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Helicobacter pylori and gastric cancer: correlation with gastritis, intestinal metaplasia, and tumour histology

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Abstract

This study aimed to examine the association between *Helicobacter pylori*, histological gastritis, and intestinal metaplasia in gastric cancers of different histological types. A total of 169 gastrectomy specimens received in one pathology department were studied. Altogether 156 were adenocarcinomas (intestinal type 87, diffuse type 50, mixed type 19). Gastritis occurred in 137 of 163 body specimens (84%) and in 126 of 131 antral specimens (96%). Its presence was unrelated to tumour histology. Atrophic gastritis was more common in both body and antral mucosa in intestinal type compared with diffuse type carcinoma. This was also true for intestinal metaplasia of the body, but not of the antral mucosa. *H pylori* was present in 101 of 163 (62%) body specimens and 56 of 131 (43%) antral specimens. In intestinal type carcinoma, *H pylori* was found in 52/84 (62%) body specimens and in 24/70 (34%) antral specimens, while the corresponding figures for diffuse type carcinoma were 29/48 (60%) and 17/38 (45%) respectively. Tumour histology therefore had no influence on the occurrence of *H pylori*. Tumour site had no effect on the presence or absence of gastritis, atrophic changes, intestinal metaplasia, or *H pylori*. While both *H pylori* and gastritis are associated with gastric cancer, the association is unrelated to tumour histology and may not be a causal one.

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The incidence of gastric cancer varies up to 20 fold between high risk and low risk countries and between different regions within the same country. Migrants from high risk to low risk areas have an intermediate risk. In the last few decades there has been a significant fall in the incidence of gastric cancer worldwide. Such marked geographical and temporal variations in gastric cancer frequency suggest that environmental influences are important in its cause.[1-3]

To date, most investigators have emphasised the role of diet in gastric carcinogenesis.[4] Atrophic gastritis with intestinal metaplasia has been identified as a precursor lesion for the commoner intestinal form of gastric carcinoma.[5,6] In contrast, no relation was shown between gastritis and intestinal metaplasia on one hand and the diffuse form of gastric carcinoma on the other.[7] Recently, *Helicobacter pylori* was shown to be closely associated, clinically and epidemiologically, with non-autoimmune gastritis and a causal relation has been postulated.[8,9] Three case-control studies established an association between *H pylori* infection and the subsequent development of gastric cancer.[10-12] The possibility that *H pylori* is involved in gastric carcinogenesis has therefore been raised.[6] This study examines the relations between *H pylori*, histological gastritis, and intestinal metaplasia in gastric cancers of different histological types.

TABLE I *Demographic characteristics*

	Adenocarcinoma type				
	Intestinal	*Diffuse*	*Mixed*	*Others*	*All*
No	87	50	19	13	169
M:F	63:24*	21:29*	15:4	8:5	107:62
Race:					
(Chinese *v* others)	79:8	45:5	18:1	13:0	155:14
Site:					
Body	48	26	8	8	90
Antrum	38	21	10	5	74
Other	1	3	1	0	5
Age:					
Mean (SD) (years)	62:5† (11·1)	57:6† (16.1)	65:6 (11·9)	52:1 (19·6)	60:6 (14·0)

Intestinal *v* diffuse disease: *p<0·001; †p=0·058.

Methods

A retrospective study of all gastrectomy specimens of gastric tumours received by the Pathology Department at the National University Hospital from January 1986 to September 1990 was performed. The specimens were fixed in 10% buffered formalin. Multiple blocks were taken from the tumour, antrum, and body (proximal resection margins in most cases). Paraffin embedded histological sections were cut at 4 μm thickness and stained with haematoxylin and eosin. Periodic acid-Schiff reaction with and without diastase digestion, mucicarmine, other special stains, and immunohistochemical techniques were performed whenever necessary. All the sections were reviewed independently by two of the authors (AW and TM) for histological typing. In instances when there was inter-observer variation, agreement was reached after review and discussion of the cases. The sections were also reviewed for the occurrence of gastritis and intestinal metaplasia. All tissue blocks of antral and body mucosa and representative blocks of tumour areas were recut and stained by the Giemsa method for the identification of *H pylori*. The sections representing antral and body mucosa were taken as far from the tumour site as possible.

HISTOLOGICAL CLASSIFICATION OF GASTRIC CARCINOMA

Gastric adenocarcinoma was typed according to a modified Lauren[13] classification into:

(i) Intestinal type, characterised by cohesive cell clusters showing distinct glandular elements with well defined lumina lined by well polarised epithelial cells. Papillary structures, solid components, brush borders, and pools of extracellular mucin may be present.

(ii) Diffuse type, showing dissociated malignant cells infiltrating surrounding tissue as single or small clusters of cells. Glandular lumina are lacking. Intracellular or extracellular mucin production is variable.

(iii) Mixed type, encompassing the features of both intestinal and diffuse subtypes.

HISTOLOGICAL ASSESSMENT

Histological gastritis was defined by a modification of the system used by McNulty *et al*,[14] which has previously been described in detail and shown to be highly reproducible.[15 16] In brief, mononuclear and polymorphonuclear cellular infiltration were each graded from zero to four. If the acute inflammatory score was one or more, or the chronic inflammatory score was three or more, gastritis was considered to be present. Gastritis was said to be acute if the polymorphonuclear cellular score was one or more with a mononuclear cellular score of under three. Chronic gastritis was diagnosed if the mononuclear cellular score was three or above while the acute inflammatory score was zero. Acute on chronic inflammation was diagnosed when the acute and chronic inflammatory scores were greater than zero and two respectively. The occurrence of atrophic gastritis, intestinal metaplasia, and *H pylori* was recorded separately.

STATISTICAL ANALYSIS

Categorical data were compared by the χ^2 test but the McNemar's test was used for comparing the frequency of atrophic gastritis, intestinal metaplasia, and *H pylori* in the body *v* antral mucosa. Numerical data were compared by Student's *t* test. Probability values of $<0{\cdot}05$ were considered significant.

Results

The numbers of tissue blocks studied were: (mean (SD)) 3·7 (1·7) for tumour tissue, 2·8 (1·8) for body mucosa, and 1·1 (1·0) for antral mucosa. Of 169 specimens available for study, 156 (92%) were carcinomas (Table I). Intestinal type carcinoma was the largest group, followed by the diffuse type, while 12% showed features of both. Patients with intestinal type were more likely to be male compared with patients with the diffuse type of carcinoma. They also tended to be older, although the difference was not statistically significant. For all types of gastric cancer, the proportion of Chinese exceeded the 75% found in the general population of Singapore.

Some specimens contained only body or antral mucosa. Gastritis was present in 84% of body mucosa and 96% of antral mucosa: its frequency and type were unrelated to tumour histology (Table II). Lymphoid follicles were identifiable in most specimens irrespective of tumour histology or type of mucosa. For both body and antral mucosa, atrophic gastritis was commoner in intestinal type carcinoma than in the diffuse type (Table II). Intestinal metaplasia was also more frequently seen in the intestinal type cancer but the differences were not statistically significant. The differences between the diffuse and intestinal forms of gastric carcinoma in the frequency of atrophic gastritis remained even after correction for the older ages of patients with intestinal type carcinoma. No *H pylori* was identified over the tumour areas. There was no obvious correlation between tumour histology and the occurrence of *H pylori* in either the body or antrum. *H pylori* was identified in intestinal type carcinoma in 52/84 (62%) specimens of body mucosa and 24/70 (34%) specimens of antral mucosa, while the corresponding figures for diffuse type carcinoma were 29/48 (60%) and 17/38 (45%) respectively (Table II).

Overall, *H pylori* occurred in body or antral mucosa, or both, in 64 of 87 patients (74%) with intestinal type carcinoma compared with 39 of 50 (78%) with diffuse type carcinoma. There was no difference between the frequency of *H pylori* in intestinal type carcinoma and that in diffuse type cancer, even after allowance was made for the presence or absence of intestinal metaplasia. Among subjects with intestinal metaplasia of either body or antral mucosa, 53 of 70 (76%) with intestinal type carcinoma were positive for *H pylori* compared with 31/34 (91%) with diffuse type carcinoma ($p=0{\cdot}11$). In the case of subjects without intestinal metaplasia of either body or antral mucosa, 11 of 17 (65%) with intestinal type carcinoma were positive for *H pylori* compared with 8/16 (50%) with diffuse type carcinoma ($p=0{\cdot}6$).

Considering only the 125 patients for whom both body and antral mucosal specimens were available for study, the frequency of gastritis, intestinal metaplasia, and positivity for *H pylori* were 121 (97%), 88 (70%), and 86 (69%) respectively. Both atrophic gastrtis and intestinal metaplasia were more common in antral than body

TABLE II *Frequency (%) of gastritis and* Helicobacter pylori *infection in different histological subsets of gastric cancer*

	Adenocarcinoma				
	Intestinal	*Diffuse*	*Mixed*	*Others*	*All*
Body mucosa:					
No	84	48	18	13	163
Gastritis:					
All	71 (85)	39 (81)	16 (89)	11 (85)	137 (84)
Chronic	7 (9)	4 (8)	0	0	11 (7)
Acute	17 (20)	13 (27)	4 (22)	4 (31)	38 (23)
Acute on chronic	47 (56)	22 (46)	12 (67)	7 (54)	88 (54)
Atrophic gastritis	24 (29)*	5 (10)*	4 (22)	2 (15)	35 (21)
Intestinal metaplasia	19 (23)	4 (8)	2 (11)	1 (8)	26 (16)
Lymphoid follicles	71 (85)	40 (83)	15 (83)	10 (77)	136 (83)
Helicobacter pylori	52 (62)	29 (60)	13 (72)	7 (54)	101 (62)
Antral Mucosa:					
No	70	38	15	8	131
Gastritis:					
All	69 (99)	35 (92)	14 (93)	8 (100)	126 (96)
Chronic	12 (17)	10 (26)	1 (6)	0	23 (18)
Acute	2 (3)	0	0	1 (13)	3 (2)
Acute on chronic	55 (79)	25 (66)	13 (87)	7 (87)	100 (76)
Atrophic gastritis	59 (84)†	23 (61)†	13 (87)	6 (75)	101 (77)
Intestinal metaplasia	52 (74)	21 (55)	13 (87)	6 (75)	92 (70)
Lymphoid follicles	70 (100)	37 (97)	14 (93)	8 (100)	129 (98)
Helicobacter pylori	24 (34)	17 (45)	9 (60)	6 (75)	56 (43)

125 patients had both body and antral mucosa studied. In the others only body or antral mucosa was available. Intestinal *v* diffuse disease type: *, †$p<0{\cdot}05$.

TABLE III *Correlation of* Helicobacter pylori *with gastritis*

		Helicobacter pylori		
		Present	*Absent*	*p*
Body or antral gastritis	Present	86	35	
	Absent	0	4	<0·01
Body gastritis	Present	98	39	
	Absent	3	23	<0·001
Antral gastritis	Present	56	70	
	Absent	0	5	0·058

mucosa ($p<0·001$ in each case). In contrast, *H pylori* occurred more commonly in the body than in the antrum ($p<0·001$). The infrequency of *H pylori* in the gastric antrum was partly but not entirely accounted for by the higher frequency of intestinal metaplasia in the antrum. *H pylori* was present in 75 of 105 body specimens (71%) that did not show intestinal metaplasia compared with 17 of 38 antral specimens (45%: $p<0·01$). None of four subjects without gastritis of the body or antrum was positive for *H pylori* compared with 86 of 121 with gastritis of the body or antrum (Table III: $p<0·01$). The presence of *H pylori* in body mucosa also correlated with body gastritis but the correlation for antral mucosa just failed to reach statistical significance.

Tumour location, whether body or antral, had no influence on the occurrence or type of gastritis. The frequency of atrophic gastritis, intestinal metaplasia, identifiable lymphoid follicles, or *H pylori* was likewise similar whether the tumour occurred in the body or antrum.

Discussion

Of the two main histological types of gastric carcinoma, the less common diffuse variety is not associated with well defined precancerous lesions.[7] However, the intestinal type of gastric cancer which predominates in high risk populations is thought to originate from cancer precursor lesions, namely, chronic atrophic gastritis and intestinal metaplasia.[5] The initiation and promotion of these precancerous lesions are thought to be influenced by several factors including gastric hypoacidity, bacterial overgrowth, and diets low in antioxidants but high in irritants and mutagen precursors.

Epidemiological and clinical studies have established a close association between *H pylori* and non-autoimmune gastritis, and many believe there is a causal relationship.[8,9] Correa[6] has hypothesised that *H pylori* may be a cause of multifocal chronic atrophic gastritis, dysplasia, and hence gastric cancer. Infection with *H pylori* was found to increase the risk of subsequent development of gastric cancer.[10] In different counties within China, the frequency of gastric cancer correlated significantly with the prevalence of antibody against *H pylori*.[17] Populations at high risk of gastric cancer have high prevalence rates of *H pylori* infection, and acquisition of infection occurs at younger ages than in low risk populations.[18,19] Subjects from lower socioeconomic backgrounds have the highest rates of infection, again conforming to their increased gastric cancer risk.[20] An anecdotal report suggested a relationship between *H pylori*, gastric dysplasia, the occurrence of intestinal metaplasia at a young age, and familial gastric cancer.[21] It must be pointed out, however, that *H pylori* is associated with both the diffuse type of antral gastritis, for which there is less evidence of a relationship with gastric cancer, and the multifocal atrophic variety which is thought to lead to intestinal metaplasia and the intestinal type of gastric carcinoma.[6]

Both *H pylori* and gastritis therefore occur commonly in association with gastric carcinoma. However, if *H pylori* is indeed causally related to atrophic gastritis, intestinal metaplasia, and the intestinal type of gastric cancer we would expect a close association between the organism and the intestinal type rather than the diffuse type of gastric cancer. Although intestinal metaplasia was, as expected, more common in the intestinal type of carcinoma compared with the diffuse type, *H pylori* occurred equally frequently in the two groups. Our findings therefore argue against Correa's hypothesis that *H pylori* is a causal factor in the atrophic gastritis-intestinal metaplasia-intestinal form of gastric cancer sequence. One possibility is that the association between *H pylori* and gastric cancer is a fortuitious one, secondary to separate associations with gastritis. It is also possible that *H pylori* may be causally related to both forms of gastric cancer via an unknown mechanism.

Our results concur with those from a recent European study which found no difference in the occurrence of *H pylori* between intestinal and diffuse type gastric carcinoma.[22] These findings differ from those of an American study in which *H pylori* was found more frequently in intestinal type gastric carcinoma than in the diffuse type.[23] The reason for these discrepancies is unclear. Three recent case-control studies indicated an association between infection with *H pylori* and subsequent development of gastric cancer.[10–12] In two of these,[11,12] tumour histology was studied and no difference was found between the intestinal and the diffuse type of gastric carcinoma in their relationship with *H pylori*.

H pylori does not colonise metaplastic epithelium. This is a potential confounding factor in the present study since even if *H pylori* infection was the cause of atrophic gastritis and intestinal metaplasia initially, it might not be demonstrable subsequently in metaplastic tissue. This problem was overcome by considering patients with and without intestinal metaplasia separately.

Another possible confounding factor is that *H pylori* was only looked for in the resection specimens. It is possible that as gastritis becomes severe, *H pylori* colonisation is reduced because of hypoacidity and over-growth with other bacteria. The frequency of previous exposure to *H pylori* may therefore be underestimated. A third possible confounding factor is that the histological identification of *H pylori* in fixed resection specimens may not be optimal, although Ormand *et al* reported otherwise.[24] Both of these confounding factors are expected to affect tumours of different histological types equally and will probably not influence the conclusions of the present study.

In Singapore gastric cancer frequency varies

widely between the three main racial groups, Chinese being more susceptible than either Malays or Indians.[25 26] Because of the small numbers of specimens from Malay and Indian patients, no inter-racial comparisons can be made in the present study.

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