



Investigation of *Helicobacter pylori* Antigen Positivity and Intestinal Parasite Coexistence in Stool Samples

Gaita Örneklerinde *Helicobacter pylori* Antijen Pozitifliği ile Intestinal Parazit Birlikteliğinin Araştırılması

✉ Filiz Demirel, ✉ Kübra Evren

Ankara Training and Research Hospital, Department of Medical Microbiology, Ankara, Turkey

Abstract

Aim: *Helicobacter pylori* and intestinal parasitic infections are commonly seen, especially in areas with low socioeconomic status and poor hygiene conditions. *H. pylori* and *Giardia duodenalis* can be commonly found in patients with upper gastrointestinal system complaints. It is thought that the urease activity of *H. pylori* may help intestinal parasites pass into the intestines without being affected by the acidic environment of the stomach. In this study, it was aimed to investigate the association of *H. pylori* and intestinal parasites (IP) in patients with gastrointestinal system complaints.

Material and Method: A total of 408 patients, who were admitted to our hospital with gastrointestinal complaints between 2018 and 2020 and whose *H. pylori* rapid antigen test was studied simultaneously with intestinal parasite examination in the stool, were evaluated retrospectively.

Results: Out of 408 patients whose stool samples were examined, one or more intestinal parasites were detected in 80 (19.6%), and *H. pylori* antigen test was positive in 65 (15.9%). While there was no statistically significant difference between *H. pylori* positivity and age groups, the rate of IP detection was found to be significantly higher in children aged 6-18 years. The most prevalent IP was *Blastocystis* sp. in 74 (18.1%) patients. Intestinal parasite and *H. pylori* antigen co-positivity in stool samples was detected in eight patients and it was not found statistically significant.

Conclusion: *H. pylori* and intestinal parasites are common all over the world. The relationship between *H. pylori* and IP is still controversial, and more studies that are comprehensive are needed to understand the association of *H. pylori* and IP, especially in patients with upper gastrointestinal system complaints.

Keywords: *Helicobacter pylori*, parasites, *Blastocystis*

Öz

Amaç: *Helicobacter pylori* enfeksiyonu ve intestinal parazitler enfeksiyonlar, özellikle düşük sosyoekonomik düzey ve kötü hijyen koşullarına sahip bölgelerde yaygın olarak görülmeye devam etmektedir. Özellikle üst gastrointestinal sistem şikayeti olan hastalarda *H. pylori* ve *Giardia intestinalis* gibi patojenler etken olarak saptanabilmektedir. *H. pylori*'nin üreaz aktivitesinin intestinal parazitlerin midenin asidik ortamından etkilenmeden bağırsaklara geçişine yardımcı olabileceği düşünülmektedir. Bu çalışmada gastrointestinal sistem şikayeti olan hastalarda *H. pylori* ve intestinal parazit (IP) birlikteliğinin araştırılması amaçlanmıştır.

Gereç ve Yöntem: Hastanemize 2018-2020 yılları arasında gastrointestinal şikayetlerle başvurmuş ve gaitada intestinal parazit incelemesi ile eş zamanlı olarak *H. pylori* hızlı antijen testi çalışılmış toplam 408 hasta retrospektif olarak değerlendirilmiştir.

Bulgular: Gaita örnekleri incelenen toplam 408 hastanın 80'inde (%19,6) bir ya da daha fazla sayıda intestinal parazit, 65'inde ise (%15,9) gaitada *H. pylori* antijen testi pozitif olarak tespit edilmiştir. IP saptanma oranı 6-18 yaş arasındaki çocuklarda anlamlı derecede yüksek bulunmuştur. En fazla tespit edilen IP *Blastocystis* sp. olup 74 (%18,1) hastada saptanmıştır. Gaitada *H. pylori* antijen pozitifliği ile birlikte intestinal parazit pozitifliği toplam sekiz hastada tespit edilmiş ve istatistiksel olarak anlamlı bulunmamıştır.

Sonuç: *H. pylori* ve intestinal parazitler tüm dünyada yaygın olarak görülmektedir. *H. pylori* ve IP arasındaki ilişkili halen tartışmalı olup özellikle üst gastrointestinal sistem şikayeti olan hastalarda *H. pylori* ile IP birlikteliğinin anlaşılabilmesi için daha geniş kapsamlı çalışmalara ihtiyaç vardır.

Anahtar Kelimeler: *Helicobacter pylori*, parazit, *Blastocystis*



INTRODUCTION

Intestinal parasitic infections (IPI) maintain their importance as a common public health problem, especially in low socioeconomic areas. Low education level, inadequate sanitation of drinking water and food, and lack of personal hygiene are the main factors that increase the prevalence of intestinal parasitic infections.^[1] IPI causes malnutrition, anemia, growth retardation, impaired cognitive skills, and reduced school performance, as well as gastrointestinal complications, especially in children.^[2,3]

Helicobacter pylori infection is more common in developing countries with low socioeconomic status. It is estimated that half of the world's population is infected with *H. pylori*. Although the mode of transmission of *H. pylori* is not known exactly, it is thought that it might be transmitted from person to person directly or from the environment through water and food.^[4,5]

The common transmission routes of *H. pylori* and intestinal parasites (IP) and similar predisposing factors bring to mind the possibility of co-infection. It is thought that *H. pylori*, which is predominantly located in the stomach corpus, increases the pH of the stomach with its urease activity and facilitates the passage of intestinal parasites to the intestine.^[6] In some studies, *Mycobacterium tuberculosis*, *Campylobacter* spp., and IP co-infections of HIV-infected patients have been shown to lead to further progression of gastrointestinal disorders and even death in some cases. Similarly, there is evidence to suggest that IP and *H. pylori* co-infection probably affect the development and exacerbation of gastrointestinal complications. It has been known that IP and *H. pylori* co-infection could worsen the cellular immune response by modulating the host immune response, thereby exacerbating gastric mucosal damage.^[7]

The relationship between *H. pylori* and IP is still controversial, and more and more comprehensive studies are needed on this subject. This study aimed to retrospectively evaluate the data of patients who applied to our hospital with gastrointestinal complaints and were requested to test for stool *H. pylori* rapid antigen test and intestinal parasites and to investigate the association of *H. pylori* positivity in stool with intestinal parasites.

MATERIAL AND METHOD

Ethics Committee Approval

For the study ethics committee approval dated 10.11.2021 and numbered E-21-638 was obtained from Ankara Training and Research Hospital Ethical Committee of Non-Invasive Clinical Research. All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Selection of Patient Group

In this study, a total of 408 patients who applied to our hospital with gastrointestinal complaints between 2018 and

2020 and whose *H. pylori* rapid antigen test was studied simultaneously with intestinal parasite examination in the stool were evaluated retrospectively.

Parasitologic Examinations

Fresh stool samples taken from the patients were evaluated for the presence of intestinal parasites after macroscopic examination, native-lugol direct microscopic examination, and microscopic examination after formol ether concentration technique. *Entamoeba* spp. and *Dientamoeba fragilis* suspected specimens were stained with Wheatley trichrome staining method. The immunochromatographic rapid antigen test (*H. pylori* Antigen Rapid Test Cassette, All Test©, Germany) was studied by the kit insert for the detection of *H. pylori* antigen in stool.

Statistical Analysis

Statistical analysis was performed using SPSS 23 (IBM Inc, New York, USA) program. The chi-square test was used for categorical variables while making comparisons between groups. As a result of the evaluation, $p < 0.05$ value was considered statistically significant.

RESULTS

Of the 408 patients included in the evaluation, 159 (39%) were male and 249 (61%) were female. Of these patients, 41 (10%) were younger than 6 years old, 334 (81.9%) were between 6-18 years old, and 33 (8.1%) were older than 19 years old.

In 65 of the patients (15.9%), the *H. pylori* antigen test in the stool was positive. Of the patients with positive antigen test, 21 were (32.3%) male and 44 were (67.7%) female. There was no statistically significant difference between *H. pylori* positivity and gender ($p = 0.230$). Of the patients with positive antigen tests, 4 (6.2%) were younger than 6, 54 (83.1%) were between 6-18, and 7 (10.7%) were older than 19 years old. There was no statistically significant difference between *H. pylori* positivity and age groups ($p = 0.393$).

One or more intestinal parasites were found in 80 (19.6%) of the patients. 30 of the patients (37.5%) were male, and 50 (62.5%) were female. There was no statistically significant difference between intestinal parasite positivity and gender ($p = 0.764$). Of the patients with intestinal parasites, 4 (5%) were < 6, 74 (92.5%) were 6-18, 2 (2.5%) > 19 years old. The chance to find intestinal parasite was significantly higher in children age 6-18 ($p = 0.02$).

Blastocystis sp. was detected in 61 (15%), *Dientamoeba fragilis* in 3 (0.7%), *Giardia intestinalis* in 3 (0.7%), *Blastocystis* sp. + *Entamoeba* sp. in 5 (1.2%), *Blastocystis* sp. + *D. fragilis* in 7 (1.7%), and *Blastocystis* sp. + *G. intestinalis* in 1 (0.2%) patient.

Eight patients had both intestinal parasites and *H. pylori* antigen positivity in stool, and was not statistically significant ($p = 0.106$).

DISCUSSION

The prevalence of *H. pylori* may vary according to socioeconomic status, geographical region and age. In developing countries, the frequency in the general population is approximately 60-80% and it is widely accepted that *H. pylori* is acquired during childhood.^[8] Intestinal parasites remain an important public health problem especially in developing countries. Intestinal parasitic infections decrease quality of life and may increase susceptibility to other infections.^[9]

The increasing rates of *H. pylori* and IP co-infections in recent studies suggest that there may be a relationship in the pathogenesis of these infections. In a study conducted in Pakistan, stool samples from 161 patients with chronic diarrhea and 114 individuals without symptoms were evaluated for the presence of *H. pylori* and IP by molecular methods. One or more IP were detected in 27 (81.8%) of 33 patients with *H. pylori* in the patient group, and 17 (63%) of 27 patients with *H. pylori* in the control group. It was reported that *H. pylori* infection is more likely to be co-infected with *Blastocystis* sp. and *E. histolytica*.^[10] In our study, the higher detection rates of both *H. pylori* and IP were thought to be related to the sensitivity of the methods.

In a study conducted in Sudan, IP was detected in 23% of patients with *H. pylori* infection, and *E. histolytica* and *G. intestinalis* were found most frequently in these patients. The frequency of co-infection was statistically significant in the patient group compared to the control group.^[11] In a study conducted in Ethiopia, IP in stool samples and *H. pylori* IgG in serum samples were investigated in 363 adult patients with upper gastrointestinal system complaints. IP positivity was detected in stool samples of 44.3% of 225 patients with *H. pylori* IgG positivity. In this study, the most detected IP was *G. intestinalis* (22.3%) and the relationship with *H. pylori* IgG positivity was found to be statistically significant.^[6]

In another study conducted in Ethiopia, 23% of 434 school-age children showed *H. pylori* and IP co-infection, and *G. intestinalis* was detected most frequently, similar to the previous study.^[12] In a study conducted in Iran, co-infection with *G. intestinalis* was shown in 29.7% of patients with abdominal pain and *H. pylori* positivity. In another study conducted on children in Africa, it was reported that the probability of *H. pylori* co-infection is three times higher in people infected with *G. intestinalis*.^[13] In two separate studies conducted in Egypt, it was shown that the highest rate of *H. pylori* positivity was most frequently associated with *G. intestinalis* co-infection.^[14,15] In our study, however, *G. intestinalis* was seen in only 4 patients and *H. pylori* was not detected in any of these patients.

It is known that *H. pylori* and IP infections are more common in underdeveloped and developing countries, so the frequency of co-infection is higher in these regions. However, few data are indicating the frequency of co-

infection in developed countries. In a study conducted in Italy, IP was detected in 74% of the patients who were found to be positive for *H. pylori* by molecular methods, and *H. pylori* and *Blastocystis* sp. co-infection was found more prevalent.^[16] In a study conducted in China, the incidence of *H. pylori* was found to be positively correlated with *Blastocystis* sp. infection.^[13] In our study, although *Blastocystis* sp. was detected in 18.1% of the patients whose stool samples were examined, the coexistence of *H. pylori* and *Blastocystis* sp. was not statistically significant. In a study conducted in Iraq, it was found that *H. pylori* and *Blastocystis* sp. co-infection may have a synergistic effect on colorectal cancer.^[17] More studies are needed to understand the importance of the coexistence of *H. pylori* and *Blastocystis* sp.

There are very few studies investigating *H. pylori* and IP co-infection in our country. In the study of Uğraş et al., the frequency of IP was investigated in patients with histopathologically proven presence of *H. pylori*, and *Blastocystis* sp. was detected in 5.7% and *G. intestinalis* in 1.9% of the patients.^[18] In our study, *Blastocystis* sp. was present in 8 (12.3%) of *H. pylori* antigen positive stool samples.

One of the limitations of our study is that the sensitivity of the diagnostic methods is quite low compared to molecular methods. Due to the retrospective nature of the study, it was not possible to compare the data obtained from the patient group with healthy controls.

CONCLUSION

To better understand the relationship between *H. pylori* and IPs, there is a need for more comprehensive studies that include more patients, use high-sensitivity methods such as PCR, and evaluate healthy controls without any symptoms.

ETHICAL DECLARATIONS

Ethics Committee Approval: Ethics committee approval dated 10.11.2021 and numbered E-21-638 was obtained from Ankara Training and Research Hospital Ethical Committee of Non-Invasive Clinical Research.

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The author has no conflicts of interest to declare.

Financial Disclosure: The author declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Wong LW, Ong KS, Khoo JR, Goh CBS, Hor JW, Lee SM. Human intestinal parasitic infection: a narrative review on global prevalence and epidemiological insights on preventive, therapeutic and diagnostic strategies for future perspectives. *Expert Rev Gastroenterol Hepatol* 2020;14(11):1093-105.
2. Olopade BO, Idowu CO, Oyelese AO, Aboderin AO. Intestinal parasites, nutritional status and cognitive function among primary school pupils in Ile-Ife, Osun State, Nigeria. *Afr J Infect Dis* 2018;12(2):21-8.
3. Ostan I, Kilimcioğlu AA, Girginkardeşler N, Ozyurt BC, Limoncu ME, Ok UZ. Health inequities: lower socio-economic conditions and higher incidences of intestinal parasites. *BMC Public Health* 2007;7:342.
4. Lehours P. Actual diagnosis of *Helicobacter pylori* infection. *Minerva Gastroenterol Dietol* 2018;64(3):267-79.
5. Öztekin M, Yılmaz B, Ağagündüz D, Capasso R. Overview of *Helicobacter pylori* Infection: Clinical Features, Treatment, and Nutritional Aspects. *Diseases* 2021;9(4):66.
6. Seid A, Tamir Z, Kसानew B, Senbetay M. Co-infection of intestinal parasites and *Helicobacter pylori* among upper gastrointestinal symptomatic adult patients attending Mekanesalem Hospital, northeast Ethiopia. *BMC Res Notes* 2018;11(1):144.
7. Taghipour A, Bahadory S, Badri M, et al. A systematic review and meta-analysis on the co-infection of *Helicobacter pylori* with intestinal parasites: public health issue or neglected correlation?. *Int J Environ Health Res* 2022;32(4):808-18.
8. Maçın S, Demir H, Özen H, Yüce A, Akyön Y. Determination of *Helicobacter pylori* antibiotic resistance patterns in pediatric gastroenterology patients: the Hacettepe experience. *Turk J Pediatr* 2015;57:254-57.
9. Maçın S, Kaya F, Çağdaş D, et al. Detection of parasites in children with chronic diarrhea. *Pediatr Int.* 2016;58(6):531-3.
10. Yakoob J, Abbas Z, Khan R, Tariq K, Awan S, Beg MA. Association of *Helicobacter pylori* and protozoal parasites in patients with chronic diarrhoea. *Br J Biomed Sci* 2018;75(3):105-09.
11. Yousif Abd Elbagi Y, Abd Alla AB, Saad MBE. The relationship between *Helicobacter pylori* infection and intestinal parasites in individuals from Khartoum state, Sudan: a case-control study. *F1000Res* 2019;8:2094.
12. Spotts H, Walelign S, Tesfaye M, Desta K, Tsegaye A, Taye B. Concurrent infection of intestinal parasites and *Helicobacter pylori* among school-age children in Central Ethiopia. *Parasite Epidemiol Control* 2020;11:e00177.
13. Krzyżek P, Gościński G. Frequency and immunological consequences of *Helicobacter pylori* and intestinal parasite co-infections: a brief review. *Ann Parasitol* 2017;63(4):255-63.
14. Ghallab M, Morsy S. *Helicobacter pylori* co-infected with common intestinal protozoa in gastrointestinal symptomatic patients. *J Egypt Soc Parasitol* 2020;50(2):390-3.
15. Ibrahim A, Ali YBM, Abdel-Aziz A, El-Badry AA. *Helicobacter pylori* and enteric parasites co-infection among diarrheic and non-diarrheic Egyptian children: seasonality, estimated risks, and predictive factors. *J Parasit Dis* 2019;43(2):198-08.
16. Pomari E, Ursini T, Silva R, Leonardi M, Ligozzi M, Angheben A. Concomitant Infection of *Helicobacter pylori* and Intestinal Parasites in Adults Attending a Referral Centre for Parasitic Infections in North Eastern Italy. *J Clin Med* 2020;9(8):2366.
17. Majeed GH, Mohammed NS, Muhsen SS. The synergistic effect of *Blastocystis hominis* and *H. pylori* in Iraqi colorectal cancer patients. *J Pharm Sci Res* Vol 2019;11(2):523-26.
18. Uğraş M, Miman O. The prevalence of intestinal parasites in children with *Helicobacter pylori* gastritis evaluated retrospectively. *Turkiye Parazitol Derg* 2013;37(4):245-48.