Comparison between resectable gastric adenocarcinomas seropositive and seronegative for *Helicobacter pylori*

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The preoperative seropositivity of *Helicobacter pylori* was determined in 128 patients who had undergone gastrectomy for primary gastric adenocarcinoma during the past 5 years. The overall seroprevalence of *H. pylori* was 64 per cent. Gastric cancers positive for *H. pylori* were associated with tumours located in the lower third of the stomach which were of localized type (Borrmann I and II) ($P < 0.05$), but not with age, sex, blood type, tumour size, invasion depth, lymph node metastases, histological type, DNA ploidy or type of surgery. The cumulative 5-year survival curves after surgical resection were significantly

better in patients who were positive for *H. pylori*. Multivariate analysis revealed that seropositivity for *H. pylori* was not an independent prognostic factor. Pathological tumour node metastasis staging was the only prognostic indicator. Better prognosis for those with *H. pylori*-seropositive gastric cancer may be attributed to the more advanced stage of *H. pylori*-seronegative gastric cancers. The potential role of *H. pylori* in gastric cancer carcinogenesis and its biological significance warrant further investigation.

Gastric cancer is common worldwide¹, and surgery remains the main treatment in about half the patients^{2–4}. The reported survival after resection varies, but Japanese studies have shown an improved survival with an overall 5-year survival rate ranging between 40 and 50 per cent after resection^{5,6}. The survival rate has not appreciably changed in western countries, where the overall 5-year survival rate varies between 21 and 25 per cent after resection^{3,4}. It is possible that biological differences exist between Japanese and western patients but there are little convincing data⁷.

The association between previous infection with *Helicobacter pylori* and subsequent development of gastric cancer has been suggested from epidemiological studies^{8–14}. Aside from the putative role in carcinogenesis, the specific biology of gastric cancer associated with *H. pylori* may be an important clinical problem.

In the present study, the serum samples taken before operation from 128 patients who underwent gastrectomy were analysed to examine any association with the bacteria. This has allowed a retrospective assessment of the clinicopathological and prognostic association which may exist between *H. pylori* and gastric cancer.

Patients and methods

Between July 1988 and March 1993, 128 patients who had undergone gastric resection for primary gastric adenocarcinoma were studied. No patient had undergone previous treatment such as chemotherapy or radiotherapy and all patients were followed clinically for a median of 3 years. The sex and age of the patient, the location of the tumour within the stomach, tumour size as measured in the surgical specimen, gross appearance (Borrmann type), and type of surgical resection were obtained from medical records. The operations were performed according to the

guidelines of the Japanese Research Society for Gastric Cancer¹⁵. Radical lymph node dissection R_{2/3} was routinely performed. When the tumour involved the proximal stomach, total gastrectomy with splenectomy and subtotal pancreatectomy was performed. The operative mortality rate was 1.6 per cent. A detailed review, including histopathological examination of available tissue slides, was performed to determine the depth of invasion on the gastric wall and the extent of metastases within regional lymph nodes. The stage was rated according to the tumour node metastasis (TNM) classification¹⁶. The histology was classified into diffuse and intestinal types in accordance with Laurén's classification¹⁷. DNA ploidy was examined by flow cytometry using fresh frozen tissue as described previously¹⁸.

Serological detection of immunoglobulin G antibodies against *Helicobacter pylori* in serum

Serum samples were stored at -70°C until examination. The presence of immunoglobulin G (IgG) antibodies against *H. pylori* was tested using a highly sensitive and specific enzyme-linked immunosorbent assay (ELISA: HEL-p test, AMRAD, Kew, Victoria, Australia). The antigen was an inactivated native antigen of *H. pylori*. Each serum sample was diluted 1:200. The secondary antibody was a sheep anti-human IgG-conjugated horseradish peroxidase. A specimen was considered positive for IgG antibodies to *H. pylori* if its optical density value was greater than or equal to the upper cut-off value which was calculated from positive control supplied by the manufacturer. The IgG assay had a sensitivity of 96 per cent and a specificity of 93 per cent and correlated well with other methods in detection of *H. pylori* infection¹⁹. All assays were performed without knowledge of clinical details.

Statistical analysis

Clinicopathological comparisons were made between patients positive and negative for *H. pylori* using the χ^2 test with Yates' correction and Student's *t* test. A *P* value of less than 0.05 was considered to be significant. Cumulative survival rates were calculated using the Kaplan–Meier method. Analysis was performed using the Biomedical statistical package (BMDP Statistical Software, Los Angeles, California, USA).

Results

An overall seropositivity for *H. pylori* was seen in 82 of 128 patients (64 per cent). The clinicopathological data of the resected tumours in both groups are summarized in Table 1. Significant correlations were found between the location of tumours and gross type (Borrmann) and *H. pylori*-positive gastric cancer was associated with tumours located in the lower third of the stomach which were of localized type (Borrmann I and II) ($P < 0.05$).

There were no differences in the extent of gastric resection, operative mortality rate and provision of postoperative chemotherapy (Table 2).

Histopathological data were compared for the two groups. There were no differences in distribution of invasion depth, lymph node metastases, distant metastases, clinical stages, histological type, DNA ploidy, duodenum invasion and section line involvement.

Fig. 1 shows the survival curves of both groups after gastric resection. Patients seropositive for *H. pylori* had a better 5-year survival rate than those determined to be seronegative, but multivariate analysis using the Cox

proportional hazards model revealed that TNM staging was the only significant prognostic indicator (hazard ratio 3.122, $P < 0.0001$).

Discussion

Since the first successful culture in April 1982, *H. pylori* has attracted increasing attention because of its potential role in gastroduodenal diseases^{20,21}. It is considered to be the main cause of non-autoimmune chronic antral gastritis and, therefore, may lead to the development of gastric cancer. This hypothesis has recently been suggested in many epidemiological studies⁸⁻¹⁴. In addition to its role in carcinogenesis, *H. pylori* might also influence the biology of this specific type of gastric carcinoma.

Although declining in incidence, gastric cancer remains the third leading cause of cancer death in Taiwan, with an annual death rate in 1990 of 15.4 and 7.2 deaths per 100 000 for men and women, respectively²². The overall seroprevalence of *H. pylori* in Taiwan has been reported to be 54.4 per cent in a normal population, 87.4 per cent

Table 1 Clinical data of 128 patients with resectable gastric adenocarcinoma

Variable	Positive for <i>H. pylori</i>	Negative for <i>H. pylori</i>	Total	<i>P</i>
Mean(s.d.) age (years)	57(13.6)	62(14.5)	59(13.7)	n.s.
Sex ratio (M:F)	51:31	22:24	73:55	n.s.
Location				
Upper	14 (17)	7 (15)	21	0.027
Middle	16 (20)	19 (41)	35	
Lower	52 (63)	20 (43)	72	
Gross appearance				
Localized (Borrmann I/II)	19 (23)	3 (7)	22	0.011
Infiltrative (Borrmann III/IV)	43 (52)	36 (78)	79	
Unclassified	20 (24)	7 (15)	27	
Size				
< 4 cm	34 (41)	13 (28)	47	n.s.
4-8 cm	35 (43)	22 (48)	57	
> 8 cm	13 (16)	11 (24)	24	
Blood type				
A	28 (34)	15 (33)	43	n.s.
B	14 (17)	9 (20)	23	
O	35 (43)	19 (41)	54	
AB	5 (6)	3 (7)	8	
Total	82 (64)	46 (36)	128	

Values in parentheses are percentages unless otherwise stated. n.s., not significant

Table 2 Operative and postoperative data

Variable	Positive for <i>H. pylori</i> (<i>n</i> = 82)	Negative for <i>H. pylori</i> (<i>n</i> = 46)	Total (<i>n</i> = 128)	<i>P</i>
Gastrectomy				
Total	20 (24)	17 (37)	37	n.s.
Subtotal	62 (76)	29 (63)	91	
Operation				
Morbidity	17 (21)	16 (35)	33 (26)	n.s.
Mortality	1 (1)	1 (2)	2 (2)	
Postoperative chemotherapy				
Yes	40 (49)	27 (59)	67	n.s.
No	42 (51)	19 (41)	61	
Recurrence				
Yes	21 (26)	21 (46)	42	0.038
No	61 (74)	25 (54)	86	

Values in parentheses are percentages. n.s., not significant

Fig. 1 Cumulative survival curves for 82 patients seropositive (—) and 46 seronegative (---) for *Helicobacter pylori* after resection for gastric cancer. $P < 0.05$ (generalized Wilcoxon test)

in those with duodenal ulcers, 76.2 per cent in those with gastric ulcers and 62.2 per cent in those with gastric adenocarcinoma^{23–25}. Infection in early childhood may be a contributing factor to gastric carcinogenesis¹⁴, but 30–40 per cent of patients with gastric cancer in Taiwan have neither recurrent nor previous infection with *H. pylori*. It would be of interest to determine differences in the behaviour of *H. pylori* between these two groups of patients.

In the present study, there were significant differences in patients with previous *H. pylori* infection with regard to the location and gross appearance of the tumours. Some researchers have noted a positive correlation between *H. pylori* infection and intestinal type metaplasia and the presence of non-cardia cancer^{11,26}. This has not been confirmed by others^{12,27}. The present study supports the positive association of distal site gastric cancer and *H. pylori* infection, but no difference was found between intestinal and diffuse type gastric cancer and *H. pylori* infection. This observation supports the hypothesis of Sipponen *et al.*²⁸ that *H. pylori*-associated chronic gastritis provides the background for gastric cancer carcinogenesis whereas the intestinal type may be promoted by an atrophic stomach and the diffuse type by a non-atrophic gastritis mucosa. Thus, both types of gastric cancer share similar features in *H. pylori* association. The correlation of localized gross type (Borrmann type I and II) gastric carcinoma has not been reported. This finding may also imply a different biology in gastric cancer associated with *H. pylori*. The improved prognosis after surgical treatment of those with *H. pylori*-seropositive gastric cancer may reflect such differences in cancer behaviour. Although the TNM stage remained the most reliable prognostic indicator after resection, different surgical approaches may result in stage migration and thus render valid comparison between different areas difficult²⁹.

The seroprevalence of *H. pylori* infection varies widely. For example, there were high prevalence areas of India (98 per cent)³⁰, Brazil (94 per cent)²⁷ and Africa (85 per cent)³¹, and low prevalence areas in the UK (30 per cent)³² and the Caucasian population of the USA (37 per cent)³³. It is possible for a survival difference to exist where different environmental factors are involved in carcinogenesis. Fortner *et al.*³⁴ observed a significantly poorer survival rate in patients born in the USA than that in patients born outside the USA. Their finding also implied that prognosis may vary with the role of environmental factors involved in carcinogenesis.

The present results suggest that although *H. pylori* seropositivity is not useful as a prognostic factor, it may influence the overall prognosis in different areas. *H. pylori*-seropositive gastric cancer has a better prognosis after surgical resection, which might be attributed to the more advanced stage of *H. pylori*-seronegative cancer. It might be one reason to explain the discrepancy between cancer treatment results between Japanese and Western patients. Further investigations are needed to elucidate the inherent biological characteristics and clinical significance of this specific type of gastric cancer.

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