

Mast cells and IgE-containing cells in gastric mucosa of *Helicobacter pylori* infected and non-infected patients with chronic urticaria

M Liutu,^{†*} K Kalimo,[†] H Kalimo,[‡] J Uksila,[§] R Leino[¶]

[†]Department of Dermatology, [‡]Department of Pathology, [§]Department of Medical Microbiology, and [¶]Department of Internal Medicine, University of Turku, FIN-20520 Turku, Finland. *Corresponding author, Department of Dermatology, University of Turku, PL 52, 20520 Turku, Finland, tel. +358-2-3132 600; fax +358-2-3131 610; E-mail: mervi.liutu@tyks.fi

ABSTRACT

Background Several studies have indicated that antibiotic therapy aimed at eradication of *Helicobacter pylori* has effects on symptoms of chronic urticaria (CU) patients. However, the possible connections and pathomechanism by which *H. pylori* might be linked to CU have remained largely unknown. The IgE-mediated pathway might be a possible link between *H. pylori* infection and CU. We therefore clarified the role of *H. pylori* as an inducer of IgE response.

Materials and methods Gastroscopy was performed and mucosal biopsy specimens were taken to evaluate the histology, as well as the presence of *H. pylori* bacteria, mast cells and IgE-containing cells in the antral mucosa, in 21 CU patients. Controls ($n = 48$) included 19 patients with lichen planus, nine patients with atopic dermatitis and 20 patients with no skin or allergic disease.

Results The mean densities of IgE-containing cells were significantly higher in *H. pylori*-infected patients and in patients with skin disease compared to non-*H. pylori*-infected patients with no skin or allergic disease. No significant difference was found in the number of IgE-containing cells between *H. pylori*-infected and non-infected patients with CU. There was no significant difference in the mean densities of mast cells in the different patient groups.

Conclusions Our findings suggest that *H. pylori* gastritis leads to increased IgE production. However, we could not show a significant difference in IgE staining between *H. pylori*-infected and non-infected patients with CU.

Key words: chronic urticaria, *Helicobacter pylori*, IgE, mast cell

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Introduction

Several studies have indicated that antibiotic therapy aimed at eradication of *Helicobacter pylori* has effects on symptoms of chronic urticaria (CU) patients.^{1–4} However, the possible connection and pathomechanism by which *H. pylori* might be linked to extraintestinal disorders, such as CU, have remained largely unknown. Chronic infection with *H. pylori* normally causes production of specific IgM, IgG and IgA class antibodies. More recently, increases in non-specific IgE levels as well as in production of specific IgE antibodies against *H. pylori* have also been detected, suggesting that IgE might contribute to extraintestinal symptoms associated with *H. pylori*.^{5–8}

During *H. pylori* gastritis, a release of histamine from the mast cells has been described.^{9,10} Interest is being shown in the ability of *H. pylori* to release histamine through an IgE-mediated pathway from the mast cells. For example, in their *in vitro* study, Gala-Ortiz *et al.*¹¹ showed specific binding of IgE from a single CU patient to an extract of *H. pylori*, suggesting an IgE-mediated relationship between *H. pylori* gastritis and CU.

In order to clarify the role of IgE antibody production in *H. pylori* infection in general, and specifically the possible association of CU with *H. pylori*, we have studied the accumulation of mast cells and IgE-binding cells in the antral mucosa of *H. pylori*-infected and non-infected individuals. In addition, the activity of gastritis was evaluated. Patients with CU were

compared with patients with lichen planus (LP) and atopic dermatitis (AD), and also with patients with no skin or allergic disease.

Materials and methods

Patients

The 69 patients participating in the study were outpatients from the Department of Dermatology and Internal Medicine of the University Central Hospital of Turku, Finland. Among the patients 21 had CU (14 male, seven female, mean age 47 years), 19 had LP (five male, 14 female, mean age 57 years), nine had AD (two male, seven female, mean age 39 years) and 20 had no history of skin or allergic disease (nine male, 11 female, mean age 53 years).

Upper gastrointestinal endoscopy, biopsy samples and identification of *H. pylori*

Upper gastrointestinal endoscopy was performed in all patients using an Olympus GIF-XQ 10 endoscope. Biopsies from the gastric antrum were taken to determine the numbers of mast cells and IgE-containing cells and to identify *H. pylori* infection. The criterion for *H. pylori* infection was the presence of bacteria in gastric mucosa studied by histology and elevated serum *H. pylori* IgG antibodies. Patients who were negative both in histology and in serology were regarded as *H. pylori* non-infected. The activity of gastritis was evaluated by the Sydney system.¹²

Histopathology and morphometry

The biopsy specimens were routinely fixed in 4% phosphate-buffered formaldehyde and embedded in paraffin. *H. pylori* infection was assessed in sections stained with haematoxylin and eosin and toluidine blue. For the identification of mast cells, the sections were stained with the prolonged toluidine blue method. For immunohistochemical demonstration of IgE-positive cells, consecutive sections were first treated with H₂O₂ and normal serum to block non-specific peroxidase activity and non-specific protein binding. Polyclonal rabbit antihuman IgE (Dakopatts, Glostrup, Denmark) was used as the antiserum at a dilution of 1:5000 with incubation at 4 °C overnight. The bound antibodies were visualized by the avidin–biotin–peroxidase method (Vector Laboratories, Burlingame, CA, USA) using diaminobenzidine as the chromogen.

The densities of the toluidine-blue-stained mast cells and IgE-containing cells were quantified light microscopically in consecutive sections of antral mucosa. The cells located in the lamina propria were counted at a magnification of 312.5 (objective, ×25; ocular, ×12.5) using an ocular grid composed of 100 small squares. The area of the lamina propria was measured by point counting, i.e. by counting the number of intercepts falling on the lamina propria. Gastric glands and sur-

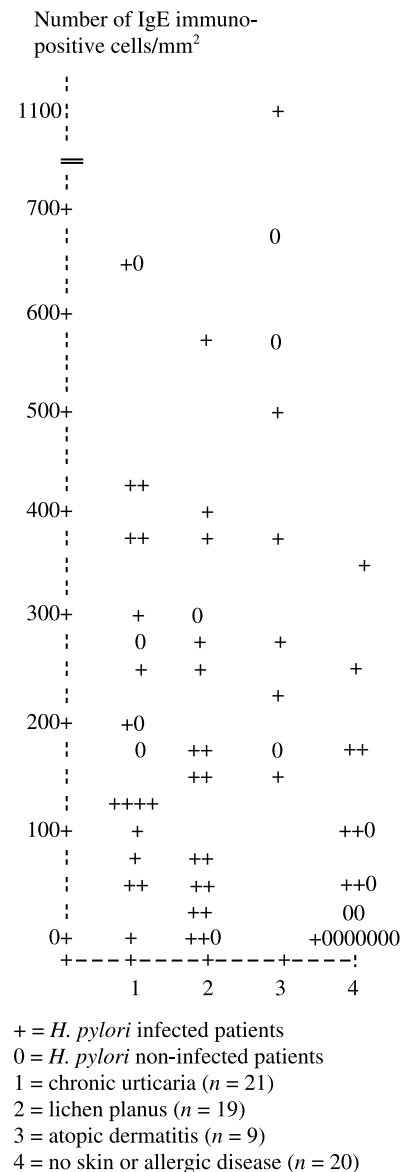


fig. 1 Number of IgE immuno-positive cells/mm² for each patient.

face epithelium were thus excluded. Cell countings were performed in five randomly selected areas above the muscularis mucosae. Mean values for each patient were expressed as the number of cells per square millimetre (fig. 1). For statistical analyses, the Wilcoxon rank-sum test was used.

Results

Toluidine-blue-stained and IgE-containing cells in antral mucosa

The mean densities of antral toluidine-blue-stained mast cells and IgE-containing cells, as well as the activity of gastric inflammation in different *H. pylori*-infected and non-infected

groups, are shown in Table 1. The mean densities of mast cells did not statistically differ between different patient groups. Remarkably, the number of mast cells as such in patients with no skin or allergic diseases did not differ from those with skin diseases. The number of both mast cells and IgE-containing cells was highest in patients with AD. In *H. pylori*-infected patients with no skin or allergic disease, the density of IgE-containing cells was significantly higher than in non-infected patients ($P = 0.0186$), but the density of IgE-containing cells remained lower than in the three groups with skin disease. There was no significant difference in the density of IgE-containing cells between *H. pylori*-infected and non-infected patients with skin disease. The mean values of IgE-containing cells in individual patients are shown in fig. 1.

Discussion

Gastric mucosa responds with inflammation to *H. pylori* infection. While numerous reports have focused on specific IgG, IgA and IgM isotype anti-*H. pylori* antibodies, *H. pylori*-specific IgE production has gained less attention. However, some studies indicate that specific IgE might also be associated with the pathophysiology of *H. pylori* infection. Aceti *et al.*⁵ found *H. pylori*-specific IgE bound to blood basophils in 84% of patients with *H. pylori* infection and serum-specific IgE in 69%. Studenikin *et al.*⁶ showed, in their study, a considerable increase of total serum IgE titres in children with *H. pylori* infection compared with the controls. Recently, Berczi *et al.*⁸ reported a significant accumulation of IgE-positive cells in the gastric mucosa in *H. pylori*-positive patients, suggesting a role of IgE-mediated immune response in the development of *H. pylori*-associated gastritis.

The increased numbers of IgE-containing cells in the antral mucosa of CU patients supported our earlier study¹³ in which we described serum IgE levels as being slightly elevated in several CU patients, even when they did not have a history of atopy. However, there was no significant difference in the number of IgE-containing cells between the *H. pylori*-infected and non-infected CU patients. Patients with AD had higher mean densities of IgE-containing cells than the other patients, which is an expected result for IgE-mediated disease.¹⁴ Interestingly, IgE-containing cells were also found in both *H. pylori*-infected and non-infected patients with LP, which is not regarded as an IgE-mediated disease. Similarly, we demonstrated that in patients with no skin or allergic disease, the mean density of antral IgE-containing cells was significantly higher in *H. pylori*-infected patients than in *H. pylori* negative ones. This is in accordance with earlier studies, which suggest that *H. pylori* infection is associated with an induction of IgE response. The fact that the number of IgE-positive cells was elevated even in *H. pylori*-negative patients in all patient groups with skin disease demonstrates that this increase is not solely due to the *H. pylori* infection, but that other factors, in gastric antigens and microbes, are

Table 1 Mean densities of mast cells and IgE-containing cells, and the activity of inflammation in antral mucosa of different *H. pylori*-positive and *H. pylori*-negative patient groups

Patients (n)	Mean density of gastric inflammation												P
	Mast cells		IgE-containing cells		Severe		Moderate		Mild		Normal		
	HP pos (n)	HP neg (n)	HP pos (n)	HP neg (n)	HP pos (n)	HP neg (n)	HP pos (n)	HP neg (n)	HP pos (n)	HP neg (n)	HP pos (n)	HP neg (n)	
CU (21)	252 (16)	366 (4)	219 (17)	329 (4)	5	0	6	0	6	1	0	3	
LP (19)	257 (17)	281 (2)	165 (17)	147 (2)	5	0	6	0	6	0	0	2	
AD (9)	398 (5)	528 (2)	408 (6)	495 (3)	0	0	3	0	3	0	0	3	
NO SAD (20)	283 (4)	329 (4)	131 (9)	17 (11)	2	1	6	3	1	1	0	6	

HP, *H. pylori*; NO SAD, no skin or allergic disease; n, number of patients.

contributing to the increase. Actually, the number of IgE-containing cells was higher in *H. pylori*-negative CU and AD patients, indicating the complex regulation of IgE synthesis and deposition of IgE to gastric mucosa in such clinical conditions as CU and AD.

Our results suggest that, in patients with manifest skin disease, the local immune response in the gastrointestinal tract may be skewed towards the Th2-type response as reflected by the increased staining of IgE of gastric mucosal cells. Because also dermatological patients with no evidence of *H. pylori* infection had increased numbers of IgE-containing cells, it is probable that other intestinal antigens, under the same conditions, could initiate this process. Our results agree with the previous results that IgE production is induced in *H. pylori*-associated gastritis. However, there may be other more prominent mechanisms, associated with CU, that result in generation of IgE production, and therefore the triggering effect of *H. pylori* on IgE synthesis in patients with CU is not as clearly detectable as in control subjects with no skin or allergic disease.

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