Pernicious anaemia and Campylobacter like organisms; is the gastric antrum resistant to colonisation?

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SUMMARY Gastric biopsies from 86 patients with pernicious anaemia were examined for Campylobacter like organisms with particular attention to those showing an antral gastritis in addition to the usual pattern of body gastritis. All the patients had chronic atrophic gastritis in the body but Campylobacter like organisms were found at this site in only three patients. Thus the Type A pattern of gastritis (autoimmune) seen in patients with pernicious anaemia is only rarely associated with Campylobacter like organisms. Forty four of these patients had biopsies from body and antrum, 16 showed an antral gastritis of whom only one had Campylobacter like organism present. Twenty five of this latter group of patients were rebiopsied after five years. There was no change in the pattern of gastritis, and the same single patient remained colonised. The frequency of an antral gastritis in patients with pernicious anaemia was 36% yet the frequency of antral colonisation by Campylobacter like organisms was very low (6%). These results show that, as in the body, Campylobacter like organisms are not associated with gastritis when it occurs at this site in pernicious anaemia. The antral gastritis that may accompany body gastritis in pernicious anaemia seems more likely therefore to be an extension of primary type A body gastritis (autoimmune) rather than a secondary type B (chronic) gastritis and, it is argued, the antrum may exhibit resistance to colonisation.

Campylobacter pylori is strongly associated with gastritis.¹⁻³ Of the main patterns of gastritis,^{4,6} the association of *Campylobacter pylori* is with type B.⁵ This corresponds approximately to the hypersecretory pattern described by Correa⁴ and is characterised by predominantly antral inflammation with extension into the body with age. Whether *Campylobacter pylori* is a pathogen rather than an opportunist is still disputed but the experimental production of gastritis in man⁷ and the pig⁸ after ingestion of the organisms favours a pathogenic role. Its absence in patterns of gastritis other than type B might also be evidence in favour of a primary role. In this respect

the organisms are absent in the gastritis associated with bile reflux⁹ and probably in pernicious anaemia. Only limited correspondence^{10,11} documents the situation in the latter, however, which is the major cause of type A⁴ or autoimmune⁵ gastritis. Here in the majority of cases the gastritis characteristically involves the body sparing the antrum. O'Connor *et al*¹⁰ found *Campylobacter pylori* in only three of 14 patients with pernicious anaemia, but the detailed location of colonisation and the state of the antrum were not made clear. When the antrum is normal in patients with pernicious anaemia a low frequency of *Campylobacter pylori* is to be expected for association with a normal mucosa is very rare.¹² It is the frequency of *Campylobacter pylori* in those patients with pernicious anaemia who have antral inflammation that is of interest. Do such individuals simply

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have type B or hypersecretory gastritis superimposed on a type A or autoimmune pattern and with this a correspondingly high rate of *Campylobacter pylori* colonisation?

To answer this question this study looked at the *Campylobacter* status of a large number of patients with pernicious anaemia but in particular that group with an antral gastritis.

Methods

PATIENTS

Gastric biopsies from 86 patients with pernicious anaemia were studied. These were identified from the pathology records of Northwick Park Hospital and the Middlesex Hospital. The diagnosis of pernicious anaemia was based on the finding of a megaloblastic anaemia, a low serum B₁₂ and an abnormal Schilling test which corrected with intrinsic factor. The patients comprised 39 men and 47 women aged 20–88 (median age 58.6 years). Endoscopic biopsies from the gastric body were available in all patients and antral biopsies from 44 of these patients. Twenty five of this group of 44 patients were included in a follow up study for other reasons and a second series of body and antral endoscopic biopsies five years later were available.

Routine paraffin embedded 3 µm sections were cut and stained with haematoxylin and eosin. Although organisms can be seen with this routine stain an additional cresyl fast violet stain was used to help identify *Campylobacter* like organisms.¹³ This has been shown to be a simple and reliable technique. Because of the absence of confirmatory microbiology in this study the term *Campylobacter* like organisms, rather than *Campylobacter pylori*, is used when referring to the histological findings. Chronic gastritis was classified according to the method of Whitehead¹⁴ and the polymorphonuclear leucocyte infiltrate and extent of intestinal metaplasia assessed in a simple semi-quantitative manner (Table 1). The Mann-Whitney U test was used for statistical analysis of the results.

Results

All patients had chronic atrophic gastritis of the body, and 16/44 (36%) had an abnormal antral biopsy (Table 1). The grades of gastritis in the body did not differ in patients with a normal antral biopsy (two mild, 12 moderate, 12 severe body gastritis, and two unclassified) from those with an antral gastritis (one mild, six moderate, eight severe atrophic gastritis, and one unclassified). Intestinal metaplasia was common in the body but rare in the antrum (Table 1). There were no statistical differences

Table 1 Gastric histological findings in patients with pernicious anaemia

	Body	Antrum
Patients (n)	86 (3)	44 (1)
Normal mucosa	0	28
Superficial gastritis	0	3
Atrophic gastritis		
mild	5	7 (1)
moderate	41 (3)	2
severe	33	4
Unsuitable for classification	7	0
Neutrophils		
absent	74 (2)	40 (1)
few	12 (1)	4
moderate	0	0
numerous	0	0
Intestinal metaplasia		
absent	26 (1)	38 (1)
mild	28	2
moderate	23 (2)	1
diffuse	9	3

() *Campylobacter pylori* positive patients.

Table 2 Five year histological follow-up in 25 patients with pernicious anaemia

	Body		Antrum	
	1st biopsy	2nd biopsy	1st biopsy	2nd biopsy
Normal mucosa	0	0	19	19
Superficial gastritis	0	0	3	2
Atrophic gastritis				
mild	0	0	2 (1)	3 (1)
moderate	11 (1)	10 (1)	0	0
severe	11	15	1	1
Unsuitable for classification	3	0	0	0

() *Campylobacter pylori* positive patients.

between the ages of patients with mild/moderate and severe atrophic gastritis in the body, but patients with a normal antrum were significantly younger than patients with antral gastritis (20–79 years, median 53, v 34–88 years, median 69, $p < 0.05$). *Campylobacter* like organisms were seen in the body in only three of the 86 patients (Table 1) of whom one was from the group of 44 in whom there was also an antral biopsy. This showed a mild atrophic gastritis also with *Campylobacter* like organisms present. Apart from this single case all other abnormal antral biopsies were negative for *Campylobacter* like organisms (15/16) (Table 1). In the 25 follow up biopsies at five years no significant histological modification was observed in the body or antral biopsies (Table 2), and the same single patient was again found to be positive for *Campylobacter* like organisms in both body and antrum.

Discussion

Campylobacter like organisms were identified on

microscopy in only three of 86 patients with pernicious anaemia. It is clear that these bacteria only rarely colonise the abnormal body in pernicious anaemia and are unlikely to be involved in the pathogenesis of the disease.^{10,11} In those patients with antral and body biopsies the figure is one of 44 patients. If the observations are restricted further to only patients with abnormal antral mucosae, then the frequency of *Campylobacter* like organisms is one of 16 patients (6.5%). This figure is well below that for the frequency of *Campylobacter pylori* in the hospital population with gastritis not caused by pernicious anaemia^{3,12,15} and even below the carriage level for gastritis found incidentally amongst symptomless controls.^{16,17}

Before considering the significance of this result, it is pertinent to examine the available data on the pathology of the antrum in pernicious anaemia. The classification of gastritis is broadly in three patterns.^{4,6} Type A⁴ corresponds approximately to autoimmune gastritis⁵ and predominantly affects the body of the stomach. Type B⁴ relates to hypersecretory gastritis⁵ that predominantly involves the antrum with variable proximal spread. This is the pattern associated with peptic ulcer disease in general, and duodenal ulceration in particular. The third pattern environmental chronic gastritis⁵ is the type seen in high incidence areas of gastric cancer. It may correspond to the AB type.⁶ Topographically it is multifocal and centred along the junction of antrum and body. Most of the studies on *Campylobacter pylori* and gastritis have been on the type B or hypersecretory pattern.^{1-3,12,15} Type A or autoimmune gastritis, the form associated with pernicious anaemia, is believed to spare the antrum.⁴ As *Campylobacter pylori* rarely colonises the normal antrum,¹² this would seem an adequate explanation for their absence in pernicious anaemia. From our results, however, and several other studies it is clear that the antrum can be abnormal in pernicious anaemia. Indeed Lewin *et al*¹⁸ claim the antrum to be invariably abnormal, Stockbrugger *et al*¹⁹ quote an incidence of 21% for antral inflammation whilst Kekki *et al*²⁰ suggest the changes in the antrum are dynamic. Thus early on in the natural history of pernicious anaemia antral gastritis is common but Kekki *et al* claim there is a return to normality as fully established gastric atrophy develops in the body. These reports¹⁸⁻²⁰ do not make it clear whether the antral involvement is all part of the pattern of type A gastritis or is merely coincidental development of type B gastritis. Day²¹ and Owen²² favour the latter suggestion. Whichever is the correct interpretation, in this study 36% of patients with pernicious anaemia were found to have antral gastritis. The failure to find any changes in the degree of gastritis in those patients

who were followed up and biopsied at five years argues against Kekki *et al*'s hypothesis.²⁰

Several factors might contribute to the low frequency of *Campylobacter pylori* found in pernicious anaemia, be it in the antrum or body. The simplest explanation is that the aetiology of the antral gastritis in patients with pernicious anaemia is the same as that in the body, namely an autoimmune one, and that *Campylobacter pylori* has no role at either site. On the other hand there was no apparent histological difference in the antral gastritis seen here as part of the picture of pernicious anaemia and that seen in the more common type B pattern.³ Special stains to detect changes in lymphocyte subpopulations or other immunological differences were, however, not carried out. A clinical difference observed was that the patients with antral disease were older than those with a normal antrum. This is a point in favour of the antral involvement being a superimposed type B or hypersecretory gastritis, as is finding the frequency of antral gastritis to be 36% which is similar to that of type B gastritis in randomly selected populations.²³ Set against this are the data from those patients who were rebiopsied at five years. These might have been expected to show increased antral abnormalities but this was not the case. Another explanation for the rarity of *Campylobacter pylori* in pernicious anaemia might be on the basis of widespread intestinal metaplasia, for *Campylobacter pylori* do not associate with this mucosal pattern.^{24,25} This seems unlikely here as intestinal metaplasia in the antrum was widespread in only three cases (Table 1). Few of the biopsies showed activity in the form of neutrophils infiltrating the surface epithelium and gastric pits. Some workers^{1,12} suggest the frequency of organisms correlates with this activity but the differences in colonisation frequency seen here are too great for this explanation. Moreover, not all investigators acknowledge this correlation.²³

Irrespective of whether the antral gastritis in pernicious anaemia is an extension of an autoimmune process into the antrum or a superimposed pattern of hypersecretory (type B) gastritis the frequency of *Campylobacter* like organisms in the involved antrum is still well below that found in control groups.^{16,17} Therefore, the antrum in pernicious anaemia appears to be susceptible to gastritis but not to colonisation by *Campylobacter pylori*; a suggestion reinforced by the absence of any change in the incidence of colonisation in the patients rebiopsied after five years and given the ubiquitous nature of the organism.^{2,3,15,24,26}

Contrasting situations are seen therefore between duodenal ulcer disease and pernicious anaemia in relation to *Campylobacter pylori* status. In patients with duodenal ulcer disease acid levels are high and

up to 90% of patients exhibit antral gastritis associated with the organisms.^{3,26} In this situation the duodenal mucosa is also colonised if there is gastric metaplasia.²⁶ On the other hand in patients with pernicious anaemia acid is absent, and even when antral gastritis is present, as shown here, there is almost a total absence of *Campylobacter pylori*. Moreover the frequency of *Campylobacter pylori* is low in patients with bile reflux which too might reflect neutralisation of gastric acid.⁹ Thus situations in which antral [pH] is likely to be raised are those in which *Campylobacter pylori* are absent and *vice-versa*. The mechanism may simply reflect competitive exclusion of *Campylobacter pylori* by other bacteria which are known to colonise the stomach in large numbers once the [pH] rises.²⁷ Conditions for antral colonisation seem unlikely to depend directly on acid and a low [pH]. *Campylobacter pylori* are inhibited by acid though can survive a [pH] of 2.6 in the presence of urea²⁸ but their urease activity²⁹ is inhibited below a [pH] of 4.5.³⁰ Furthermore the bacteria are closely associated with the epithelial cell surface which is below the mucus layer^{3,25} and therefore protected from acid exposure. In addition periods of hypoacidity seem a necessary part of experimental colonisation susceptibility in man⁷ as well as being a feature of epidemic outbreaks.^{31,32} Finally, H₂ antagonists fail to eliminate the organisms.³³ Thus gastric acidity at the most only contributes to a suitable environment for *Campylobacter pylori*, or at the least is simply a marker of some other function of the gastric body mucosa essential to the organisms successful antral association. Indeed there are data suggesting that antral colonisation might be dependent on the integrity of the gastric body mucosa.³⁴

In conclusion we have shown that antral gastritis unassociated with *Campylobacter* like organisms occurs in the type A or autoimmune gastritis of pernicious anaemia. It is possible that the former is immune mediated and represents the same process as in the gastric body. On the other hand, the antral involvement may represent superimposed hypersecretory or type B gastritis. This would seem unlikely unless either *Campylobacter pylori* is merely an opportunist and the antrum in pernicious anaemia resistant to colonisation, or that the gastric environment in pernicious anaemia selects out that small percentage of patients found in studies of type B gastritis^{2,3,15,24,26} that have no associated *Campylobacter pylori* and for whom another aetiology must be found.

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