and 90.7% in those with stone. The frequency of combination with stone in the cases of dysplasia was 86.9%, and was higher than that of 44% in the cases of carcinoma.

Table II. Size and Grade of Atypia of Isolated Dysplasia

Grade of atypia	-0.5	0.5–0.9	1.0–1.4	1.5–1.9	2.0–2.9	3.0–	Number and frequency (%) of cases
Mild	21	3	1	1		1	27 (50.0)
Moderate	6	4	6	2	1	3	22 (40.7)
Severe	1	1		1	1	1	5 (9.3)
Number and frequency (%) of cases	28 (51.8)	8 (14.8)	7 (13.0)	4 (7.4)	2 (3.7)	5 (9.5)	54

Dysplasias adjacent to gallbladder carcinoma with frequent surrounding intestinal metaplasia (89.1%) and stone (61.1%) were found in 46 of 110 cases, 41.8% (average age of 65.1), and they were more frequently found in lesions of the early stage, of well-differentiated type, and of various sizes (Tables III–V). Remote isolated dysplasia was found in 3 of 110 cases, 2.7%. From the histochemical examination, goblet cells, which usually existed in the intestinal mucosa containing blue-stained sialomucin demonstrated by HID-AB stain, were found at the rate of 26% in the lesions of dysplasia and 35% in those of carcinoma (Fig. 4). Although CEA stained by the PAP method was sometimes slightly positive (approximately 40% of cases) in the apical surface of non-metaplastic epithelium and intestinal metaplasia (negative in pseudopyloric gland metaplasia), it was positive in 80% of lesions of dysplasia (stained mainly in the apical surface) and in 98% of lesions of carcinoma (stained usually in the cytoplasm) (Fig. 5).

Table III. Gross Findings of Carcinoma with Adjacent Dysplasia

	Number of cancer cases	Number and frequency (%) of carcinoma with adjacent dysplasia
Early		
papillary	8	4 (50)
nodular	3	1 (33.3)
flat (granular)	14	9 (64.3)
Advanced		
papillary	13	8 (61.5)
nodular	31	8 (25.8)
infiltrative	41	16 (39.0)
Total	110	46 (41.8)

Table IV. Histological Findings of Carcinoma with Adjacent Dysplasia

Histological findings	Number of cancer cases	Number and frequency (%) of carcinoma with adjacent dysplasia
Well-dif.	65	32 (49.2)
Moderately-dif	25	11 (44.0)
Poorly-dif.	13	2 (15.4)
Adenosquamous	7	1 (14.2)
Total	110	46 (41.8)

Table V. Size of Carcinoma with Adjacent Dysplasia

Size (cm)	Number of cancer cases	Number and frequency (%) of carcinoma with adjacent dysplasia
–0.5	6	2 (33.3)
0.5–0.9	4	2 (50.0)
1.0–1.9	10	6 (60.0)
2.0–2.9	12	6 (50.0)
3.0–3.9	19	6 (31.6)
4.0–4.9	18	9 (50.0)
5.0–	41	15 (36.6)
Total	110	46 (41.8)

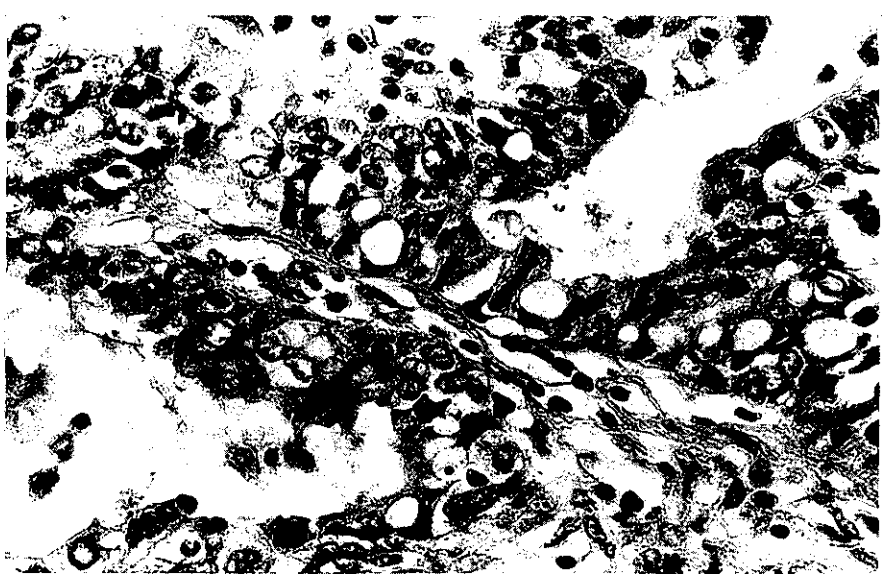

Fig. 4. Well-differentiated adenocarcinoma with goblet cells. HE staining, ×320.

Fig. 5. Positive staining is seen in the apical surface and cytoplasm in the well-differentiated adenocarcinoma. Immunohistochemical staining of CEA by the PAP method. ×120.

DISCUSSION

Albores-Saavedra *et al.* observed 83% of epithelial hyperplasia, 13.5% of atypical hyperplasia and 35% of carcinoma *in situ* from the histological examination of 200 resected gallbladders.[3] Latio found dysplasia at the rate of 33.8% (severe 1.4%, moderate 8.5%, and mild 23.9%) and intestinal metaplasia at the rate of 83.3% in 71 resected gallbladders, and this author pointed out the importance of the sequence of intestinal metaplasia to dysplasia.[1] On the other hand, Dowling and Kelly reported only one case of isolated dysplasia from 277 resected gallbladders (0.4%).[2] In Japan, the frequency of dysplasia was reported to be 13.1% of 247 resected cases by Majima[4] and 7.6% of 500 resected cases by Kojima *et al.*[5] In the present investigation, the incidence of isolated dysplasia was 2.2%.

Albores-Saavedra *et al.* described epithelial hyperplasia as the earliest precursor lesion of carcinoma and stated that some cases of epithelial hyperplasia will progress to atypical hyperplasia or dysplasia.[3] However, Dowling found no evidence of epithelial hyperplasia in his own series, and this author would not regard the lesions, which were called mild dysplasia by Latio,[1] as being dysplasia.[2] Black and associates stated that isolated atypical hyperplasia is distinctly uncommon in the gallbladder.[6]

Such marked discrepancies in the reported incidences may be mostly explicable in terms of differences in the populations studied, in the methods of examination, i.e. step section or single section, and in the definitions of the precursor lesions used in the studies. For example, it is difficult to distinguish regenerative atypia and well-differentiated adenocarcinoma *in situ* from dysplasia.

In the present series, gallstone was found in 86.9%, intestinal metaplasia in 80.4% and pseudopyloric gland metaplasia in 100% of the cases with dysplasia, respectively. Probably, the prolonged mucosal damage by a stone causes desquamation and regeneration of mucosal epithelium with subsequent metaplastic change, especially intestinal metaplasia. Whether gallstones cause or result from dysplasia is a moot point which deserves study; however, such damage may cause immature atypical epithelial proliferation after a long period.[7,8] Goblet cells containing sialomucin, which were usually found in the intestinal mucosa, were not infrequently found in the tissues of dysplasia and carcinoma. Although CEA stained by the PAP method was negative in pseudopyloric glands and slightly positive in the apical surface of non-metaplastic epithelium and intestinal metaplasia in approximately 40% of cases, this substance was strongly positive mainly in the apical surface of cells constituting dysplasia and in cytoplasm of cells constituting carcinoma. From these findings, it was considered that the sequence of intestinal metaplasia-dysplasia-carcinoma may be important especially in cases with gallstone.[9,10]

As to dysplasia adjacent to carcinoma, Dowling reported the incidence of 66.7%[2] and suggested the importance of intestinal metaplasia for the basic change, as had Albores-Saavedra *et al.*[3] and Latio.[1] The possibilities are that adjacent dysplasia 1) arose as very well-differentiated carcinoma, 2) arose synchronously, 3) arose secondarily, and 4) arose as a precancerous lesion. In the formation of such adjacent dysplasia, it seems likely that abnormal epithelial proliferation may easily occur in the metaplastic mucosa adjacent to carcinoma.

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