STOMACH

Interleukin 1B proinflammatory genotypes protect against gastro-oesophageal reflux disease through induction of corpus atrophy

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Background and aims: The relationship between *Helicobacter pylori* infection and gastro-oesophageal reflux disease (GORD) is controversial but it is accepted that GORD is associated with increased exposure to gastric acidity. The proinflammatory interleukin (*IL*)-*1B* polymorphisms increase the risk of hypochlorhydria and gastric atrophy. We examined the association between proinflammatory cytokine gene polymorphisms, presence of gastric atrophy, and risk of GORD in *H pylori* positive and negative subjects in Japan.

Methods: We studied 320 consecutive dyspeptic patients without peptic ulcers or cancers. GORD symptoms were scored using the Carlsson-Dent questionnaire and erosive oesophagitis was assessed endoscopically. *H pylori* infection was diagnosed by urea breath test, histological examination, and serology. Gastric atrophy was assessed histologically, and polymorphisms in the *IL-1B, IL-10*, and tumour necrosis factor α (*TNF-A*) genes were genotyped.

Results: Two hundred and eight patients were *H pylori* positive and 112 were negative. One hundred and eight (34%) were found to have erosive oesophagitis by endoscopic criteria (grade A: 78; grade B: 23; grade C: 6; grade D: 1). Erosive oesophagitis and GORD symptoms were significantly more common in *H pylori* negative compared with *H pylori* positive subjects ($p<0.05$). *H pylori* positive subjects were more likely to have corpus gastric atrophy than *H pylori* negative subjects ($p<0.001$). Among *H pylori* positive patients, those without erosive oesophagitis or GORD symptoms were significantly more likely to have corpus atrophy than subjects with erosive oesophagitis or GORD symptoms ($p<0.05$). Among *H pylori* positive patients, subjects homozygous for the proinflammatory allele *IL-1B*−511T had a significantly lower risk of erosive oesophagitis (odds ratio (OR) 0.06 (95% confidence interval (CI) 0.006–0.51); $p=0.01$) and GORD symptoms (OR 0.10 (95% CI 0.01–0.85); $p=0.04$) compared with those homozygous for the −511C allele, while none of the two other proinflammatory cytokine gene polymorphisms had significant correlations with erosive oesophagitis or GORD symptoms.

Conclusions: A proinflammatory *IL-1B* genotype is associated with increased risk of atrophy and decreased risk of GORD in *H pylori* infected subjects in Japan. These data indicate that in some genetically predisposed subjects, *H pylori* infection may protect against GORD through induction of gastric atrophy.

Gastro-oesophageal reflux disease (GORD) refers to the reflux of gastric contents into the oesophagus, leading to mucosal damage and/or symptoms of heartburn and regurgitation. Although the symptoms of GORD are common, many patients do not have erosive oesophagitis diagnosed on endoscopy.[1] Proton pump inhibitors seem to be equally effective in treating non-erosive reflux disease (NERD) and erosive oesophagitis, indicating that excessive gastric acid secretion may be associated with both conditions.[2] Reflux disease is underlined by a number of well described pathophysiological mechanisms but recent interest has focused on the role of *Helicobacter pylori* in this condition. Specifically, there has been debate over whether this infection and its associated divergent patterns of gastritis may impact against reflux disease and its complications. Some reports have indicated a lower prevalence of *H pylori* infection in patients with GORD,[3–7] and particularly low prevalence of the virulent *cagA* positive strains in patients with complicated GORD.[4 5 8] However, there are also some studies that have found no causal relationship between *H pylori* infection and GORD.[9 10] A number of factors, including lack of use of validated reflux symptom questionnaires, differences in the definition of reflux, method of *H pylori* detection, differences in *H pylori* genotypes, inclusion of inappropriate control groups, lack of documentation of patterns of gastritis (that is, antrum predominant versus corpus predominant or pangastritis), and differences in host genetic factors probably account for the discrepancies between the various published reports.

H pylori infection can have varying effects on gastric acid secretion, and the changes largely depend on the pattern and severity of gastritis. Antrum predominant gastritis is associated with hypergastrinaemia, gastric hypersecretion, and duodenal ulcer disease, while patients with corpus predominant gastritis and/or multifocal atrophic gastritis have decreased acid secretion and increased risk of gastric ulcer and gastric carcinoma.[11–13] Host and environmental factors, as well as bacterial virulence characteristics, determine the development of *H pylori* associated upper gastrointestinal diseases.[14 15] Host genetic factors are key determinants of risk

Abbreviations: GORD, gastro-oesophageal reflux disease; IL, interleukin; NERD, non-erosive reflux disease; TNF-α, tumour necrosis factor α; ELISA, enzyme linked immunosorbent assay; PCR, polymerase chain reaction; OR, odds ratio

for many benign and malignant diseases, particularly in the stomach.[15–19]

A variety of pro- and anti-inflammatory cytokines are expressed in *H pylori* infected gastric mucosa. One of the key cytokines that is upregulated in the gastric mucosa by infection is interleukin (IL)-1β.[20] This cytokine is important in initiating and amplifying the inflammatory responses against the bacterium and is also a potent inhibitor of gastric acid secretion.[21–24] The *IL-1B* gene encoding IL-1β is highly polymorphic, and several diallelic polymorphisms have been reported. Two of these are in the promoter region at positions −511 and −31, representing C-T and T-C transitions, respectively. Several studies have shown that these two polymorphisms are in near total linkage disequilibrium.[16 25] These polymorphisms have been shown to significantly affect gastric mucosal IL-1β production in response to *H pylori* infection.[26 27] The effect of these polymorphisms on gastric acid secretion is therefore most likely mediated through higher production of IL-1β, which is a potent inhibitor of acid secretion.[21 23] In addition to the *IL-1* gene cluster, proinflammatory genotypes of tumour necrosis factor α (*TNF-A*) and *IL-10* were each associated with the risk of non-cardia gastric cancer.[18 28]

Recently, Queiroz *et al* reported that *IL-1B* and *IL-1RN* proinflammatory genotypes were associated with a decreased risk of erosive oesophagitis.[29] The reflux group in that study excluded NERD patients. In the current study, we hypothesised that the proinflammatory *IL-1B* polymorphisms might decrease the risk of GORD symptoms and erosive oesophagitis by increasing the development of hypochlorhydria and gastric atrophy, especially among *H pylori* infected subjects. We examined prospectively the effect of the proinflammatory cytokine polymorphisms (*IL-1B*−511 C/T, *IL-10*−819T/C, and *TNF-A*−1031T/C) on *H pylori* induced gastric atrophy, reflux symptoms, and erosive oesophagitis.

METHODS

Study subjects

We studied consecutive outpatients with dyspeptic symptoms who underwent upper gastrointestinal endoscopy between June 2002 and January 2003 at the Gastroenterology Department of Nagoya University Graduate School of Medicine (table 1). Patients who were found to have peptic ulcers, cancers, polyps, or any other specific lesions in the upper gastrointestinal tract were excluded. A total of 320 patients were recruited and these comprised subjects with macroscopically normal upper gastrointestinal mucosa or with gastritis and/or oesophagitis. None of these patients had taken non-steroidal anti-inflammatory drugs, proton pump inhibitors, antibiotics, or bismuth compounds in the three months preceding upper gastrointestinal endoscopy. Two biopsy specimens per site were obtained from the antrum and corpus and were used for routine histological examination (haematoxylin-eosin and Giemsa stains). *H pylori* infection was assessed by ^{13}C urea breath testing, histological examination by Giemsa stain, and the presence of serum IgG antibody against *H pylori*. All subjects gave written informed consent, and the study was approved by the ethics committee of the Nagoya University Graduate School of Medicine.

Evaluation of GORD symptoms

GORD symptoms in all patients were evaluated using the Carlsson-Dent reflux scale.[30] This scale uses seven items to evaluate the nature of the sensations experienced by patients and the temporal relationship of symptom occurrence to factors that are known to provoke (meals, bending, stooping, lifting), exacerbate (fatty or spicy food), or relieve (antacids) gastro-oesophageal reflux. Each response was assigned a positive, neutral, or negative score, and the score for each item was weighted so that the highest positive values were assigned to factors considered strongly indicative of the diagnosis of GORD. A score ranging from −7 to +18 was calculated by adding the individual positive and negative scores from the questionnaire. Patients were regarded as having reflux symptoms if their scores were higher than 6. This cut off point was estimated by the authors to have a sensitivity of 54 % and specificity of 60%.

Endoscopic study

A total of 320 patients with or without oesophagitis were included (table 1). Oesophagitis was assessed by endoscopic examination, and mucosal breaks were graded as A, B, C, or D according to the Los Angeles classification of oesophagitis[31]: grade A: one or more mucosal breaks confined to the mucosal folds, each no longer than 5 mm; grade B: at least one mucosal break more than 5 mm long confined to the mucosal folds but not continuous between the tops of two mucosal folds; grade C: at least one mucosal break continuous between the tops of the two or more mucosal folds but not circumferential; and grade D: circumferential mucosal break.

H pylori and CagA antibody

Fasting blood samples were analysed for *H pylori* antibody and CagA status. Serum was sampled before endoscopy and all samples were stored at −70℃ until they were analysed in the same batch. Anti-*H pylori* IgG antibody tests, high molecular weight campylobacter associated protein with antigens extracted from clinically isolated Japanese *H pylori* strains (J-HM-CAP), and enzyme linked immunosorbent assay (ELISA) (Kyowa Medex, Tokyo, Japan) were used for identification of *H pylori* infected subjects. An ELISA value of 2.3 or over was regarded as positive for both tests. CagA serology was performed by ELISA, with horseradish peroxidase as the enzyme tracer (CagA IgG EIA; Sceti Co. Ltd,

Table 1 Patient characteristics

Endoscopic oesophageal findings	No	Age (mean (SD))	Reflux symptom		*H pylori* status		Corpus gastric atrophy	
			+	−	Positive	Negative	Yes	No
No erosive oesophagitis	212	47 (8)	47	165	159	53	113	99
Erosive oesophagitis								
Grade A	78	43 (10)	59	19	37	41	16	62
Grade B	23	47 (7)	18	5	10	13	4	19
Grade C	6	47 (5)	5	1	2	4	0	6
Grade D	1	46	1	0	0	1	0	1
Total	108	45 (10)	83	25	49	59	20	88
Total	320	46 (9)	130	190	208	112	133	187

Table 2 Polymerase chain reaction (PCR) conditions for genotyping of interleukin (*IL*)-*1B*, *IL-10*, and tumour necrosis factor α (*TNF-A*) markers

Polymorphism	Primers	Temperature, time, and cycles for PCR; polymerase; PCR method; and definitions of the allele
IL-1B C to T at −511	F 5′-CTG CAT ACC GTA TGT TCT CTG CC R 5′-GGA ATC TTC CCA CTT ACA GAT GG	5 min at 94°C, 30 cycles of 30 s at 94°C, 59°C, and 72°C, and 5 min at 72°C; AmpliTaq Gold; PCR-RFLP (*Eco*811); C allele: 194 bp, T allele: 109 bp and 85 bp
IL-10 T to C at −819	F1 5′-TTT AGA CTC CAG CCA CAG AAG R1 5′-GCA AAC TGA GGC ACA GAG ATA F2 5′-CCC TTG TAC AGG TGA TGT AAC R2 5′-TAC TTT CCA GAG ACT GGC TTC	5 min at 94°C, 30 cycles of 1 min at 94°C, 57°C, and 72°C, and 5 min at 72°C; AmpliTaq Gold; PCR-CTPP; C allele: 407 bp and 275 bp, T allele: 407 bp and 177 bp
TNF-A T to C at −1031	F1 5′-AAG GCT CTG AAA GCC AGC TG R1 5′-CCA GAC CCT GAC TTT TCC TTC A F2 5′-GAA GCA AAG GAG AAG CTG AGA AGA C R2 5′-CTT CCA TAG CCC TGG ACA TTC T	10 min at 95°C, 30 cycles of 1 min at 95°C, 66°C, and 72°C, and 5 min at 72°C; AmpliTaq Gold; PCR-CTPP; C allele: 444 bp and 174 bp, T allele: 444 bp and 316 bp

RFLP, restriction fragment length polymorphism; CTPP, confronting two pair primers.

Tokyo, Japan). The anti-CagA IgG antibody concentration in standards and samples was measured in a spectrophotometer at 450 nm.

Histology

Glandular atrophy was assessed by a single expert gastrointestinal pathologist who used the updated Sydney system[32] without knowledge of the experimental results. An assessment of *H pylori* was made after staining of serial tissue sections with Giemsa.[33 34] Two biopsies per site (antrum and corpus) were used and when the scores between the two biopsies differed, the more severe scores were selected.

Genetic analysis

DNA was extracted from 200 µl of buffy coat preserved at −40°C by the use of QIAamp DNA Blood Mini Kit (Qiagen Inc., Valencia, California, USA). Polymerase chain reaction (PCR) amplification was conducted using the primers listed in table 2. Genomic DNA (50 ng) was used in a volume of 25 µl with 0.1 mM dNTPs, 25 pmol of each primer, 0.5 units of ''Takara Taq'' (Takara Shuzo Co. Ltd, Otsu, Japan) or ''Ampli Taq Gold'' (Perkin-Elmer Corp., Foster City, California, USA), and 2.5 µl of 10× PCR buffer, including 15 mM $MgCl_2$. The PCR-restriction fragment length polymorphism method was used for genotyping the *IL-1B*−511 C/T polymorphism. For genotyping the *IL-10*−819T/C and *TNF-A*−1031T/C polymorphism, a PCR-confronting two pair primers method was applied. This method for single nucleotide polymorphism genotyping does not require a step to digest DNA products (table 2).[35] All PCR products were visualised on a 2% agarose gel with ethidium bromide staining.

Table 3 Localisation of glandular atrophy in the different clinical groups based on the presence or absence of erosive oesophagitis and stratified by *Helicobacter pylori* infection

	Glandular atrophy		
	None	Antrum only	Antrum and corpus
H pylori negative cases			
Oesophagitis (n = 59)	54	3	2
No oesophagitis (n = 53)	48	2	3
Total (n = 112)	102	5	5
H pylori positive cases			
Oesophagitis (n = 49)	4	27	18
No oesophagitis (n = 159)	13	36	110
Total (n = 208)	17	63	128

Statistical analysis

Statistical analysis was performed using the χ^2 test, and an unconditional logistic model was applied for estimating odds ratios (ORs) and interaction terms by a computer program (STATA version 6; STATA Corp., College Station, Texas, USA). The product variable between gene and gene was included in the logistic model when evaluating the multiplicative interactive effects of selected gene pairs. ORs for erosive oesophagitis, GORD symptoms, and the presence of corpus glandular atrophy were adjusted for age and sex. A p value < 0.05 was considered statistically significant.

RESULTS

A total of 320 patients with or without oesophagitis were studied, with a mean age of 46 years (range 35–60). Of the 320 patients studied, 130 (41%) satisfied the criteria for GORD symptoms (table 1). A total of 208 patients (65%) were *H pylori* positive and 112 were negative. Two hundred and four (98%) of 208 *H pylori* infected patients were CagA positive. One hundred and thirty three (42%) of 320 patients had glandular atrophy in the gastric corpus.

Endoscopic findings for erosive oesophagitis

A total of 108 patients (34%) were found to have erosive oesophagitis endoscopically, according to the Los Angeles classification of oesophagitis: grade A: 78 (72%); grade B: 23 (21%); grade C: 6 (6%); and grade D: 1 (1%) (table 2). Erosive oesophagitis was significantly more common in *H pylori* negative compared with *H pylori* positive patients (59/112 = 53% *v* 49/208 = 24%; $p<0.05$) (tables 2, 3). GORD symptoms were reported in 83 (77%) of 108 patients with erosive oesophagitis compared with 47 (22%) of 212 without erosive oesophagitis ($p<0.05$).

Effect of *H pylori* infection on pattern of gastric atrophy and erosive oesophagitis

Glandular atrophy was assessed histologically in the gastric antrum and corpus, respectively. Subjects were divided into two groups: one with corpus atrophy and one without corpus atrophy. The latter group included subjects without atrophy and subjects with antrum atrophy only. A total of 133 of 320 (42%) patients had corpus atrophy and all of these patients also had antrum atrophy. *H pylori* positive patients were more likely to have corpus atrophy than *H pylori* negative patients. Of the 208 who were *H pylori* positive, 128 (62%) had corpus

Table 4 Genotype distribution of the interleukin (*IL*)-*1B*, *IL-10*, and tumour necrosis factor α (*TNF-A*) markers among *Helicobacter pylori* infected and non infected subjects

Polymorphism	Total (n = 247)	*H pylori* positive (n = 190)	*H pylori* negative (n = 57)
IL-1B C−511T			
C/C	78 (31%)	62 (33%)	16 (28%)
C/T	128 (52%)	94 (49%)	34 (60%)
T/T	41 (17%)	34 (18%)	7 (12%)
IL-10 T−819C			
T/T	99 (40%)	75 (39%)	24 (42%)
C/T	116 (47%)	89 (47%)	27 (47%)
C/C	32 (13%)	26 (14%)	6 (11%)
TNF-A T−1031C			
T/T	173 (70%)	141 (74%)	32 (56%)
C/T	71 (29%)	49 (26%)	22 (39%)
C/C	3 (1%)	0 (0%)	3 (5%)

atrophy compared with only five of 112 *H pylori* negative patients (4%) ($p<0.001$) (table 3). Among *H pylori* positive patients, the presence of corpus atrophy was protective against erosive oesophagitis. Of the 159 *H pylori* positive patients who had no erosive oesophagitis, 110 (69%) had corpus atrophy. In contrast, of the 49 *H pylori* positive patients with erosive oesophagitis, only 18 (37%) had corpus atrophy ($p<0.05$).

Effect of the *IL-1B−511C/T*, *IL-10−819T/C*, and *TNF-A−1031T/C* polymorphisms on corpus gastric atrophy

Of the 320 patients participating in this study, 247 (190 *H pylori* positive and 57 *H pylori* negative) were approved for genetic studies and their samples were analysed for the *IL-1B*−511C/T, *IL-10*−819T/C, and *TNF-A*−1031T/C polymorphisms. The *IL-1*, *IL-10*, and *TNF-A* markers had no impact on risk of *H pylori* infection, with the different genotypes being equally prevalent among infected and non-infected subjects (table 4). Among *H pylori* positive patients, there were significantly more subjects with corpus gastric atrophy among those homozygous for the proinflammatory allele *IL-1B*−511T compared with those homozygous for the C allele (OR 4.85 (95% CI 1.86–12.6); $p = 0.001$). This suggests that a proinflammatory *IL-1B* profile is associated with an increased risk of gastric atrophy. None of the other polymorphisms had any significant effect on the risk of corpus gastric atrophy (table 5), and there were no interactions with the *IL-1B*−511 genotypes.

IL-1B−511C/T, *IL-10−819T/C*, and *TNF-A−1031T/C* polymorphisms and GORD

Among *H pylori* positive patients, subjects homozygous for the proinflammatory alleles *IL-1B*−511T had a significantly lower risk of erosive oesophagitis (OR 0.05 (95% CI 0.01–0.26); $p<0.001$) (table 6) and GORD symptoms (OR 0.14 (95% CI 0.03–0.64); $p = 0.01$) (table 7) compared with those homozygous for the −511C alleles. Comparing *IL-1B*−511T/T relative to C/C, age, sex, *H pylori*, and atrophy adjusted OR for GORD was 0.13 (95% CI 0.03-0.48). Taking gastric atrophy out of the model, age and *H pylori* adjusted OR for GORD comparing *IL-1B*−511T/T relative to C/C was 0.09 (95% CI 0.25–0.32). The results indicate that *IL-1B*, *H pylori*, and atrophy are independent markers of decreased risk of GORD.

None of the other cytokine gene polymorphisms had any significant correlation with erosive oesophagitis or GORD symptoms and there was no apparent interaction between the different genetic markers.

DISCUSSION

In the present study, we have shown that both endoscopic (erosive oesophagitis) and symptomatic (NERD) manifestations of GORD are significantly more common among *H pylori* negative subjects compared with those who are *H pylori* positive. We have also shown that subjects with evidence of *H pylori* induced gastric atrophy (documented by histological assessment) are protected from both erosive oesophagitis and NERD. The most novel finding in our study is the demonstration that both gastric atrophy and the associated

Table 5 Age adjusted odds ratios (ORs) and 95% confidence intervals (95% CI) for histological atrophy according to the interleukin (*IL*)-*1B*, *IL-10*, and tumour necrosis factor α (*TNF-A*) genotypes in the *Helicobacter pylori* positive group

	H pylori positive			
Polymorphism	Case/control	OR	p Value	95% CI
IL-1B C−511T				
C/C	26/36	1	Referent	
C/T	60/34	2.42	0.009	1.25–4.68
T/T	26/8	4.85	0.001	1.86–12.6
IL-10 T−819C				
T/T	44/31	1	Referent	
C/T	48/41	0.83	0.55	0.45–1.54
C/C	20/6	2.37	0.10	0.85–6.59
TNF-A T−1031C				
T/T	80/61	1	Referent	
C/T	32/17	1.44	0.29	0.73–2.83
C/C	0/0	–	–	–

Table 6 Age adjusted odds ratios (ORs) and 95% confidence intervals (95% CI) of gastro-oesophageal reflux disease symptoms according to the interleukin (*IL*)-*1B*, *IL-10*, and tumour necrosis factor α (*TNF-A*) genotypes in *Helicobacter pylori* positive and negative groups

	H pylori negative				*H pylori* positive			
Polymorphism	**Case/control**	**OR**	**p Value**	**95% CI**	**Case/control**	**OR**	**p Value**	**95% CI**
IL-1B C−511T								
C/C	5/11	1	Referent		29/33	1	Referent	
C/T	16/18	1.12	0.85	0.34–3.73	48/46	2.03	0.04	1.02–4.04
T/T	1/6	0.14	0.10	0.01–1.43	2/32	0.14	0.01	0.03–0.64
IL-10 T−819C								
T/T	13/11	1	Referent		37/38	1	Referent	
C/T	9/18	1.54	0.28	0.17–1.67	34/55	1.22	0.55	0.63–2.37
C/C	0/6	0.27	0.19	0.04–1.85	8/18	1.50	0.40	0.59–3.82
TNF-A T−1031C								
T/T	13/19	1	Referent		62/79	1	Referent	
C/T	8/14	0.59	0.40	0.17–2.03	17/32	1.13	0.72	0.57–2.25
C/C	1/2	0.35	0.43	0.03–4.61	0/0	–	–	–

protection from GORD complications are mediated by an increased prevalence of the proinflammatory *IL-1B* genetic profile. The work presented here complements the recent report by Queiroz *et al* who showed that the *IL-1B*−511/−31 proinflammatory alleles protect against erosive oesophagitis.[29] We have further expanded these findings by showing that the link between these proinflammatory *IL-1B* polymorphisms and protection from GORD is mediated through induction of gastric atrophy. This is the first demonstration of a direct link between *H pylori* infection, host genetic makeup, gastric atrophy, and protection from GORD and its complications. The findings are likely to have a significant impact on the understanding of the pathogenesis of GORD and the role of *H pylori*.

The most likely explanation for the protective role of these *IL-1B* genotypes against erosive oesophagitis in *H pylori* infected subjects is their association with increased production of IL-1β. Apart from being a very potent proinflammatory cytokine that initiates and amplifies the host's immune response against *H pylori* infection, it is also a powerful acid inhibitor.[24] Thus high production of this cytokine in the gastric mucosa of infected subjects is associated with functional inhibition of gastric acid secretion, which is followed in time by permanent loss of acid secretory capacity through the destructive effects of chronic inflammation on parietal cells. Reduction in gastric acidity is the logical explanation for protection against GORD, a primarily acid related condition. There is evidence that the proinflammatory *IL-1B* genotypes are associated with hypochlorhydria in both Caucasian and Japanese populations.[16 19] Furuta and colleagues[19] reported that Japanese subjects with the *IL-1B*−511 T/T genotype had a median gastric juice pH of 6.5 (range 4.2–7.3) (that is, almost neutral). The same genotype was associated with the highest atrophy scores and lowest pepsinogen I/II ratios. Thus it seems very likely that the reported protective effect of these same *IL-1B* genotypes in this study is also mediated through lower acid secretion due to corpus atrophy.

It is now well established that the damaging effects of the host proinflammatory polymorphisms on the gastric mucosa are significantly increased in the presence of virulent strains of *H pylori*. Figueiredo *et al* have shown that for each combination of bacterial/host genotype, the odds of having gastric carcinoma were greatest in those with both bacterial and host high risk genotypes.[17 18 27] The high risk combination is likely to not only increase the risk of gastric atrophy and cancer but also reduce the risk of developing GORD and its complications.

H pylori infection has divergent clinical outcomes that are in some instances mutually exclusive. Thus patients who develop duodenal ulcer disease are protected from gastric cancer.[36] Although both conditions are associated with *H pylori*, the pathophysiological consequences of the infection are diametrically opposite. Duodenal ulcers are associated with an antrum predominant pattern of gastritis, low prevalence of gastric atrophy, and very high acid secretion. In contrast, gastric cancer patients develop severe pan- or corpus predominant gastritis, multifocal atrophic gastritis,

Table 7 Sex and age adjusted odds ratios (ORs) and 95% confidence intervals (95% CI) of erosive oesophagitis according to the interleukin (*IL*)-*1B*, *IL-10*, and tumour necrosis factor α (*TNF-A*) genotypes in *Helicobacter pylori* positive and negative groups

	H pylori negative				*H pylori* positive			
Polymorphism	**Case/control**	**OR**	**p Value**	**95% CI**	**Case/control**	**OR**	**p Value**	**95% CI**
IL-1B C−511T								
C/C	9/7	1	Referent		18/44	1	Referent	
C/T	20/14	1.94	0.30	0.55–6.81	42/52	1.24	0.52	0.64–2.40
T/T	1/6	0.35	0.39	0.03–3.84	2/32	0.05	<0.001	0.01–0.26
IL-10 T−819C								
T/T	15/9	1	Referent		22/53	1	Referent	
C/T	13/14	0.42	0.13	0.13–1.31	30/59	0.63	0.14	0.34–1.17
C/C	2/4	–*	–*	–	10/16	0.44	0.10	0.17–1.16
TNF-A T−1031C								
T/T	19/13	1	Referent		45/96	1	Referent	
C/T	10/12	0.78	0.70	0.22–2.75	17/32	0.70	0.25	0.34–1.32
C/C	1/2	0.68	0.77	0.05–8.90	0/0	–	–	–

*The number of the case is 0. The frequency of erosive gastritis between TT and CC was compared by Fisher's exact test (p=0.02)

and hypo or achlorhydria. Thus it is apparent that an outcome characterised by low acid secretion protects from acid related disorders such as duodenal ulcers. This pattern also protects from GORD, a condition in which the lower oesophagus has increased exposure to gastric acid. There have been several reports in the literature indicating that infection with virulent strains of *H pylori* (for example, *cagA*$^+$ strains) protects from the more severe outcomes of GORD, such as Barrett's oesophagitis, oesophageal strictures, dysplasia, and oesophageal adenocarcinoma.[4 5 8] It has been speculated that the higher risk of developing gastric atrophy and its associated reduction in gastric acid secretion by *cagA*$^+$ strains explain why these patients are protected from GORD and its complications. It is now apparent that a major factor in this sequence of events is the host genetic constitution. In our population (and in common with most Asian populations), all subjects were infected with *cagA*$^+$ strains yet there was still a divergence of outcomes in relation to GORD. By examining the host genetic markers relating to IL-1β, we could clearly demonstrate that the proinflammatory acid inhibiting and atrophy inducing genotypes were crucial for development of the protective phenotype. Another important observation in our study was the high prevalence of gastric atrophy. Based on histological assessment, 42% of the examined subjects had evidence of atrophy in the corpus as well as in the antrum. The findings in this study confirm that progression of gastric atrophy may protect from erosive oesophagitis in *H pylori* positive individuals.

Demonstration that patients with a proinflammatory *IL-1B* genetic makeup have increased *H pylori* induced atrophy and a reduced risk of GORD must sound a cautionary note about the potential consequences of eradicating the infection in such subjects. It is entirely possible that eradication of the infection in those with a proinflammatory profile would allow the corpus mucosa to recover sufficiently so that acid secretion returns. In such circumstances, one might expect an increased incidence of GORD in those who have a predisposition to developing reflux. While this may be adequately handled with acid suppressive therapy, it is essential that health care providers are aware of the potential for the increased incidence of GORD and its possible complications. This point is particularly relevant if one contemplates eradication of the infection on a large population wide scale, particularly in areas of high gastric cancer incidence. There is currently no convincing evidence that *H pylori* eradication either increases the incidence of de novo GORD or worsens pre-existing GORD.[37] However, most of these studies have come from Western countries in which gastric atrophy and hypochlorhydria are much less common than in countries such as Japan, China, and Korea. In these Asian countries, the outcome might be different and it is essential that this be taken into consideration when planning a global eradication strategy. It is apparent and indeed very desirable that prospective studies addressing these issues should be undertaken in Asian countries.

In conclusion, we have shown that proinflammatory genotypes of the *IL-1B* gene increase the risk of gastric atrophy and reduce the risk of erosive oesophagitis and GORD symptoms, most probably through induction of gastric atrophy and gastric acid inhibition. These findings are very relevant to understanding the pathogenesis of GORD and the role of *H pylori* infection. They are also relevant to the ongoing debate about the long term consequences of *H pylori* eradication in the context of gastric cancer prevention.

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REFERENCES

1 **DeVault KR**, Castell DO. Updated guidelines for the diagnosis and treatment of gastro-oesophageal reflux disease. The Practice Parameters Committee of the American College of Gastroenterology. *Am J Gastroenterol* 1999;**94**:1434–42.
2 **Chiba N**, De Gara CJ, Wilkinson JM, *et al*. Speed of healing and symptom relief in grade II to IV gastrooesophageal reflux disease: a meta-analysis. *Gastroenterology* 1997;**112**:1798–810.
3 **Werdmuller BF**, Loffeld RJ. Helicobacter pylori infection has no role in the pathogenesis of reflux oesophagitis. *Dig Dis Sci* 1997;**42**:103–5.
4 **Chow WH**, Blaser MJ, Blot WJ, *et al*. An inverse relation between cagA+ strains of Helicobacter pylori infection and risk of oesophageal and gastric cardia adenocarcinoma. *Cancer Res* 1998;**58**:588–90.
5 **Vicari JJ**, Peek RM, Falk GW, *et al*. The seroprevalence of cagA-positive Helicobacter pylori strains in the spectrum of gastro-oesophageal reflux disease. *Gastroenterology* 1998;**115**:50–7.
6 **Varanasi RV**, Fantry GT, Wilson KT. Decreased prevalence of Helicobacter pylori infection in gastrooesophageal reflux disease. *Helicobacter* 1998;**3**:188–94.
7 **Wu JC**, Sung JJ, Ng EK, *et al*. Prevalence and distribution of Helicobacter pylori in gastro-oesophageal reflux disease: a study from the East. *Am J Gastroenterol* 1999;**94**:1790–4.
8 **Vaezi MF**, Falk GW, Peek RM, *et al*. CagA-positive strains of Helicobacter pylori may protect against Barrett's oesophagus. *Am J Gastroenterol* 2000;**95**:2206–11.
9 **Hackelsberger A**, Schultze V, Gunther T, *et al*. The prevalence of Helicobacter pylori gastritis in patients with reflux ooesophagitis: a case-control study. *Eur J Gastroenterol Hepatol*. 1998;10: 465–8?).
10 **Malfertheiner P**. Helicobacter pylori eradication does not exacerbate gastro-ooesophageal reflux disease. *Gut* 2004;**53**:312–13.
11 **Sipponen P**, Hyvarinen H. Role of Helicobacter pylori in the pathogenesis of gastritis, peptic ulcer and gastric cancer. *Scand J Gastroenterol* 1993;**196**:S3–6.
12 **El-Omar EM**, Oien K, El-Nujumi A, *et al*. Helicobacter pylori infection and chronic gastric acid hyposecretion. *Gastroenterology* 1997;**113**:15–24.
13 **Meining A**, Stolte M, Hatz R, *et al*. Differing degree and distribution of gastritis in Helicobacter pylori-associated diseases. *Virchows Arch* 1997;**431**:11–15.
14 **Graham DY**. Helicobacter pylori infection in the pathogenesis of duodenal ulcer and gastric cancer: a model. *Gastroenterology* 1997;**113**:1983–91.
15 **El-Omar EM**, Chow WH, Rabkin CS. Gastric cancer and H. pylori: Host genetics open the way, *Gastroenterology* 2001;**121**:1002–4.
16 **El-Omar EM**, Carrington M, Chow WH, *et al*. Interleukin-1 polymorphisms associated with increased risk of gastric cancer. *Nature* 2000;**404**:398–402.
17 **Figueiredo C**, Machado JC, Pharoah P, *et al*. Helicobacter pylori and interleukin 1 genotyping: an opportunity to identify high-risk individuals for gastric carcinoma. *J Natl Cancer Inst* 2002;**94**:1680–7.
18 **Machado JC**, Figueiredo C, Canedo P, *et al*. A proinflammatory genetic profile increases the risk for chronic atrophic gastritis and gastric carcinoma. *Gastroenterology* 2003;**125**:364–71.
19 **Furuta T**, El-Omar EM, Xiao F, *et al*. Interleukin 1 polymorphisms increase risk of hypochlorhydria and atrophic gastritis and reduce risk of duodenal ulcer recurrence in Japan. *Gastroenterology* 2002;**123**:92–105.
20 **Yamaoka Y**, Kita M, Kodama T, *et al*. Induction of various cytokines and development of severe mucosal inflammation by cagA gene positive Helicobacter pylori strains. *Gut* 1997;**41**:442–51.
21 **Wallace JL**, Cucala M, Mugridge K, *et al*. Secretagogue-specific effects of interleukin-1 on gastric acid secretion. *Am J Physiol* 1991;**261**:G559–64.
22 **Kondo S**, Shinomura Y, Kanayama S, *et al*. Interleukin-1 beta inhibits gastric histamine secretion and synthesis in the rat. *Am J Physiol* 1994;**267**:G966–71.
23 **Beales IL**, Calam J. Interleukin 1 beta and tumour necrosis factor alpha inhibit acid secretion in cultured rabbit parietal cells by multiple pathways. *Gut* 1998;**42**:227–34.
24 **El-Omar EM**. The importance of interleukin 1beta in Helicobacter pylori associated disease. *Gut* 2001;**48**:743–7.
25 **Hamajima N**, Matsuo K, Saito T, *et al*. Interleukin 1 polymorphisms, lifestyle factors, and Helicobacter pylori infection. *Jpn J Cancer Res* 2001;**92**:383–9.
26 **Hwang IR**, Kodama T, Kikuchi S, *et al*. Effect of interleukin 1 polymorphisms on gastric mucosal interleukin 1beta production in Helicobacter pylori infection. *Gastroenterology* 2002;**123**:1793–803.
27 **Rad R**, Dossumbekova A, Neu B, *et al*. Cytokine gene polymorphisms influence mucosal cytokine expression, gastric inflammation, and host specific colonisation during Helicobacter pylori infection. *Gut* 2004;**53**:1082–9.
28 **El-Omar EM**, Rabkin CS, Gammon MD, *et al*. Increased risk of noncardia gastric cancer associated with proinflammatory cytokine gene polymorphisms. *Gastroenterology* 2003;**124**:1193–201.

29 **Queiroz DM**, Guerra JB, Rocha GA, *et al*. IL1B and IL1RN polymorphic genes and Helicobacter pylori cagA strains decrease the risk of reflux oesophagitis. *Gastroenterology* 2004;**127**:73–9.
30 **Carlsson R**, Dent J, Bolling-Sternevald E, *et al*. The usefulness of a structured questionnaire in the assessment of symptomatic gastrooesophageal reflux disease. *Scand J Gastroenterol* 1998;**33**:1023–9.
31 **Armstrong D**, Bennett JR, Blum AL, *et al*. The endoscopic assessment of oesophagitis: a progress report on observer agreement. *Gastroenterology* 1996;**111**:85–92.
32 **Dixon MF**, Genta RM, Yardley JH, *et al*. Classification and grading of gastritis. The updated Sydney system. *Am J Surg Pathol* 1996;**20**:1161–81.
33 **Kusugami K**, Ando T, Ohsuga M, *et al*. Mucosal chemokine activity in Helicobacter pylori infection. *J Clin Gastroenterol* 1997;**25**:S203–10.
34 **Ohsuga M**, Kusugami K, Ina K, *et al*. Comparison between in vivo and in vitro chemokine production in Helicobacter pylori infection. *Aliment Pharmacol Ther* 2000;**14**:S205–15.
35 **Hamajima N**, Saito T, Matsuo K, *et al*. Polymerase chain reaction with confronting two-pair primers for polymorphism genotyping. *Jpn J Cancer Res* 2000;**91**:865–8.
36 **Hansson LE**, Nyren O, Hsing AW, *et al*. The risk of stomach cancer in patients with gastric or duodenal ulcer disease. *N Engl J Med* 1996;**335**:242–9.
37 **Raghunath AS**, Hungin AP, Wooff D, *et al*. Systematic review: the effect of Helicobacter pylori and its eradication on gastro-oesophageal reflux disease in patients with duodenal ulcers or reflux oesophagitis. *Aliment Pharmacol Ther* 2004;**20**:733–44.

EDITOR'S QUIZ: GI SNAPSHOT

An unusual cause of pancreatitis

Robin Spiller, *Editor*

Clinical presentation

A 67 year old woman had experienced more than 10 episodes of acute pancreatitis in five years. She was referred to our hospital for recurrent acute pancreatitis despite prior endoscopic papillotomy and laparoscopic cholecystectomy. She denied consumptions of alcohol or herbal medications. No other family member had a similar illness.

On physical examination neither jaundice nor abdominal tenderness was noted. There was no palpable abdominal mass. Laboratory values were as follows: amylase 100 U/l (<220), lipase 36 U/l (<190), triglycerides 120 mg/dl (<200), and calcium 2.37 mmol/l (2.02–2.6). Liver function tests and tumour markers were within normal limits.

Magnetic resonance cholangiopancreatography was performed (fig 1) and the findings were confirmed by endoscopic retrograde cholangiopancreatography (ERCP). At ERCP, the major papilla was not widely open but the minor papilla was prominent. The major papilla was first cannulated and opacified and the minor papilla was subsequently cannulated and opacified (fig 2).

Question

What accounts for her recurrent attacks of pancreatitis?

See page 171 for answer

This case is submitted by:

Supplementary figure 1 (Endoscopic view of the major papilla. Note the lack of profuse mucus at the orifice in this case) and supplementary figure 2 (endoscopic view of the minor papilla), can be viewed on the Gut website at http://www.gutjnl.com/supplemental.

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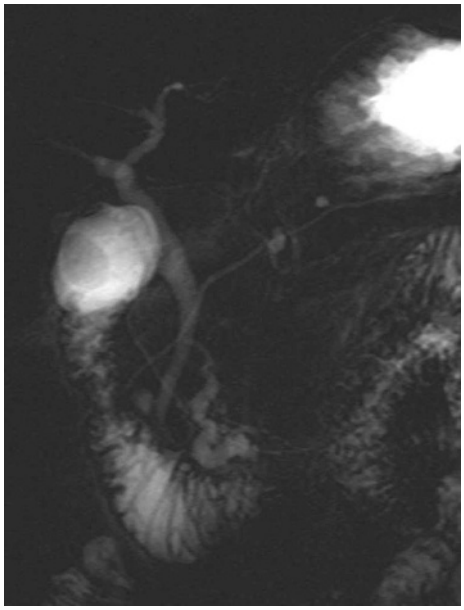

Figure 1 Magnetic resonance cholangiopancreatography of a 67 year old woman.

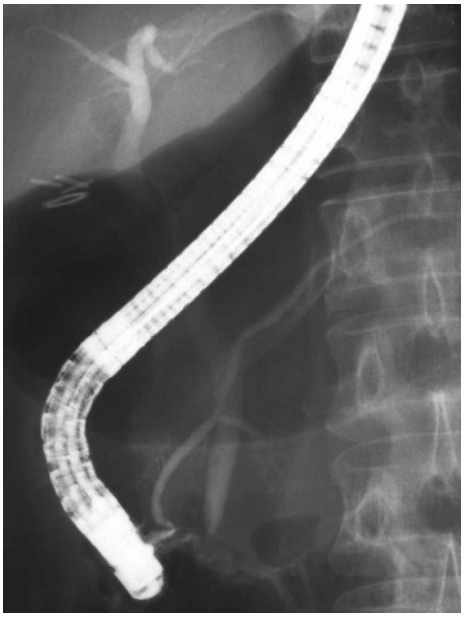

Figure 2 Endoscopic retrograde cholangiopancreatography with sequential opacification at the major papilla and then at the minor papilla.