

ASSOCIATION OF INTESTINAL PARASITES WITH HELICOBACTER PYLORI INFECTION IN PATIENT ATTENDING BENHA UNIVERSITY HOSPITAL

By

AMIRA SALAH EL-GHANNAM^{1*}, AMIRA K. ELALFY², MARWA S.E. ABD ELRAOUF³, DINA ABD ELHADY MOHAMMED¹, GHADA H. OMAR¹, and NAGAT AHMED SOLIMAN¹

¹Department of Parasitology, ²Department of Internal Medicine, ³Department of Public Health and Preventive and Social Medicine, Faculty of Medicine, Benha University, Benha, Egypt (*Correspondence: amira.elghnnam@fmed.bu.edu.eg).

Abstract

This cross sectional study was conducted from October 2023 to March 2024, on 170 patients attending Gastroenterology outpatient clinic in Benha University hospital and aimed to investigate the association of intestinal parasites with *H. pylori* infection and the associated symptoms.

Stool samples were collected and examined within an hour by: direct wet smear technique, concentration method and Modified Ziehl-Neelsen stain. A rapid chromatographic immuno-assay was used for the qualitative detection of *H. pylori* copro-antigens in stool. All patients with aged ranged from 20 to 60 years old, were 73 males (42.94%) and 97 females (57.06%) presenting with gastrointestinal manifestations.

The results showed that 103 (60.59%) patients were positive *H. pylori* copro-antigen in stool and 24 (14.12%) had different intestinal parasites. There was a significant association between intestinal parasites and *H. pylori* infection (P= 0.0001). Besides, intestinal parasites were associated with *H. pylori* were in (18.45%) of positive patients, and in (7.46%) of *H. pylori* negative patients.

There was significant difference for different types of intestinal parasites as regarding *H. pylori* infection (P=0.007). *Entamoeba histolytica* 11 (10.68%) were the most frequently detected parasite in association with *H. pylori*, followed by *Giardia lamblia* 5 (4.85%) and *Enterobius vermicularis* 2 (1.9%).

Keywords: Intestinal parasites, *H.pylori*, gastrointestinal manifestations, association.

Introduction

Intestinal parasites infect millions of people worldwide with high probability of associated infection with *Helicobacter pylori* (Pomari *et al*, 2020). In some developing countries, the intestinal parasites can cause morbidity and mortality due to polluted water, hot humid atmosphere, poor human safety, and sanitation (Eyayu *et al*, 2021). *H. pylori*, Gram-negative helical bacilli, affect about 4.4 billion people (more than 50% of the world population). The World Health Organization (WHO) considers it a class 1 carcinogen causing peptic ulcer and gastric cancer (Alexander *et al*, 2021). The *H. pylori* infection rate is 30-50% in developed and 85-95% in some countries (Bashir and Khan, 2023). *H. pylori* and intestinal parasitic infections had recognized as possible etiological factors for

gastrointestinal symptoms such as diarrhea and abdominal pain (Abd El Hameed *et al*, 2021). Multiple noninvasive tests are used to detect *H. pylori* antigens in stool samples, and are widely used for diagnosis and follow-up after antibiotic treatment (Kayali *et al*, 2018). Infection with intestinal protozoa in association with *H. pylori* can aggravate host immune responses (Krzyżek and Gościński, 2017). Co-infection with protozoa caused gastric mucosal damage (Hussain *et al*, 2020)

This study aimed to evaluate the association between intestinal parasites and *H. pylori* among symptomatic out-patients attended Benha University Hospitals.

Materials and Methods

Study design: This cross sectional study was included 170 patients who were attending Gastroenterology outpatient clinic

in Benha University hospital and complaining of gastrointestinal symptoms during the period from October 2023 to March 2024. Patients who had chronic illness such as liver, kidney or heart disease, diabetes mellitus, antibiotic or anti-parasitic therapy before 3 weeks of the study were excluded. A questionnaire was designed to obtain socio-demographic features about the patients such as age, sex, education, economic status, drinking water, residence, raising animals, employment status and their complaints. Stool samples were collected in clean plastic containers after written informed consents obtained from all patients. Stool samples were examined within an hour at Parasitology department, Faculty of Medicine, Benha University by direct wet smear technique, concentration method (Garcia, 2007) and modified Ziehl-Neelsen stain (Henriksen and Pohlens, 1981).

A rapid chromatographic immunoassay was used for the qualitative detection of *H. pylori* copro-antigen in stool specimens (Rightsign[®], *H. pylori* antigen rapid test Cassette, (feces) REF IHPG-C61, Hamburg, Germany) with sensitivity of > 99.9% and specificity of 98.1% .

Sample size: the sample size was calculated using Epi-info software version 7.2.5.0 based on a previous study by Seid *et al.* (2018) who reported a 70.3% prevalence of *H. pylori* among upper gastrointestinal symptomatic adult patients. The minimum sample size needed will be 165 patients. The confidence level and margin of error were adjusted at 95% & 7%, respectively

Ethical approval: This study was approved by the Research Ethical Committee, Faculty of Medicine, Benha University (No. RC 1-9-2023).

Statistical analysis: Data were collected, computerized and analyzed by using SPSS v. 20.0 (Statistical Package for Social Sciences). Categorical data were expressed as frequency and percentages. Chi squared test was used to determine the difference

and association between groups. McNemar test was used to determine the association between matched groups with categorical variables. Accepted significant level was at $P=0.05$, and $P<0.001$ highly significant.

Results

The 170 patients with age ranges between 20 to 60 years old, included 73 males (42.94%) and 97 females (57.06%) with gastrointestinal manifestations. After stool examination and *H. pylori* copro-antigen detection test, we had get 4 groups; GI: intestinal parasitic infection positive patients included 24 (14.12%) of patients, GII: *H. pylori* infection positive patients included 103(60.59%) of patients, GIII: (associated infection) positive patients for both parasitic and *H. pylori* infection included 19 (11.18%) of patients, and G IV: patients negative for both parasites and *H. pylori* infection included 62 (36.47%) of total patients. There was a significant association between intestinal parasites and *H. pylori* ($P=0.0001$). The intestinal parasites were in (18.45%) of *H. pylori* patients, and in (7.46%) of *H. pylori* negative ones.

There was a significant difference with different intestinal parasites as to *H. pylori* infection ($P=0.007$). Intestinal protozoa, 21(87.5%) of patients positive for intestinal parasites, were more frequent than intestinal helminthes 3(12.5%). *Entamoeba histolytica* 11(10.68%) were the common parasite associated with *H. pylori* patients, followed by *Giardia lamblia* 5(4.85%) and *Enterobius vermicularis* 2 (1.9%).

Socio-demographic characters of participants showed that 128 (75.29%) were educated, 96 (56.47%) were rural residents, 99 (58.24%) were employed, 164 patients (96.47%) used tap water, 11 (6.47%) had pet animal(s).

There was significant difference for *H. pylori* in different age groups ($P=0.0004$), but intestinal parasites was insignificant in different age groups ($P=0.053$). *H. pylori* patients were higher (39.80%) among age group (31-40years) and lower

(6.79%) among (41-50).

Intestinal parasites were higher (33.3%) among age group (41-50years) and lower (12.5%) among age group (50-60). *H. pylori* was more in females 62 (60.19%), without significant (P= 0.305), and intestinal parasites were more in males 15(62.5%) with significant (P=0.036).

Education and employment were insignificant in *H. pylori* patients (P=0.571 & P=0.205) respectively, but were significant in intestinal parasites (P<0.001& P=0.001) respectively.

As regards *H. pylori* infection, residence, and keeping pet animals were insignificant (P=0.0001) (P=0.136) respectively. While

in parasitic infection, residence and pet animals were insignificant (P=0.256 & 0.688) respectively. Water consumption showed a significant high *H. pylori* difference was in patients on tap water (94.17%) (P= 0.044), but insignificant in intestinal parasites (95.8%) (P=0.855).

Patients were bloating 24 (100%) was a highly significant symptom (P1=001), followed by decreased appetite 20 (83.3%) in intestinal parasitic infection, for *H. pylori* patients, abdominal pain 90 (87.38%) was a highly significant (P2<.0001), followed by bloating 85(82.5%).

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

Table 1: Association between intestinal parasites and *H. pylori* infection:

Variations	Parasite positive (n=24)	Parasite negative (n=146)	P(McNemar's test)
<i>H. pylori</i> positives (n=103)	19 (18.45%)	84 (81.55%)	0.0001
<i>H. pylori</i> negatives (n=67)	5 (7.46%)	62(92.54%)	

Table 2: Intestinal parasites among *H. pylori* (n=24)

Parasites	<i>H. pylori</i> positive (n=103)	<i>H. pylori</i> negative (n=76)	Parasites (n=24)	P value
<i>E. histolytica</i>	11 (10.68%)	1 (13.16%)	12 (50%)	0.007**
<i>G. lamblia</i>	5 (4.85%)	0 (00.00%)	5 (20.8%)	
<i>C. parvum</i>	1 (0.97%)	3 (3.94%)	4 (16.67%)	
<i>H. nana</i>	0 (00.00%)	1 (13.16%)	1 (4.17%)	
<i>E. vermicularis</i>	2 (1.9%)	0 (00.00)	2 (83.33%)	

Table 3: Socio-demographic features for *H. pylori* among participants:

Items	<i>H. pylori</i> positive (60.59%)	<i>H. pylori</i> negative (39.41%)	P value
Age (years): 20-30	39 (37.86%)	12 (17.91%)	0.0004**
: 31-40	41 (39.80%)	28 (41.79%)	
: 41- 50	7 (6.79%)	19 (28.35%)	
: 51-60	16 (15.53%)	8 (11.94%)	
Males	41 (39.80%)	32 (47.76%)	0.305
Females	62 (60.19%)	35 (52.23%)	
Education: Illiterate	27 (26.21%)	15 (22.39)	0.571
: Educated	76 (73.78%)	52 (77.61)	
Residence: Urban	33 (32.04%)	41 (61.19)	0.0001**
: Rural	70 (67.96%)	26 (38.81)	
Pet animal: Yes	9 (8.74%)	2 (2.98%)	0.136
: No	94 (91.26%)	65 (97.01%)	
Employment: Yes	56 (54.37%)	43 (64.17)	0.205
: No	47 (45.63)	24 (35.82)	
Tap water: Yes	97 (94.17%)	67 (100%)	0.044*
: No	6 (5.82%)	0 (00.00)	

P<0.05= significant*,P<.001= highly significant**.

Table 4: Socio-demographic characteristics of parasites among participants:

Variations	Parasite positive (14.12%)	Parasite negative (85.88%)	P value(x ² test)
Age (years): 20-30	7(29.2%)	44(30.1%)	0.053
: 31-40	6(25%)	63(43.2%)	
: 41- 50	8(33.3%)	18(12.3%)	
: 51-60	3(12.5%)	21(14.4%)	
Males	15(62.5%)	58(39.7%)	0.036*
Females	9(37.5%)	88(60.3%)	
Education: Illiterate	18(75%)	24(16.4%)	<0.001**
: Educated	6(25%)	122(83.6%)	
Residence: Urban	13(54.2%)	61(41.8%)	0.256
: Rural	11(45.8%)	85(58.2%)	
Employment: Yes	21(87.5%)	78(53.4%)	0.001**
: No	3(12.5%)	68(46.6%)	
Tap water: Yes	23(95.8%)	141(96.6%)	0.855
: No	1(4.2%)	5(3.4%)	
Pet animal: Yes	2(8.3%)	9(6.2%)	0.688
: No	22(91.7%)	137(93.8%)	

Table 5: Associated complaints in study participants:

Symptoms	Parasite +ve	Parasite -ve	<i>H. pylori</i> +ve	<i>H. pylori</i> -ve	P value
Perianal Itching: Yes	1(4.2%)	0(0)	1(0.1)	0(0)	=.013*
: No	23(95.8%)	146(100%)	102(99.1%)	67(100%)	=0.418
Saliva: Yes	16(66.7%)	41(82.1%)	55(53.4%)	2(3%)	<.001**
:No	8(33.3%)	105(71.9%)	48(46.6%)	65(97%)	=<.0001**
Appetite: Yes	20(83.3%)	81(55.5%)	84(81.6%)	17(25.4%)	=0.010**
: No	4(16.7%)	65(44.55)	19(18.4%)	50(64.6%)	=<.0001**
Diarrhea: Yes	14(58.3%)	19(13%)	28(27.2%)	5(7.5%)	<.0001**
: No	10(41.7%)	127(87%)	75(72.8%)	62(92.5%)	=.001**
Constipation: Yes	5(20.8%)	36(24.7%)	37(35.9%)	4(6%)	=0.684
: No	19(79.2%)	110(75.3%)	66(64.1%)	63(94%)	<.0001**
Bloating: Yes	24 (100%)	81(55.5%)	85(82.5%)	20(29.9%)	<.001**
: No	0(0%)	65(44.5%)	18(17.5%)	47(70.1%)	<.0001**
Abdominal Pain: Yes	19(79.25%)	113(77.4%)	90(87.4%)	34(50.7%)	=0.847
: No	5(20.8%)	33(22.6%)	13(12.6%)	33(49.3%)	<.0001**
Nausea and vomiting: Yes	4(16.7%)	63(43.2%)	56(54.4%)	11(16.4%)	=0.013*
: No	20(83.3%)	83(56.8%)	47(45.6%)	56(83.6%)	<.0001**

P<0.05= significant*,P<.001= highly significant**.

Discussion

The present results showed that (60.59%) of patients were positive *H. pylori* copro-antigen in stool and (14.12%) had different intestinal parasitic infection.

This agreed with Pomari et al. (2020) who reported (66%) subjects were infected with *H. pylori* in a study in Italy. It was disagreed with Kaya et al. (2023), who reported *H. pylori* positivity rate was (27.79%).

Different rates were reported by Abd El Hameed et al, (2021) who reported that (16%) of cases were positive for *H. pylori* and (32.67) of cases were positive for intestinal parasitic infection.

In the present study, there was a significant association between intestinal parasites and *H. pylori* infection (P = 0.0001). As the results showed that intestinal parasites were

detected in (18.45%) of *H. pylori* positive patients, (associated infection), this was agreed with Abd Elbagi et al, (2021) who reported that the occurrence of intestinal parasite among *H. pylori* patients was (23%) in a study was done in Sudan. Pomari et al, (2020) reported a higher rate of coinfection between *H. pylori* and parasitic infection as (74%) of cases had a concomitant infection, and this association was significant. This results was disagreed with Abd El Hameed et al, (2021) who reported that co-infected cases represented (61.9%) of the *H. pylori* positive group.

No doubt, association of both infections could result from sharing the same mode of infection, the feco-oral route, also low socio-economic status and poor sanitary conditions may be risk factors for both infections

(Moreira *et al.*, 2005).

The results showed that *E. histolytica* (10.68%) were the most frequently detected parasite in association with *H. pylori* patients, followed by *G. lamblia* (4.85%) and *E. vermicularis* (1.9%). This agreed with Abd Elbagi *et al.*, (2021) who reported that *E. histolytica* was detected in 12% of the *H. pylori* cases followed by *E. coli* in 7% of cases and *G. lamblia* in 4%. A significant relationship between *E. histolytica* positivity and *H. pylori* positivity was reported by (Kaya *et al.* 2023), as it was (27.3%) among *H. pylori* positive cases and (1.8%) for *G. lamblia*, and it was (2.1%) for *E. vermicularis*.

Our results disagreed with Pomari *et al.*, (2020) who reported that *Blastocystis* was the most frequent as (67%) of cases were infected, followed by *E. coli* (20%). It was disagreed with Abd El Hameed *et al.* (2021) who reported that *G. lamblia* was the most frequently (35.9%) detected parasite associated with *H. pylori* infection.

In the present study the highest prevalence rates of *H. pylori* patients (39.80%) were found among the age groups 31-40 years old, and the lowest rate (6.79%) among the age group 41- 50 years old. This was agreed with (Kaya *et al.* 2023), who reported the highest prevalence rates of *H. pylori* positive patients (75.7%) were found among the age groups more than 40 years old. This disagreed with Abd Elbagi *et al.* (2021), who reported the highest prevalence rates (40%) of *H. pylori* patients were among the age groups 1-15, while the lowest rate (10.7%) was among age group 31-45. Also, Abd El Hameed *et al.* (2021) showed that the highest prevalence of *H. pylori* was (45.8%) among 5-10 years age group.

The present study showed that the intestinal parasites rate of (33.3%) was highest among the age group 41- 50 years old and the lowest rate (12.5%) was among the 50-60 age group. Abd El Hameed1 *et al.* (2021) reported that the highest rate (42.8%) of intestinal parasites was at 1-5 year old and the lowest rate (22.4%) among the age group of

10-15 years old.

In the present study, *H. pylori* infection was more in females (60.19%), with insignificant ($P= 0.305$), while the intestinal parasites was more in males (62.5%), with significant ($P=0.036$). This agreed with Kaya *et al.* (2023), who reported that *H. pylori* infected females (76.6%) were significantly higher than males (23.4%). This disagreed with Abd Elbagi *et al.* (2021), who reported that the rate of intestinal parasites was (24%) among males without significant difference.

The present results showed a significant difference for type of water consumption, *H. pylori* infection rate (94.17%) was high in patients on tap water. This disagreed with Kaya *et al.* (2023), who reported lower rate of *H. pylori* (40.2%) in patients on tap water with significant difference.

This study showed that in *H. pylori* infected patients, abdominal pain was a highly significant symptom in (87.38%) of patients ($P2<.0001$), followed by bloating (82.5%). This agreed with Pomari *et al.* (2020), who reported that epigastric pain was the most significant symptom in *H. pylori* infected cases.

The present study showed that bloating (100%) was a highly significant symptom ($P1=001$), followed by decreased appetite (83.3%) ($P1=0.010$), in intestinal parasitic infected patients. The difference in site of *H. pylori* (upper gastrointestinal tract), and intestinal parasites (lower gastrointestinal tract) showed variations in patients' complaints. This agreed with Abd El Hameed *et al.* (2021), who found that abdominal pain, distension and diarrhea and other symptoms were the most frequent complaints among different patients .

Conclusions

Abdominal pain, bloating and decrease appetite were the most common symptom for intestinal parasites and *H. pylori* infection. The association of parasitic infection and *H. pylori* modulated clinical symptoms of each other. The intestinal parasites and *H. pylori*

must be routinely examined in patients with gastrointestinal symptoms.

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Explanation of figures

Fig. 1: Prevalence of parasitic and *H. pylori* infection among participants.

