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ASSOCIATION BETWEEN *ENTAMOEBA HISTOLYTICA/DISPAR* AND *HELICOBACTER PYLORI* INFECTIONS IN PATIENTS WITH GASTROINTESTINAL COMPLAINTS

By

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Abstract

This research studied the prevalence of different intestinal parasitic infections amongst *H. pylori* infected patients and assessed the association impact in the pathogenesis of *H. pylori* infection at the Tropical Medicine and Gastroenterology outpatient clinic, South valley University Hospitals. A total of 80 stool samples were collected. *H. pylori* infection was diagnosed by detecting the *H. pylori* antigen in stool. Accordingly, patients were divided into 2 groups: *H. pylori* positive cases (group I) and was 50 cases (62.5%) and *H. pylori* negative cases (group II). *H. pylori* positive cases were subjected to upper endoscopy. There was a significant association between parasitic infections and *H. pylori* in patients with gastrointestinal complaints ($P \leq 0.005$). *Entamoeba histolytica*/dispar (55.5%) was the commonest parasite detected in *H. pylori* positive cases. Upper gastrointestinal endoscopy of the stomach showed characteristic mucosal pattern (mosaic appearance with focal hyperemic area), indicated *H. pylori* infection.

Key words: Egypt, *Helicobacter pylori*, Intestinal parasites, *Entamoeba histolytica*/dispar.

Introduction

Helicobacter pylori (*H. pylori*) are gram-negative bacilli responsible for chronic gastritis. *H. pylori* being recognized as strongly associated with gastric and duodenal ulcers in all age groups (Mutaz and Carmen, 2015). *H. pylori* produce a sufficient amount of active urease enzyme to survive the acidic environment of the stomach (Smoot, 1997), and some enzymes with destructive effect to the epithelial lining of the stomach as: protease enzyme, vacuolating cytotoxin A (VacA) and phospholipases (Cameron *et al*, 2001)

Poor socioeconomic condition is a major risk factor for acquiring not only *H. pylori* infection but also parasitosis (Hotez *et al*, 2004). Moreover, prevalence of *H. pylori* infection is much higher in developing countries than in developed ones (Cave, 1997; Elsaied *et al*, 2009, Eldash *et al*, 2013).

Gastrointestinal tract parasites may induce mild, acute or chronic infections (Mazigo *et al*, 2010). The intestinal parasites cause gastrointestinal symptoms such as diarrhea, dysentery, vomiting, lack of appetite and abdominal distention (Garcia, 2004). In Egypt, *Entamoeba histolytica* was the most prevalent protozoan infection followed by *Giardia lamblia* infection (Bakr *et al*, 2009; Zaytoun *et al*, 2013).

In Egypt, there is more or less little information about the magnitude of parasitic infections amongst patients infected with *H. pylori* and suffering from recurrent GIT symptoms. No doubt, differentiation of parasitic agents plays an important role treatment.

This study aimed to examine the prevalence of different intestinal parasitic infections amongst *H. pylori* infected patients and to assess the impact of this association in the pathogenesis of *H. pylori* infection at the Tropical Medicine and Gastroenterology Outpatient Clinic, South Valley University Hospital, Qena Governorate, Egypt.

Material and methods

The study included 80 patients attended the

Tropical Medicine & Gastroenterology, Department, Faculty of Medicine, South Valley University, during the period from April 2015 to September 2016. They suffered from either upper GIT symptoms (heartburn, vomiting, hematemesis & melena) or of upper along with lower GIT symptoms (diarrhea, diarrhea alternating with constipation, abdominal distention, dysentery & tenesmus). Sheets were filled out on each patient. A patient with any of the gastric surgery, gastric malignancy, severe hepatic, renal, and cardiopulmonary impairment, bleeding tendency, smoking, addictive tea or coffee, on non-steroidal anti-inflammatory drugs, antiplatelet agents, anticoagulants, steroids, antibiotics, proton pump inhibitors or bismuth within last 30 days was excluded.

Stool examination: Two fresh stool samples were collected from each patient at the time of enrollment in the study. The first sample diagnosed *H. pylori* by detecting antigen using *H. pylori* stool antigen test (One step *H. pylori* antigen test device; ACON Laboratories Inc., San Diego, CA, USA) according to manufacturer's directions (Silva *et al*, 2010). So, patients were divided into 2 groups: *H. pylori* positive cases (GI) and *H. pylori* negative cases (GII).

The second stool sample was transferred to the Clinical Pathology laboratory, Faculty of Medicine to be examined immediately by different techniques for parasitic infections. In the laboratory, slides were then prepared directly for wet mount in saline as well as in iodine and then were microscopically examined initially under low power then under high power. Finally the sample was concentrated by applying the formalin-ethyl acetate technique (Dubey *et al*, 1990). The iodine stained slides were prepared and examined microscopically. Fixed smears were stained with Modified Ziehl-Neelsen stain (El-Shazly *et al*, 2006) and Giemsa stain (Garcia, 2004). Also, stool culture for nematode larvae on agar plates was done for detection of *S. stercoralis* or hookworm larvae (Knopp *et al*, 2008).

Endoscopic examination: To assess the degree of severity of gastritis related to *H. pylori* infection and to determine the role of parasitic infection in changing the mucosal pattern of the stomach in these patients, all patients included in the study who were proved to have *H. pylori* positive stool antigen were subjected to upper GIT endoscopy after conscious sedation using 5mg intravenous Midazolam, with upper video gastroduodenoscopes (EPK-I 5000 Olympus Europe, Hamburg, Germany).

Ethical considerations: Verbal consent was obtained from the patients. All procedures were conducted according to the ethical standards approved by the Institutional Human Ethics Committee, Faculty of Medicine, South Valley University.

Statistical analysis: Data were entered and analyzed by SPSS 19, quantitative data presented as mean and standard deviation, qualitative data presented as frequency, chi square, z test and one way ANOVA test. P value of less than 0.05 considered as cut off for significance.

Results

H. pylori infection was detected in 50/80 patients (62.5%). The prevalence of intestinal parasitic infection in *H. pylroi* positive cases (GI) was 36/50 (72%) versus 13/30 (43.3%) in *H. pylori* negative cases (GII) were significant (P= 0.005). Intestinal parasites detected included *Entamoeba histolytica/dispar* (55.5%), *Blastocystis hominis* (27.7%), *Giardia lamblia* (38.8%), *Ascaris lumbricoides* (19.4%) and *Enterobius vermicularis* (2.7%). Eight samples with mixed infections: *E. histolytica/dispar* and *B. hominis* in three samples, *E. histolytica/dispar* and *E. vermicularis* in one sample, *G. lamblia* and *B. hominis* in two sample, *A. lumbricoides* and *B. hominis* in two samples. There was significant association between *E. histolytica/dispar* and *H. pylori* (P<0.05).

Table 1: Prevalence of intestinal parasites amongst *H. pylori* positive and *H. pylori* negative patients.

Intestinal parasites	GI positive patients (n=50) No (%)	G II negative patients (n=30) No (%)	Z test	P value
Intestinal parasites (+ve)[a]	36 (72)	13 (43.3)	4.4	0.005*
Entameba histolytica/dispar	20 (55.5)	2 (15.3)	3.3	0.003*
Blastocystis spp.	10(27.7)	8 (61.5)	2.1	0.01*
Giardia lambia	14(38.8)	3 (23.1)	0.9	0.1
Cryptosporidium spp.	0 (0.00)	5 (23.3)	2.9	0.001*
Ascaris lumbricoides	7 (19.4)	2 (38.5)	1.3	0.08
Entrobiuos vermicularis	1 (2.7)	0 (0.00)	0.5	0.3
Intestinal parasites (-ve)	14 (28)	17 (56.7)	1.01	0.1

Discussion

H. pylori colonizes gastric mucosa and one of the major cause of gastric ulcer, autoimmune gastritis, gastric cancer and B cell lymphoma of mucosa associated lymphoid tissue (Kusters *et al*, 2006). *H. pylori* stool antigen test has the advantage of being directed noninvasive (Sabbi *et al*, 2012).

The current study observed significant high prevalence of parasitic infection among *H. pylori* positive patients (P<0.005), as compared to *H. pylori* negative patients. This result was in concordance with few previous studies (El-Massry *et al*, 2003; Kazemian *et al*, 2014). *H. pylori* escaped the destructive effect of stomach acidity by neutralization, as it produces large amounts of urease enzyme which breaks down plasma urea in the stomach wall to ammonium ion. *H. pylori* proved risky infection by destroying stomach acidity (Smoot, 1997) an important body defense against ingested pathogens (Smith, 2003; David and William, 2006).

The present study showed a significant association between *E. histolytica/ dispar* and *H. pylori* infections (*P*<0.05). This strong association is attributed to the fact that *H. pylori* has been found to affect the sensorymotor system of the gut including the colon, in addition to its urease enzyme neutralization effect on stomach acidity. Kazemian *et al*. (2014) in Iran among 37 children with *H. pylori* infection they reported *G. lamblia* and *E. histolytica/dispar* in 29.7%, & 10.8% respectively. Fouad *et al*. (2014) in Egypt stated that *H. pylori, G. intestinalis* and coeliac disease were common causes of dyspepsia and found *G. intestinalis* genotype A greatly associated with dyspeptic symptoms. Sabah *et al*. (2015) in Egypt among 72/140 with *H. pylori* co-infection with *E. histolytica*, or *G. lamblia* found (51.4%), and added that *H. pylori* was about 70% in Tanta City in patients with different gastrointestinal symptoms. But, others found association between *G. lamblia* and *H. pylori* infections (Zeyrek *et al*, 2008; Escobar-Pardo *et al*, 2011), the present study did not show significant association (P>0.05). All these parasites are transmitted faeco-orally either directly or through consumption of food or water contaminated with their infective stages; prevalence rate of these parasitic infections mainly depends on sanitary conditions. Most patients were from rural areas with poor sanitary conditions and of low socioeconomic level which are major risk factors for acquiring both *H. pylori* and parasites (Cameron *et al*, 2001). *H. pylori* possibly act as an additional risk factor for those parasitosis.

In the present study, upper gastrointestinal endoscopy of stomach showed characteristic mucosal pattern of *H. pylori* that was mosaic appearance, with or without focal hyperemic areas that agreed with Yan *et al*. (2010) who reported that mosaic mucosal pattern of the stomach was an indicator for *H. pylori*-positive gastritis. But, no difference in stomach mucosal pattern was detected among patients with or without parasitic infections.

Conclusion

G. lamblia triggers symptoms of functional dyspepsia. The study showed that *H. pylori* were a major risk factor for *Entamoeba* spp. co-infection in rural areas of Qena Governorate. This needs proper intervention to re-

duce co-infection.

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