ORIGINAL ARTICLE

H. pylori positivity and various pathological, endoscopic and clinical features correlated with each other

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Abstract

Objective: To investigate the relationship between dyspepsia symptom scores and endoscopic appearances, and histopathological findings and helicobacter pylori positivity in patients having dyspepsia symptom.

Methods: The study was conducted at the gastroenterology outpatient clinic of Adnan Menderes University, School of Medicine, Aydin, Turkey from April 2012 to July 2012 and comprised patients between 18-65 years of age who were admitted with dyspepsia. Glasgow dyspepsia severity scoring was done with questions posed orally to the patients. In histopathological evaluation of biopsy specimens according to Sydney criteria, chronic inflammation, activity, atrophy, intestinal metaplasia and helicobacter pylori parameters were used. Total number of eosinophils and number of mast cells were recorded.

Results: Of the 60 patients with dyspepsia, 38(63.3%) were female and 22(36.7%) were male. The degree of activation and severity of inflammation increased significantly with increasing helicobacter pylori positivity(r=0.459'p<0.0001; r=0.475'p<0.0001). A significant relationship was found between inflammation, activation and the number of mast cells (p<0.05).There was no relationship between helicobacter pylori intensity and the eosinophil count (r=0.171; p=0.093). There was also a statistically significant correlation between severity of inflammation and activation and the number of eosinophils (r=0.313;p=0.002;r=0.245;p=0.016).

Conclusion: Mast cell density was seen to have a role in the inflammatory processes of helicobacter pylori infection.

Keywords: Helicobacter pylori, Dyspepsia, Mast cells, Eosinophils. (JPMA 65: 1305; 2015)

Introduction

Dyspepsia is a continuous or recurrent pain, discomfort or inconvenient feeling around the upper-middle region of the abdomen. Functional dyspepsia (FD) is defined as the presence of persistent or recurrent dyspepsia symptoms localised in the epigastric region without detecting any underlying organic, metabolic and systemic cause in extensive examinations. Its ethyopathogenesis and pathophysiology have not been sufficiently elucidated yet. In patients without distinctive changes of gastric mucosa, dyspepsia is considered to be secondary to a functional disorder rather than a structural disorder, and this is called FD.[1,2] Although it is suggested that diverse factors, such as increased gastric acid secretion, Helicobacter pylori (Hp) infection, gastroduodenal dysmotility, visceral hypersensitivity and psychological factors, may explain physiopathology, the exact mechanism is not clear yet, and the data of relevant studies are controversial.[3-8]

The current study was planned to investigate the relationship among dyspepsia symptom scores, endoscopic appearances, histopathological findings and Hp positivity in patients with dyspepsia.

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Materials and Methods

The study was conducted at the gastroenterology outpatient clinic of Adnan Menderes University, School of Medicine, Aydin, Turkey from April 2012 to July 2012 and comprised patients between 18-65 years of age who were admitted with dyspepsia. Glasgow dyspepsia severity scoring[9] was done with questions posed orally to the patients. Upper gastrointestinal (GI) endoscopic examination was performed (Pentax EG 2940-type video gastroscope). Patients were categorised as normal, antral gastritis, and pangastritis according to the endoscopic appearances, and two biopsies were obtained from the antrum which were examined histopathologically by the same pathologist who was blind to patients' endoscopic diagnosis and symptom scores.

After fixing in 10% neutral-buffered formalin, the biopsy samples were followed routinely. Then cuts in 4μm thickness were obtained from the samples that were embedded into paraffin blocks. The cuts were then applied haematoxylin and eosin (H&E), Giemsa and Alcian Blue dyes and examined under a light microscope. According to the Sydney criteria, chronic inflammation,

activity, atrophy, intestinal metaplasia and Hp parameters were used for assessment.[10] Additionally, total eosinophil count was recorded by examining the most intense areas (hot-spot) of the H&E-stained preparations under 10 high-power field (10HPF, X40).[11] In Toluidine Blue-stained sections, the most intense areas of mast cells were examined under HPF, and under 10HPFtotal number of mast cells at lamina propria was recorded.

Data was analysed using SPSS 13. Compatibility of continuous variables to the normal distribution was analysed with Kolmogorov-Smirnov test and descriptive statistics were shown as median (25%-75%) since none of the variables showed normal distribution. Kruskal-Wallis test was used for comparisons between groups. In statistical analysis, 2 patients with no inflammation were combined with "mild" and 1 patient with severe activation was combined with "moderate"). Kendall's correlation coefficient was used to examine the relationships between variables. P<0.05 was considered statistically significant.

Results

Of the 60 patients, 38(63.3%) were women and 22(36.7%) were men, with an overall mean age of 39±13 years.

Eosinophil count and the number of mast cells were significantly higher (p<0.05) for patients who were evaluated as antral gastritis and pangastritis than the ones with normal endoscopy results.

Dyspepsia score was higher in normal group compared to those with antral gastritis and pangastritis, but the difference was statistically insignificant (p>0.05).

Only 7(11.6%) patients were Hp-negative, whereas atrophy was detected in 7(11.6%) patients, and intestinal metaplasia was detected in 7(11.6%) patients (Table-1).

The number of mast cells, eosinophil count and dyspepsia score medians were not significantly different between the Hp groups (p=0.177; p=0.380; p=0.242) (Table-2).

The number of mast cells was significantly different with respect to activation and inflammation severity (p=0.001; p<0.0001). Also, the eosinophil count was significantly different with respect to the activation and the severity of inflammation ((p=0.029; p=0.012) (Table-3).

The severity of activation and inflammation increased

Table-3: Comparison of the number of mast cells and eosinophil countwith respect to inflammation and activation groups.

	Mast Cells Median(25%-75%)	Eosinophil Median(25%-75%)
Inflammation		
Mild	9.0(5.0-10.0)	45.0(21.0-60.0)
Moderate	15.0(12.0-25.0)	6.03(39.0-108.0)
Severe	16.0(12.0-33.8)	103.0(48.8-180.0)
P value	p<0.0001	0.012
Activation		
None	8.0(4.0-15.0)	36.0(20.0-45.0)
Mild	12.0(9.0-19.5)	61.5(39.0-103.5)
Moderate	20.0(13.5-27.5)	57.0(39.5-136.0)
P value	p=0.001	0.029

Table-1: Four-scale values of the parameters used in histopathological staging of the biopsy specimens according to the Sydney system.

	Hp n (%)	Inflammation n (%)	Activation n (%)	Intestinal metaplasia n (%)	Atrophy n (%)
None	7 (11,7)	2 (3,3)	15 (25,0)	53 (88,3)	53 (88,3)
Mild	35 (58,3)	21 (35,0)	32 (53,3)	2 (3,3)	6 (10,0)
Moderate	14 (23,3)	31 (51,7)	12 (20,0)	5 (8,3)	1 (1,7)
Severe	4 (6,7)	6 (10,0)	1 (1,7)	- -	- -
Total	60 (100)	60 (100)	60 (100)	60 (100)	60 (100)

Hp: Helicobacter pylori.

Table-2: Comparison of the number of mast cells, eosinophil count and dyspepsia score with respect to helicobacter pylori (Hp) groups.

	Hp				P value
	None	Mild	Moderate	Severe	
Mast Cells Median (25%-75%)	10.0(5.0-17.0)	12.0(8.0-18.0)	20.0(10.5-30.0)	16.0(8.3-21.5)	0.177
Eosinophils Median (25%-75%)	32.0(20.0-75.0)	48.0(36.0-90.0)	69.0(35.3-120.0)	58.5(39.8-129.0)	0.380
Dyspepsia Score Median (25%-75%)	12.0(11.0-14.0)	11.0(8.0-13.0)	10.0(7.8-12.5)	8.0(7.0-11.3)	0.242

Table-4: Analysis of correlation between variables.

		Dyspepsia Score	Hp	Inflammation	Activation	Mast	Eosinophil
Dyspepsia Score	r	1.000	-0.199	-0.055	0.013	-0.158	-0.119
	p	.	0.058	0.604	0.904	0.093	0.197
	N	60	60	60	60	60	60
Hp	r		1.000	0.475	0.459	0.212	0.171
	p		.	<0.0001	<0.0001	0.041	0.093
	N		60	60	60	60	60
Inflammation	r			1.000	0.672	0.457	0.313
	p			.	<0.0001	<0.0001	0.002
	N			60	60	60	60
Activation	r				1.000	0.396	0.245
	p				.	<0.0001	0.016
	N				60	60	60
Mast	r					1.000	0.523
	p					.	<0.0001
	N					60	60
Eosinophil	r						1.000
	p						.
	N						60

Hp: Helicobacter pylori.

with increasing Hp-positivity, which was statistically significant (r=0.459; p<0.0001; r=0.475; p<0.0001) Hp intensity did not correlate significantly with dyspepsia score(r=-0.199; p=0.058). The number of mast cells showed an increase as Hp intensity increased, and this was statistically significant (r=0,212; p=0.041). But there was no significant relationship with the eosinophil count (r=0.171; p=0.093). There was a significant correlation between the number of mast cells and severity of the inflammation and activation (r=0.457; p<0.0001; r=0.396; p<0.0001). The number of eosinophils was significantly correlated with the severity of inflammation and activation (r=0.313; p=0.002; r=0.245; p=0.016). There was no significant correlation between the degree of inflammation and the dyspepsia score induced by Hp (r=-0.055; p=0.604). Also there was no significant relationship between the degree of activation and the dyspepsia score (r=0.013; p=0.904). No significant relationship was found between the dyspepsia score and the number of mast cells as well as the eosinophil count (r=-0.158; p=0.093; r=-0.119; p=0.197) (Table-4).

Discussion

In our study, Hp positivity was determined as 88% in patients with complaints of dyspepsia. Studies investigating the role of gastric mucosal inflammation in the development of dyspeptic symptoms were mainly associated with prevalence and eradication of Hp. A study conducted in 2003 reported that 80% of women and 86% of men who had gastric complaints were infected with Hp, while the infection rate in asymptomatic men and women was 83%.[12]

In a study comparing endoscopic and histological findings of FD patients with or without Hp infection, gastritis score were significantly higher in patients infected with Hp.[4] We found a significant positive correlation between Hp intensity and the severity of both inflammation and activation (p<0.05). In other words, the severity of both activation and inflammation increased with increasing Hp positivity. This finding is similar to studies showing close relationship between Hp concentration and gastritis scores.[5,6] One study investigated 103 FD patients and 42 control subjects in terms of gastritis, Hp positivity, and activity of the disease. While Hp positivity was detected 62% in the FD group, it was 55% in the control group, with no statistically significant difference between the two groups. It was concluded that FD cannot be associated with Hp gastritis alone.[7] Several studies which have examined the relationship between the histological grade of gastritis and severity of the symptoms of gastritis, reported incompatible results. In a couple of studies no correlation was found between the severity of histological gastritis and severity of symptoms.[7,8] The most appropriate way to evaluate the relationship between Hp and FD is considered to be the assessment of the improvement in symptomatic response and in gastritis after eradication therapy.[1] A meta-analysis of recent studies concluded that Hp eradication therapy may have small but statistically

significant clinical benefit compared to placebo.[13,14] No statistically significant correlation was found between Hp intensity and dyspepsia scores in our cases (p> 0.05). A study reported no correlation between histological grading and dyspepsia scores.[8]

Mast cells (MC) can be found throughout normal tissues to play a pro-inflammatory role. MCs were considered pro-inflammatory cells since they can secrete different mediators and cytokines in the initial phase of inflammation.[15] MCs are responsible for the activation of polymorphonuclear cells as well as mononuclear cells. A study found that the increase in the density of MCs was proportional to the intensity of polymorphonuclear and mononuclear cells. In active Hp-positive gastritis, mucosal MCs are believed to play an active role in the accumulation of the neutrophils and lymphocytes.[16] Our study observed a statistically significant correlation between the intensity of Hp and the number of MCs (p< 0.05), where the number of MCs increased with the intensity of Hp. Moreover, the severity of inflammation and activation showed a significant relationship with the number of MCs (p<0.0001). No correlation was found between the dyspepsia score and the number of MCs (p>0.05). A study demonstrated the increase in the number of MCs among FD patients either with Hp-positive or Hp-negative, but it indicated that such an increase may occur in the presence of symptoms, irrespective of inflammation.[17]

Eosinophils are inflammatory cells that play a role in the pathogenesis of many diseases. The density of eosinophils in the gastrointestinal tract are known to be relatively higher than other tissues.[18] Eosinophils may also have an important role in the pathogenesis of chronic gastritis. It is observed that both serum immunoglobulin A (IgA) and IgG levels increased as part of the systemic humoral response against Hp, and Hp-specific IgA and IgM concentrations in the gastric fluid increased as part of the local humoral response.[19] Furthermore, IgA is an extremely potent stimulus for eosinophilic degranulation.[20] A study investigated Hp-infected gastric mucosa, normal antral gastric mucosa, biopsy samples of Menetrier's disease and non-specific gastritis in terms of eosinophil infiltration and degranulation. It demonstrated more pronounced eosinophilic infiltration and degranulation in Hp-infected gastric mucosa. It also noted that the eosinophil count was significantly associated with the degree of chronic gastritis. As a result, it speculated that the increase in eosinophilic degranulation may be a response to the development of Hpcolonisation.[21] Another study indicated the relationship between Hp intensity and eosinophilic infiltration, and also between eosinophil count and severity of chronic gastritis. Unlike many other studies MCs were reported to play an insignificant role.[22] Similarly, one study reported that while there was a significant increase in the number of eosinophils associated with Hp, no such relation was found in the number of MCs.[23] In our study, there was no significant relationship between the intensity of Hp and the eosinophil count. However, inflammation and activation showed a significant relationship with the number of eosinophils (p < 0.05).

In our study, the small number of Hp-negative patients can be explained by the high prevalence of Hp infection in our country and the small number of the study group. The prevalence of chronic atrophic gastritis is known to increase with age.[24] In our study, the small sample size of patients with intestinal metaplasia and atrophy can be explained by the presence of relatively young patients.

The limitation of our study is the small sample size of patients and the absence of a control group.

Conclusion

Hp infection positivity was seen to be influential in the inflammatory process by increasing mast cell density.

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