

Alterations in mucin type: an indicator for suspicion of malignant gastric transformation

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Abstract

Mucins are produced by both benign and malignant gastric epithelium. In general, mucins can be classified into neutral and acidic mucins. The latter are of 2 major types, sulphated (sulphomucins) and carboxylated (sialomucins). A retrospective study was initiated at the Department of Pathology, University Hospital, Kuala Lumpur to histochemically study the mucin profiles of cases of intestinal (IGC) and diffuse (DGC) types of gastric carcinoma in Malaysian patients to determine whether a significant change of mucin type occurs in the event of malignant transformation. 42 IGC and 37 DGC were subjected to alcian blue-periodic acid Schiff and high iron diamine-alcian blue histochemical staining. In addition, 18 cases of gastrectomies performed for benign lesions in the stomach served as normal controls. The number of cases of IGC and DGC which exhibited sulphomucin production was significantly increased ($p < 0.001$) compared to normal controls. Also, the number of cases of DGC which produced neutral mucin were significantly less ($p < 0.05$) than the control group. However, there was no significant difference between the number of IGC and DGC cases which demonstrated sialomucin production and normal controls. It appears that while not pathognomonic, a lack of neutral mucin production should alert the pathologist to the possibility of a gastric malignancy, in particular DGC. The likelihood of a malignant lesion would be further supported if there is an increased sulphomucin production.

Key words: gastric carcinoma, sulphomucin, sialomucin, neutral mucin, alcian blue-periodic acid Schiff, high iron diamine-alcian blue

INTRODUCTION

Although most major histopathology laboratories in the country look towards immunohistochemistry as the main aid to histological diagnosis, time-proven histochemical techniques should not be underrated as valuable diagnostic adjuncts. Mucin histochemistry is well-established and extensive work has been done in the last three decades or so to study the chemical types of mucin produced in the normal gastrointestinal tract as well as when malignant transformation occurs. Normal gastric epithelium is mucin producing, the predominant mucin being of the neutral variety.¹ In contrast, acid mucins, primarily sialomucins and sulphomucins, are produced only in small amounts in the foveola and neck cells of the fundus, foveola of the antrum and cardiac glands of the stomach.² Several workers in the West have recorded an increased production of sulphomucin when malignancy supervenes.^{2,4,6} There has however been no documented evidence of such in gastric carcinoma among Malaysian patients. With the advent of the widespread use of fiberoptic

endoscopes and endoscopic biopsy for the diagnosis of carcinoma of the stomach, it has become necessary that accurate histological diagnoses be made from tiny pieces of biopsy material. Like all tiny pieces of biopsy tissue, this often poses problems for the pathologist. As with many other tumours, gastric carcinoma also suffers from a lack of a reliable histological marker capable of assisting in the diagnosis of difficult biopsies. Hence, we initiated a retrospective study to evaluate whether mucin type significantly alters in the presence of gastric carcinoma in our local patients and the possibility of utilising mucin histochemistry as an inexpensive aid in the diagnosis of problematical cases.

MATERIALS AND METHODS

All cases of primary gastric adenocarcinoma diagnosed at the Department of Pathology, University Hospital, Kuala Lumpur with gastrectomy performed between January 1986 and December 1991 were retrieved. All the cases were reviewed histologically and classified

according to Lauren's classification system⁷ into intestinal (IGC) and diffuse (DGC) types of gastric carcinoma. The cases which produced large amounts of mucin were not separated into a different category but were classified as IGC or DGC according to the criteria of Lauren. All gastrectomies performed for various benign lesions in 1991 served as normal controls. These were also retrieved and reviewed histologically. Cases with intestinal metaplasia were excluded as controls. Only histologically re-confirmed cases of gastric carcinoma and normal controls were admitted into the study. Although note was made during histological review of the presence or absence of associated intestinal metaplasia in all cases of gastric carcinoma, tissue blocks which showed intestinal metaplasia in addition to the carcinoma were omitted from histochemical study. One representative paraffin block of each case of carcinoma without features of intestinal metaplasia in the block and one non-lesional block of normal gastric mucosa of the normal control cases were therefore selected for histochemical staining. Two serial sections were cut from the formalin-fixed, paraffin-embedded tissue blocks. One section was stained with alcian blue, pH 2.5-periodic acid Schiff (AB-PAS)⁸ and the other with high iron diamine-alcian blue, pH 2.5 (HID-AB)⁹ using routine established protocols. Positive controls for both the histochemical techniques were run with each batch of tests. Sections of stomach with intestinal metaplasia served as controls for the AB-PAS technique. Sections of normal colonic mucosa served as controls for sulphomucin and normal small intestinal mucosa for sialomucin in the HID-AB technique.

The AB-PAS and HID-AB stained sections were histologically examined. Acid mucins stained blue and neutral mucin magenta with AB-PAS. Presence of both types of mucin in the same cell resulted in an intermediate colour. Sulphomucins stained black while sialomucins remained blue when stained with HID-AB. Only cases with intracellular mucin production were interpreted as positive. The results were analysed and the probability (p) value calculated using the chi-square test.

RESULTS

During the 6-year period of the study, 79 cases of primary gastric adenocarcinoma had undergone gastrectomy. Of these, 42 were IGC and 37 DGC. In 1991, 18 patients had gastrectomy performed for a benign lesion.

TABLE 1: Age and sex distribution of cases of intestinal and diffuse types of gastric carcinoma

	Histological type (Lauren)	
	Intestinal-type	Diffuse-type
Age range (yrs)	37 – 98	27 – 89
Mean age (yrs)	63.5	59.1
Male:Female ratio	2:1	1.2:1

The age and sex distribution of the cases of gastric carcinoma is shown in Table 1. The mean age of patients with IGC was 4.4 years older than those with DGC. 28 (66.7%) cases of IGC were males while 14 (33.3%) were females; the male:female ratio was 2:1. In the DGC group, 20 (54.1%) cases were males while 17 (45.9%) were females and the male:female ratio was 1.2:1.

The chemical types of mucin produced in normal control stomachs, IGC and DGC are illustrated in Table 2. Table 3 compares the number of cases of IGC and DGC with respect to the chemical types of mucin produced to the normal controls. The number of cases of IGC with sulphomucin production were significantly increased compared with the normal stomach (p<0.001). In the DGC group, sulphomucin production was also seen in a significantly increased number of cases compared to the normal (p<0.001). Figure 1 illustrates a case of IGC and Figure 2 a case of DGC with sulphomucin production. In addition, the number of cases of DGC which demonstrated neutral mucin

TABLE 2: Chemical type of mucin produced in cases of normal control stomachs (n=18) and intestinal (IGC) (n=42) and diffuse (DGC) (n=37) types of gastric carcinoma

	Neutral mucin	Sialomucin	Sulphomucin
Normal control stomachs			
No.positive (%)	18 (100)	8 (44)	2 (11)
No.negative (%)	0 (0)	10 (56)	16 (89)
IGC			
No.positive (%)	36 (86)	29 (69)	29 (69)
No.negative (%)	6 (14)	13 (31)	13 (31)
DGC			
No.positive (%)	29 (78)	13 (35)	28 (76)
No.negative (%)	8 (22)	24 (65)	9 (24)

TABLE 3: Comparison of the chemical types of mucin production by the intestinal (IGC) (n=42) and diffuse (DGC) types of gastric carcinoma to normal control stomachs (n=18)

	Neutral mucin	Sialomucin	Sulphomucin
IGC (%)	36 (86)	29 (69)	29 (69)
Normal control stomachs (%)	18 (100)	8 (44)	2 (11)
p value	> 0.05	> 0.05	< 0.001
DGC (%)	29 (78)	13 (35)	28 (76)
Normal control stomachs (%)	18 (100)	8 (44)	2 (11)
p value	< 0.05	> 0.05	< 0.001

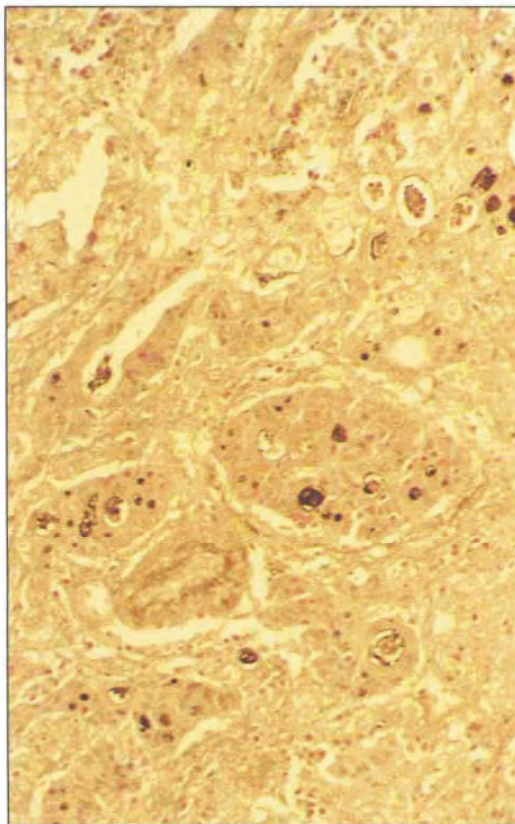


FIG 1: Intracytoplasmic sulphomucin produced by the malignant epithelial cells surrounding glandular structures in a case of the intestinal type of gastric carcinoma stained black with HID-AB (X200).

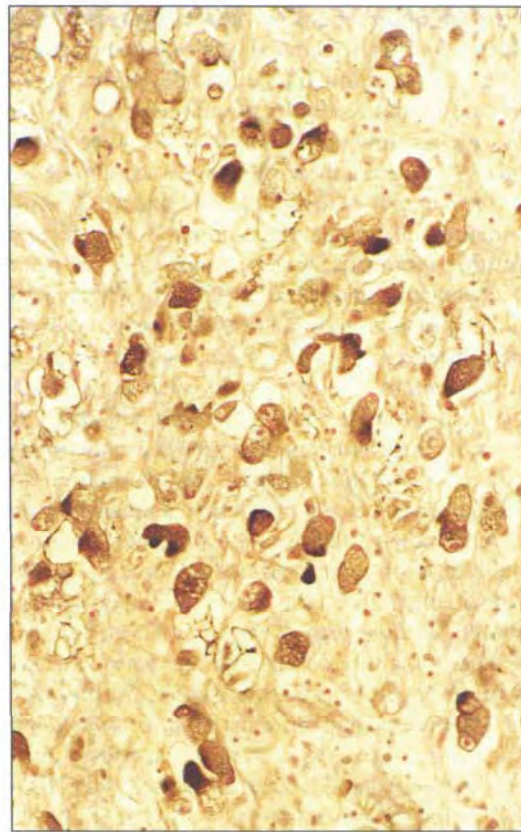


FIG 2: A case of Lauren's diffuse type of gastric carcinoma with sulphomucin produced in almost all the carcinomatous cells which appear as black intracytoplasmic staining (HID-AB X300).

production were significantly decreased compared to the normal controls ($p < 0.05$). No statistical difference was observed between the number of cases of IGC and DGC with sialomucin production and the normal controls.

DISCUSSION

Gastric carcinoma remains an important problem among Malaysians. On the whole, the results of this study show that the demographic pattern of gastric carcinoma is similar to what is generally known. The male predominance observed in IGC, the lack of sex predilection in DGC and the occurrence of IGC in older patients compared with DGC does not differ from what has also been previously noted by Lauren in a study in a large Finnish population? Also similar to what has been previously noted by other workers,¹⁻³ the predominant type of mucin produced by normal control gastric mucosa was neutral mucin.

However, the most important and interesting finding of this study is the re-confirmation of

previous observations by various workers^{2,4-6} that the predominant chemical type of mucin changes when gastric epithelial cells undergo malignant transformation. Unlike normal gastric mucosa, in which neutral mucin was the most widespread mucin produced, there was a significant increase in the number of cases of IGC and DGC with sulphomucin production compared with normal control stomachs. Furthermore, a significant decrease in the number of DGC cases with neutral mucin production compared with normal controls was observed. It can be surmised therefore that sulphomucin production is increased when malignant transformation to either IGC or DGC ensues. In contrast, neutral mucin production is decreased when neoplastic transformation to DGC occurs. In practical terms, it means that on a tiny biopsy, the loss of magenta staining, indicative of a loss of neutral mucin, of the constituent cells on the AB-PAS-stained section should alert the pathologist to look carefully for a carcinomatous component, in particular the diffuse variety of gastric carcinoma. The presence of an increased and prominent intracellular black staining, representative of sulphomucin production when the tissue is stained with HID-AB, while not confirmatory, is also suggestive of malignant change to either IGC or DGC. Nonetheless, it has to be cautioned that 11% of normal controls also demonstrate sulphomucin production.

In conclusion, gastric carcinoma occurring in Malaysians does not appear to differ considerably from that of their Western counterparts in terms of age of onset and sexual predilection. The chemical types of mucin produced in the normal stomach and in gastric carcinoma also parallel what has been noted in Western studies. From this study, it is also apparent that the simple mucin histochemistry, interpreted judiciously, can be revived to provide useful clues to the diagnosis of problematical biopsies.

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