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CONCOMITANT INFECTION OF PARASITES AND HELICOBATER PYLORI IN SOHAG UNIVERSITY HOSPITALS

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Abstract

Helicobacter pylori is a gram-negative bacterium colonizes the gastric mucosa. Chronic infections can develop various gastrointestinal complaints as gastritis, gastric and/or duodenal ulcers, cancers, or mucosa-associated lymphomas. Also, gastrointestinal diseases are caused by bacteria, viruses, and parasites. This study evaluated the intestinal parasites among positive and negative *H. pylori* infected outpatients. Stool samples (200) collected from individuals who attended outpatient clinics in Sohag University Hospitals were examined for *H. pylori* antigen and gastrointestinal parasites. They were divided into two groups of 100 each, G1: *H. pylori* positive antigen cases and G2: *H. pylori* negative ones as control. The results showed significant increase in G1 than in G2 as to *G. lamblia*, *C. parvum*, *E. histolytica*, *B. hominis*, *E. coli*, *I. buetschilii*, *Cyclospora* spp., *E. vermicularis*, *H. nana* and *A. lumbricoides*.

Key words: Patients, *Helicobacter pylori*, gastritis, gastrointestinal, parasites

Introduction

Gastrointestinal troubles are one of the main globally causes of morbidity and mortality, mainly in developing countries caused by parasites, viruses and bacteria (Shaldoum *et al,* 2017). About 50% (over 3 billion) of world populations were infected with *H. pylori* (Salih, 2009). Meanwhile, *Helicobacter pylori*, a highly mobile gram-negative, distincti- vely twisted bacterium, affects about 50% of the world's population leading to gastric pa-hologies, such as inflammation, gastrointest- inal ulcers and/or malignancies (Elbehiry *et al,* 2023). *Helicobacter pylori* colonize in as chronic infection in the gastric mucosa (Krz- yzek and Gosciniak, 2017). The frequency of co-infections adds to the complex clinical pictures, as different agents may have poten-tial synergistic or antagonistic interactions, with impact on treatment, and susceptibility to other diseases (Mcardle *et al,* 2018). The stomach acidic prevents survival of viruses, parasites and other bacteria. *H. pylori* have evolved to be uniquely suited to thrive in the harsh stomach environment by secreting a special enzyme urease that converts urea to ammonia reduces acidity (Kazemian *et al,* 2016). This risk factor allows pathogenic intestinal protozoa such as *G. lamblia* and other gastrointestinal parasites to pass the stomach's increased pH causing infections (Shaldoum *et al,* 2017). So, there is a need to explore co-infection to stop risk complications (Tay *et al,* 2016).

In Upper Egypt Dyab *et al.* (2024) reported that *G. lamblia*, *E. histolytica*, and *E. vermicularis* were among the commonest infections. El Shazly *et al.* (2006) in Lower Egypt reported that helminthes in a descending order were *S. mansoni*, *Fasciola* sp., *H. heterophyes*, *Hymenolepis nana, Trichostrongylus* sp., *A. lumbricoides*, *S. stercoralis*, *H. diminuta*, *Tsaginata, E. vermicularis*, *T. trichura,* then *A. duodenale,* and intestinal protozoa were *B. hominis*, *G. lamblia*, *E. histolytica*/*E.dispa*, *I. butschlii*, *C. parvum, E. coli*, *Isospora hominis*, *E. nana*, *E. hartmani*, *D. fragilis*, *Ch. mesnili*, *T. hominis*, *C. cayetanensis*, *E. hominis* and *E. intestinalis*.

Although transmission mechanism is unclear until now yet *H. pylori* spread among indoo- rs via close contact. Transmission may also occur directly by contaminated hands, inadequate water supply, or ingestion of contaminated raw vegetables (Larussa *et al,* 2015). Also, freshwater and fish have a role in *H. pylori* dissemination (Mubarak *et al,* 2023). Besides, Spotts *et al.* (2020) reported that *H. pylori* co-infection with one or more of the zoonotic parasites among school-aged children ranged between 22.4% to 44.3%

Patients and Methods

This case control study was done from September 2022 to April 2023 on outpatients of different ages of both sexes who attended Sohag University Hospitals. They were 200 patients: 100 with positive *H. pylori* antigen as cases &100 patients without *H. pylori* antigen as a control. *H. pylori*-Ag in stools was diagnosed by immuno-chromatographic test.

Inclusion criteria: not on antibiotics 4 weeks before sampling or proton pump inhibitors 2 weeks before sampling or anti-parasitic drugs 2 weeks before sampling. A standard questionnaire was completed by patients for demographic data; age, sex, residence, and family size.

Stool samples (2 to 5gm) were collected from patients in labeled clean, covered containers, without urine. Each sample was divided into two parts; one to diagnose *H. pylori*-Ag by immunochromatographic (Chemtrue® One-Step, Biotech Co., Ltd., Shanghai, China), after the manufacturer's instructions, and the second for microscopic examination as direct wet smear, concentration method, and stained with modified Kinyoun's Acid-Fast Stain for coccidian parasites (Garcia, 2016). Macroscopic examination was done adults and gravid segments.

Statistical analysis: Data were tabulated and analyzed by using SPSS version 25.0. Variables were presented as M ±SD, frequencies and percentages. Chi square ($\chi 2$) and Fiher's exact tests compared between qualitative data. A P-value of < 0.05 was significant.

Ethical approval: The study was approved by the Scientific Ethical Committee, Sohag Faculty of Medicine, University (Soh-Med-22-04-15) & registered at clinical trials.gov (NCT05360940). All participants were informed about the study purpose and signed a written concept.

Results

Participants' ages ranged from 2 to 72years (25.940±16.382), of whom (56.5%) were males, and (59.5%) live in rural areas. Family size (57%) was ≥ 5 members. The patients and control were cross matched ($P > 0.05$).

H. pylori and parasites were (42%) than in control (13%), with significant increase ($P < 0.001$). Cases without parasites (58%) were less than in control (87%) with ($P < 0.001$).

G. lamblia was 24%, followed by *C. parvum* 14% & *E. histolytica* (12%). In control, *G. lamblia* was 3%, *C. parvum* 2% & *E. histolytica* (10%), with significant difference as to *G. lamblia* ($P < 0.001$), *Cryptosporidium* (P= 0.002) and *B. hominis* (P= 0.01), without significant as to *E. coli* (P= 0.774), *I. butchii* (P= 0516), *Cyclospora* (P= 0.700), *E. vermicularis* (P= 0.651), *H. nana* (P= 0.651), *A. lumbricoides* (P= 0.651).

Cases < 15 years showed high infection rate (51.43%), than > 45years ones (20%), but without significant (P =0.326), also, without difference among both sexes as to with or with-out parasites (p= 0.276). Males (23) were more infected than female in co-infected cases (19) and in parasite-free (35, & 23 respectively).

High infected ages in control (> 30-45y) was (13.64%) and low (> 45y) was (12.5%), without significant (P =1). Parasites in males (13.46%) was more than females (12.5%), without significant (P =0.886).

Details were given in tables (1, 2, 3, 4, & 5).

Table 1: Demographic characteristics (n = 200).

Demographic characters	No. (%)
Ages	25.940±16.382
<15 Years	66 (33%)
15-30 Years	57 (28.5%)
>30-45 Years	51 (25.5%)
>45 Years	26 (13%)
Male	113 (56.5%)
Female	87 (43.5%)
Residence: Rural	119 (59.5%)
Urban	81 (40.5%)
Family size:< 5	86 (43%)
≥ 5	114 (57%)

Table 2: Demographic characteristics of patients as to *H.pylori* infection.

	Cases (N=100)	Control (N=100)	P-value
Ages	25.590±16.046	26.290±16.785	
<15 Years	35 (35%)	31 (31%)	0.438
15-30 Years	26 (26%)	31 (31%)	
>30-45 Years	29 (29%)	22 (22%)	
>45 Years	10 (10%)	16 (16%)	
Males	61(61%)	52(52%)	> 0.05
Females	39(39%)	48(48%)	
Residency: Rural	65(65%)	54 (54%)	> 0.05
Urban	35 (65%)	46 (46%)	
Family size: < 5	48 (48%)	38 (38%)	> 0.05
≥ 5	52 (52%)	62 (62%)	

Table 3: *Helicobacter pylori* with or without intestinal parasites.

Parasite	Cases (G1)		Control (G2)		Total		P value
	No.	%	No.	%	No.	%	
Co-infected	42	42.00	13	13.00	55	27.50	< 0.001
No parasite	58	58.00	87	87.00	145	72.50	
Total	100	100.00	100	100.00	200	100.00	

Table 4: Parasites among positive *H. pylori* versus negative *H. pylori.*

Parasites	Cases (G1)		Control (G2)		
	No.	%	No.	%	P-value
Giardia lamblia	24	24.00	3	3.00	<0.001*
Cryptosporidium	14	14.00	2	2.00	0.002*
Entamoeba histolytica	12	12.00	10	10.00	0.651
Blastocystis hominis	11	11.00	2	2.00	0.010*
Entaomeba coli	7	7.00	6	6.00	0.774
Iodamoeba Butchii	6	6.00	4	4.00	0.516
Cyclospora	4	4.00	3	3.00	0.700
Enterobius vermicularis	2	2.00	3	3.00	0.651
Hymenolepis nana	3	3.00	2	2.00	0.651
Ascaris Lumbricoides	2	2.00	3	3.00	0.651

Table 5: Distribution of co-infection versus control as to age groups and sexes.

	Heliobacter patients (G1)					Control (G2)				
Variations	With parasite		No parasite		P-value	With parasite		No parasite		P-value
	No.	%	No.	%		No.	%	No.	%	
< 15 years	18	51.43	17	48.57	= 0.326	4	12.90	27	87.10	= 1
15-30 years	11	42.31	15	57.69		4	12.90	27	87.10	
>30-45 years	11	37.93	18	62.07		3	13.64	19	86.36	
> 45 years	2	20.00	8	80.00		2	12.50	14	87.50	
Male	23	37.70	35	62.30	= 0.276	7	13.46	45	86.54	= 0.886
Female	19	48.72	23	51.28		6	12.50	42	87.50	

Discussion

H. pylori can overcome the stomach's acidity by the ammonia resulted from the urease enzyme. This allowed gastrointestinal parasites to cross the high pH (Shaldoum *et al*, 2017).

In the present study, co-existence of *H. pylori* and parasites mostly shared gastrointestinal symptoms and mode of transmission. This agreed with Cheng *et al*. (2009), who reported that *H. pylori* supported C. *parvum* and *G. intestinalis* colonization in human gastrointestinal tract. But, gastrointestinal parasitic infection may affect inflammatory response to *H. pylori* (Whary *et al*, 2011). Co-existence of *H. pylori* and intestinal parasites might interact synergistically led to serious health complications, which could be influenced by hosts and environmental factors (Marini *et al*, 2007). Association of *H. pylori* and gastrointestinal parasites increased risk factors (Cheng *et al*, 2009).

In the present study, *H. pylori* and parasites was (42%) in cases with significant associated with *G. lamblia*, *C. parvum* and *B. hominis*. This agreed with Eldash *et al.* (2013), who among Egyptian children reported *H. pylori* and *G. intestinalis* was among

36 (40.0%) patients and 11 (12.2%) controls with a significant difference (p<0.001). This agreed with Shaldoum *et al.* (2017), they found that *H. pylori* and parasites were (41.1%). This also agreed with Ibrahim *et al*. (2019), who found (43.9%) significantly associated with *G. intestinalis* and *C. parvum*. Abroad, it agreed with Ankarklev *et al*. (2012) in Uganda, who reported of *H. pylori* (44.3%) with *G. intestinalis* (20.1%) as dominant concomitant parasite with was 3-folds higher of *G. intestinalis* and *H. pylori* infections versus control. Also, this agreed with Seid *et al.* (2018) in Ethiopia, who found high (44.3%), with high significant of *G. lamblia* (22.3%) in *H. pylori*-infected patients. But, it was higher than Krzyzek and Gosciniak, (2017) in Iran, who reported that giardiasis co-infected was (29.7%) in cases. Metwally *et al.* (2022) reported that in Egyptian patients, *H. pylori* had >90% resistacce to metronidazole and amoxicillin. Mayo-Clinic (2022) reported that metronidazole is the most commonly used antibiotic for giardiasis, but side effects may include nausea and a metallic taste in the mouth. Korenková *et al.* (2024) reported that giardiasis, often presents a treatment challenge, particularly in terms of resistance to metronidazole. Despite extensive research, markers for metronidazole resistance have not yet been identified.

In contrast, this result disagreed with Ghallab and Morsy (2020) in Egypt, who found that *H. pylori* positive patients (83.8%) were co-infected with *G. intestinali* 31(19.3%), *E. histolytica* 27(16.8%), *C. parvum* 32(20%) and *B. hominis* 68(42.5%). Abroad, this disagreed with Abd El-bagi *et al. (*2019) in Sudan, who found that co-infection was 23% in *H. pylori* patients. Also, it disagreed with Demirel and Evren (2020) in Turkey, who found that parasites and positive *H. pylori* antigen in stools was (12.3%). Also, this disagreed with Yakoob *et al*. (2018) in Pakistan, who found that *H. pylori* and parasites were co-infected 27/33 (81.8%) in *H. pylori* patients versus 17/27 (63%) in control. They added that co-infect- ed *Blastocystis* spp. was (67%) and *E. histol-ytica* was (21%). It differed from Pomari *et al. (*2020) in Italy found that parasites were in 74% in *H. pylori* patients, with significant increased *B. hominis*. However, Ugraş and Miman (2014) in Turkey didn't find association between *H. pylori* and parasites.

In the present study, there was significant association between *H. pylori* and *G. lamblia* (P <0.001), *C. parvum* (P= 0.002), and *B. hominis* (P= 0.010), co-infection. High infection *G. lamblia* (24%) rate was comp-ared to *E. histolytica* (12%). This agreed with Abou El-Hoda *et al.* (2007) and Sabah *et al.* (2015) in Egypt. Abroad, it agreed with Al-Shammari *et al.* (2001) in Saudi Arabia, Andargie *et al.* (2008) in Ethiopia, Khurana *et al.* (2008) in India, Isaev and Efimova (2010) in Russia, Zaglool *et al.* (2011) in Saudi Arabia, Koohsar *et al.* (2012) and Balarak *et al.* (2016) in Iran. But, this disagreed with Ahmed *et al.* (2016) in Sudan, they didn't find significant difference of *G. lamblia* co-infected with *H. pylori* among *H. pylori* infected or uninfected ones. They added that a high *E. histolytica* rate was among *H. pylori* infected patients than uninfected ones.

In the present study, *H. pylori* and *C. parvum* was significantly associated (P= 0.002). This agreed with Ibrahim *et al.* (2019) reported (5.3%), and Ghallab and Morsy (2020) reported (20%).

In the present study, *B. hominis* (11%) agreed with Demirel and Evren (2020), in Turkey, who found *B. hominis* was (12.3%) of *H. pylori* antigen positive stools. But, this disagreed with (42.5%) reported by Ghallab and Morsy (2020), who found a significant association between *H. pylori* and *B. hominis* and with Nghaimesh *et al.* (2018) in Iraq, who found (67.2%) of *H. pylori* antigen positive stools. *H. pylori* and parasites in females (48.7%) was high than in males (37.7%), without significant difference (P= 0.276). This agreed with Bin Mohanna *et al*. (2014), in Yemen, and Shaldoum *et al*. (2017), in Egypt. But, this disagreed with Lee *et al*.

(2000) in Korea, and Shehata and Hassanein (2015) in Egypt, reported that the vise was versed.

Conclusion

Gastrointestinal parasites were more common among *Helicoobacter pylori* infected patients, with an overall rate (42%). *G. lamblia*, followed by *C. parvun* and then *B. hominis* were the most common co-infections with significant differences mainly among age group < 15years. Both *H. pylori* and *Giardia* are resistant to metronidazole.

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