

Influence of *Helicobacter pylori*, sex, and age on serum gastrin and pepsinogen concentrations in subjects without symptoms and patients with duodenal ulcers

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Abstract

The relation between *Helicobacter pylori* (*H pylori*) infection and fasting gastrin and pepsinogen-I and -II concentrations was evaluated in 278 volunteers without symptoms and the results were compared with the values obtained in 35 patients with duodenal ulcers. *H pylori* infection was determined with the ¹³C-urea breath test in subjects without symptoms and with endoscopy, biopsy (histology and culture), and quick urease test (CLO-test) in patients with duodenal ulcers. Gastrin and pepsinogen-I and -II concentrations were assayed with specific radioimmunoassay systems. The results clearly indicate that fasting gastrin and pepsinogen-I and -II concentrations were significantly higher in *H pylori* positive compared with *H pylori* negative subjects. Neither age nor sex affected basal gastrin and pepsinogen concentrations in *H pylori* negative subjects. Fasting gastrin, pepsinogen-I and -II concentrations in serum samples were similar in *H pylori* positive persons with no symptoms and those with duodenal ulcers suggesting that similar mechanisms are involved in increasing plasma concentrations of these variables in both populations. Hypergastrinaemia and hyperpepsinogenaemia are therefore probably secondary to active *H pylori* infection.

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Colonisation of the gastric mucosa by *Helicobacter pylori* (*H pylori*) is associated with chronic gastritis and peptic ulcer disease, but the role of *H pylori* in the pathogenesis of peptic ulcer disease is still unclear.¹ Thus although eradication of *H pylori* has been shown to dramatically reduce ulcer relapse rates,^{2,3} we still do not know how *H pylori* causes ulcers. Besides *H pylori*, aggressive factors (acid, pepsin) are necessary for peptic ulcer pathogenesis. Measurement of serum gastrin and pepsinogen concentrations is considered a diagnostic tool to indirectly assess these factors.⁴ Information on the interrelations between *H pylori*, chronic gastritis, and circulating gastrin or pepsinogen concentrations seems, however, confusing⁵ and in particular, there is no consensus on how the mechanisms interact.

Recently, it has been suggested that in humans gastric acid and serum pepsinogen secretion rates increase with age as does the prevalence of *H pylori* infection.¹⁶ Although *H pylori* infection was associated with decreased acid secretion rates in the few subjects studied, serum gastrin concentrations surprisingly did not differ

between *H pylori* positive and *H pylori* negative subjects.⁶

The aim of this study was therefore to further explore the potential relation between serum gastrin and pepsinogen-I and -II concentrations and *H pylori* infection in a large number of *H pylori* positive and *H pylori* negative, subjects without symptoms and to compare the results with those in patients with duodenal ulcers. We also aimed to re-evaluate the effects of age and sex on these variables.

Methods

The study was approved by the human research and review committees of the participating centres and subjects gave written informed consent.

SUBJECTS WITHOUT SYMPTOMS

Caucasian subjects without symptoms and without a history of dyspepsia, peptic ulcer disease, or other upper gastrointestinal symptoms or abdominal surgery (with the exception of appendectomy) were carefully recruited by word of mouth among coworkers, relatives, and friends of the investigators, and fellow workers. Some elderly subjects were recruited from senior citizens' homes or choirs. No one had taken any antacids, bismuth preparations, or other drugs to treat dyspepsia within the past year and nobody was taking antibiotics during the three months before the study. The 278 volunteers thus recruited comprised 155 men aged 21 to 91 years (median 42 years) and 123 women aged 18 to 83 years (median 43 years).

After an overnight fast, a ¹³C-urea breath test (UBT) was performed according to published methods⁷ and a plasma sample was obtained for gastrin, pepsinogen-I, and pepsinogen-II determinations. The plasma was stored at -20°C until analysis.

PATIENTS WITH DUODENAL ULCERS

Patients referred for upper gastrointestinal endoscopy because of suspected duodenal ulcers were invited to participate in the study. The patients gave written informed consent. Exclusion criteria included previous gastric surgery, known bleeding diathesis, oral anticoagulation, or recent (during the three months before the study) treatment with bismuth compounds, antisecretory agents (H₂ antagonists, omeprazole), or antibiotics. Thirty five patients with duodenal ulcer at endoscopy were finally

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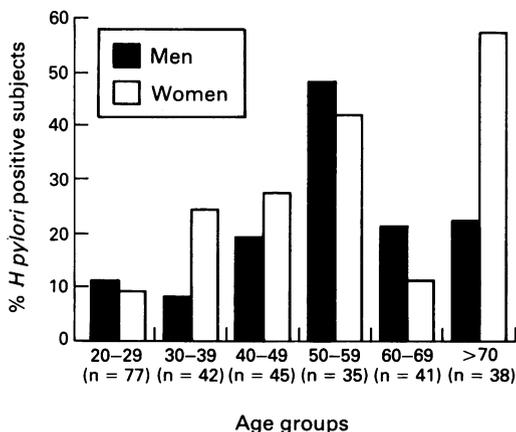


Figure 1: Prevalence of H pylori infection in a population without symptoms.

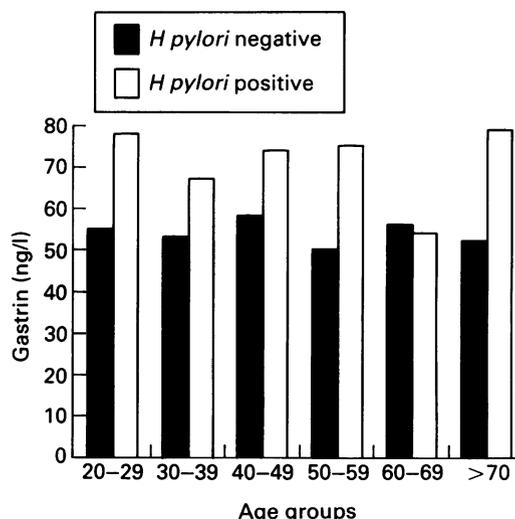


Figure 2: Mean age specific concentrations of fasting serum gastrin in H pylori positive and H pylori negative subjects without symptoms.

included (30 men aged 20 to 60 years, median 36 years; five women aged 26 to 50 years, median 36 years). During endoscopy, performed in the morning after an overnight fast, five biopsies were taken from the antrum for the detection of H pylori: one for a quick urease test (CLO-test), two for culture with standard methods,⁸ and two for histology. The endoscopes were sterilised after each examination according to local standards; the biopsy forceps were sterilised by autoclaving. Based on these tests, all 35 subjects could be unequivocally classified as H pylori positive. A plasma sample was taken for gastrin, pepsinogen-I, and pepsinogen-II determinations and stored at -20°C until analysis.

analysis of variance. If this step showed a significant difference for the variable examined, then pairs of subgroups were examined by the Newman-Keuls post hoc test. Differences in proportions in H pylori positive and H pylori negative subjects were evaluated by χ^2 test. Correlation coefficients were calculated by Pearson's method.¹⁰ A p value <0.05 was considered significant.

GASTRIN RADIOIMMUNOASSAY

Radioimmunoassay of coded serum samples was performed as described recently.⁹

PEPSINOGEN-I AND -II RADIOIMMUNOASSAY

Radioimmunoassay for serum pepsinogen-I and -II of coded serum samples were performed with commercial kits (Pepsik-I and -II, SORIN Biomedica, Saluggia, Italy).

STATISTICAL ANALYSIS

For each variable examined, the difference between men and women was tested for significance by the non-parametric Mann-Whitney U test. Differences in each variable among H pylori positive subjects (men or women) and H pylori negative subjects (men or women) were tested by

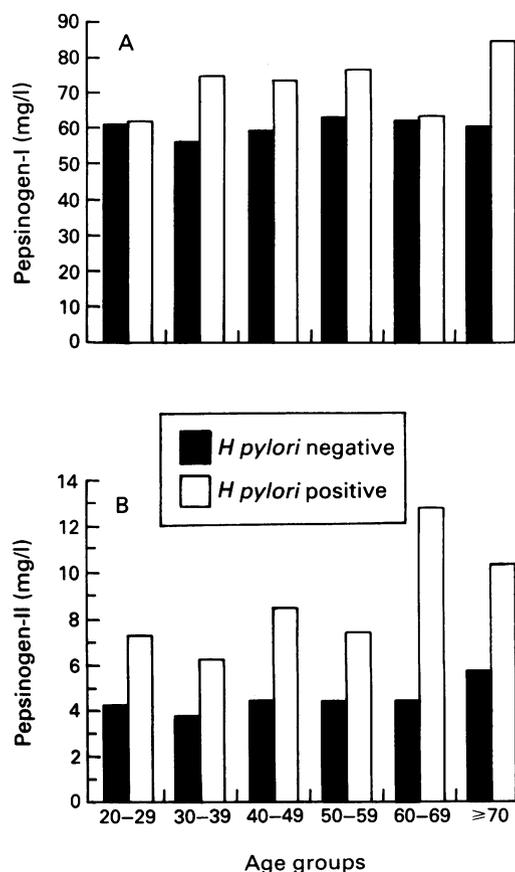


Figure 3: Mean age specific concentrations of fasting pepsinogen-I (A) and pepsinogen-II (B) in H pylori positive and H pylori negative subjects without symptoms.

Serum gastrin and serum pepsinogen concentrations (mean (SEM)) in H pylori negative and H pylori positive subjects without symptoms and patients with duodenal ulcers

	H pylori negative. No symptoms		H pylori positive. No symptoms		Patients with duodenal ulcers (n=35)
	Men (n=125)	Women (n=91)	Men (n=30)	Women (n=32)	
Gastrin (pg/ml)	52 (1)	58 (2)	73 (5)***	74 (5)††	59 (3)**
Pepsinogen-I (ng/ml)	62 (2)	57 (2)	81 (10)*	67 (5)†	72 (5)*
Pepsinogen-II (ng/ml)	4.3 (0.2)	4.3 (0.2)	9.3 (1.7)***	7.9 (1.0)†††	6.7 (0.7)**
Ratio-I:-II	15.8 (0.5)	14.9 (0.6)	11.4 (1.0)***	11.0 (1.0)††	10.4 (0.8)**

*p<0.01; **p<0.001; ***p<0.0001 v H pylori negative men by Mann-Whitney test. †p<0.05; ††p<0.001; †††p<0.0001 v H pylori negative women by Mann-Whitney test.

Results

SUBJECTS WITHOUT SYMPTOMS

H pylori

H pylori infection was present in 62 of 278 subjects – that is, in 22% of the population studied (Fig 1). The frequency of *H pylori* infection in men and women was similar and increased with age as previously reported.^{7,11-13}

Gastrin

Basal serum gastrin concentrations were significantly higher in *H pylori* positive than in *H pylori* negative subjects ($p < 0.001$). Age and sex did not significantly affect basal gastrin concentrations (Table, Fig 2), irrespective of *H pylori* state.

Pepsinogen concentrations

Basal serum pepsinogen-I and -II concentrations were significantly higher in *H pylori* positive than in *H pylori* negative subjects (Table, Fig 3). There was a significant decrease in the ratio of pepsinogen-I to pepsinogen-II in *H pylori* positive compared with *H pylori* negative subjects ($p < 0.001$), because in *H pylori* positive subjects the increase in pepsinogen-II was more pronounced than the increase in pepsinogen-I. As with serum gastrin, neither age nor sex influenced serum pepsinogen concentrations.

PATIENTS WITH DUODENAL ULCERS

H pylori

All 35 patients with duodenal ulcers were clearly identified as *H pylori* positive (35 positive by culture, 35 positive by histology, 34 positive in the CLO-test).

Gastrin

As there were only five women with duodenal ulcer disease and as there were no differences apparent, the results shown in the Table are pooled for male and female patients. Basal serum gastrin concentrations were similar in patients with duodenal ulcers compared with *H pylori* positive subjects without symptoms, but were significantly ($p < 0.001$) higher than in *H pylori* negative subjects without symptoms (Table). There was no effect of age on basal serum gastrin concentrations (data not shown).

Pepsinogen concentrations

Pepsinogen-I and pepsinogen-II concentrations in patients with duodenal ulcers were again similar to the values found in *H pylori* positive, subjects without symptoms, but significantly higher than those in *H pylori* negative subjects. Again, no age dependence was detectable (data not shown).

Discussion

The effect of age and sex on basal serum gastrin and pepsinogen secretion in healthy people as

well as in patients with duodenal ulcers has been investigated previously by different groups.^{6,13-19} Most of them did not control for a possible confounding effect of *H pylori* infection. Moreover, in a recent study it was suggested that aging is associated with increasing gastrin and pepsinogen secretion in humans, particularly in men.⁶ Although infection with *H pylori* was associated with lower acid secretion rates, no differences in basal gastrin concentrations were found between *H pylori* positive and *H pylori* negative subjects. Only 11 subjects with *H pylori* infection were examined in the study, however. Here we have examined a large group of subjects without symptoms covering a broad age range and in whom the *H pylori* state was known. Our findings can be summarised as: (1) basal gastrin and pepsinogen-I and -II concentrations were significantly influenced by *H pylori* infection, being higher in *H pylori* positive compared with *H pylori* negative subjects; (2) in a large group of *H pylori* negative subjects, neither age nor sex affected basal serum gastrin or pepsinogen concentrations.

In the present study, 62 of 278 Swiss subjects with no symptoms were *H pylori* positive (22%). This figure is comparable with data from other industrialised countries such as Great Britain, the United States, Japan, Australia, and New Zealand with an apparent overall prevalence of 15–25%.²⁰ Similarly, age was associated with an increase in *H pylori* prevalence, but the peak of *H pylori* positive subjects was found in the group aged between 50 and 60 years, whereas the rate tended to be lower in older subjects. This phenomenon as been noted previously by other groups²⁰ and could be associated with progression of the chronic *H pylori* gastritis to atrophic gastritis in elderly people, because severe gastric atrophy seems to make the stomach inhospitable to *H pylori*.^{6,21,22} Alternatively, as teeth and gums have been proposed to act as a reservoir of *H pylori* infection,²³ people becoming more edentulous with increasing age may lose their *H pylori* reservoir.

The present study has also confirmed that *H pylori* affects circulating gastrin concentrations.²⁴⁻²⁷ Gastrin is a gut hormone produced by G cells located in the gastric antrum.²⁸ The factors that regulate gastrin secretion are complex. Here we add to the complex as we have clearly shown that basal gastrin concentrations are neither affected by age nor by sex, but are influenced by *H pylori*. As basal gastrin levels can be returned to normal with eradication of *H pylori*,²⁹⁻³¹ the data indicate that the increase in gastrin is induced by the infection. These findings have physiological and clinical implications. Firstly, *H pylori* infection apparently interferes with the physiological mechanisms regulating gastrin release. Future studies evaluating mechanisms of gastrin release have to take into account the *H pylori* state. Secondly, high gastrin concentrations may induce increased parietal cell mass and acid secretion, which in turn could contribute to the pathogenesis of duodenal ulceration. It has, however, not been shown up to now that eradication of *H pylori* modifies parietal cell mass or acid secretion rates.

Recent work has focused on the gastrin acid

secretion feedback loop and identified a number of potential pathogenetic pathways.³² The finding of enhanced fasting gastrin concentrations in *H pylori* positive subjects and in duodenal ulcer disease cannot easily be explained. Firstly, *H pylori* could increase gastrin release by production of ammonia through its enzyme urease. This would raise the pH within the antrum mucus layer, which in turn would prevent inhibition of gastrin release through a feedback mediated mechanism by low intragastric acidity. Secondly, gastrin concentrations could be increased through local inflammation as gastrin concentrations are also increased in patients with non-*H pylori* induced gastritis.³³ Beardshall and coworkers have shown that suppression of *H pylori* with tripotassium dicitratobismuthate and metronidazole decreased the response of gastrin to gastrin releasing peptide stimulation.³⁴ These findings support the hypothesis that *H pylori* colonisation is directly involved in the hypergastrinaemia. Bismuth has, however, effects on the gastric mucosa apart from that on *H pylori*. It is therefore possible that the changes found in the study of Beardshall *et al* are not related to suppression of *H pylori* although this remains the most likely explanation.³⁴ Further studies are still necessary to determine the mechanism by which *H pylori* increases fasting plasma gastrin concentrations in *H pylori* positive subjects and in patients with duodenal ulcer disease.

In the present study we also analysed the relation between active *H pylori* infection and serum pepsinogen-I and pepsinogen-II concentrations. Both pepsinogens are present in the chief cells of the oxyntic glands of the gastric corpus mucosa, but only pepsinogen-II is present in the gastric antrum. We found that the serum concentrations of both pepsinogens were increased in persons without symptoms but with active *H pylori* infection compared with those without *H pylori*. Interestingly, the effect was more pronounced with pepsinogen-II, indicating that diffuse antral gastritis leads either to increased pepsinogen-II secretion or to increased leakage of pepsinogen-II into the circulation. Although the mechanism by which *H pylori* infection produces hyperpepsinogaemia remains unknown, it has been shown before that patients with gastritis have increased serum pepsinogen concentrations.³⁵⁻³⁶ In fact, measurement of pepsinogen concentrations had been proposed as a method for screening of gastritis.²¹ We have shown that the serum pepsinogen-I:-II ratio is significantly lower in *H pylori* positive subjects than in uninfected persons. Again, neither age nor sex affected pepsinogen-I or pepsinogen-II concentrations.

The distribution and severity of chronic gastritis induced by *H pylori* in patients with duodenal ulcer disease seems to be different from those in *H pylori* positive subjects without symptoms.¹³⁻³⁷⁻³⁹ It might be anticipated, therefore, that the pattern of gastrin and pepsinogen would differ in *H pylori* positive subjects without symptoms and in patients with duodenal ulcers. We have shown, however, that basal gastrin pepsinogen-I and -II concentrations are similar in these groups, suggesting that similar mecha-

nisms are involved in increasing plasma concentrations of gastrin and pepsinogen in both populations. Similar results have recently been published for basal gastrin concentrations in a small group of subjects matched for sex and age.⁴⁰

In summary, *H pylori* infection is associated with increases in basal gastrin and pepsinogen-I, and pepsinogen-II concentrations. Basal gastrin and pepsinogen-I, and pepsinogen-II concentrations were not influenced by sex or age and were similar in patients with duodenal ulcers and in *H pylori* infected volunteers without symptoms, suggesting that hypergastrinaemia and hyperpepsinogaemia are secondary to active *H pylori* infection.

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