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The prevalence of Helicobacter pylori and its effect on the prognosis of patients with COVID-19

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ABSTRACT

Objective: SARS-CoV-2 RNA positivity in stool in COVID-19 infection has been reported at rates varying between 6-83%. The purpose of this study was to determine the prevalence of H.pylori and investigate whether it determines the disease prognosis in COVID-19 patients.

Methods: This study was conducted on 117 confirmed COVID-19 patients who were hospitalized due to symptomatic pneumonia and tested for stool H.pylori antigen. Stool H. pylori test outcomes, demographic parameters, laboratory findings, and prognostic predictors of disease were recorded. The effect of the presence of H.pylori in patients with COVID-19 was analyzed.

Results: The mean age of 117 included patients was 49.68 ± 14.62 years, 78 (66.7%) had COVID PCR positivity and 32 (27.35%) had H.pylori positivity. There was no statistical difference in demographic data, prognosis, and laboratory parameters between those with and without H.pylori.

Conclusion: H.pylori positivity was detected as 27.35% in patients with COVID-19 infection. However, we could not find the positive or negative effect of H.pylori on the prognosis of COVID-19 disease. In conclusion, according to the results of this study, H. pylori positivity or negativity neither determined the severity of the COVID-19 disease nor the poor prognostic indicators of the disease.

Keywords: H. pylori, COVID-19, prognosis

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INTRODUCTION

New coronavirus (COVID-19, SARS-CoV-2) has become a pandemic that has spread all over the world, starting in China in 2019, causing severe acute respiratory failure (1). New information is being added every day about the characteristics and treatment of COVID-19 disease. However, our knowledge of COVID-19 infection and its treatment is still limited.

SARS-CoV-2 RNA positivity in stool in COVID-19 infection has been reported at rates varying between 6-83% (2-5). Detection of viral genetic material in the stool does not mean that viable infectious virions are present in the stool, but a long-term positive gastrointestinal specimen is interpreted as the virus can replicate actively in the patient's gastrointestinal system (6). ACE-2 receptors used by SARS-CoV-2 to enter the cell are highly expressed in the small intestine and the binding affinity of ACE affects the infectivity of the virus. A number of viruses, such as coronavirus, rotavirus, and noroviruses, can invade absorbent enterocytes through the ACE-2 receptors on absorbent enterocytes in the ileum and colon, causing gastroenteritis.

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There have been studies showing that intestinal epithelial cells expressing ACE-2 may be at increased risk of attack by SARS-CoV-2 and that ACE2 is highly expressed in the small intestine, especially in proximal and distal enterocytes. Therefore, the digestive system can be invaded by SARS-CoV-2 and used as an entrance of infection. It has been shown that approximately 12% of patients with SARS-CoV-2 infection have gastrointestinal symptoms including diarrhea, nausea and vomiting (7). In the meta-analysis examining the studies investigating whether the gastrointestinal system symptoms are associated with mortality in COVID-19 disease; no significant difference was shown between groups with and without gastrointestinal symptoms (8).

H.pylori is one of the most common infections affecting the human race with a high prevalence in developing countries (9). With respect to the distribution map of COVID-19 cases and mortality rates in different countries, mortality rates per million population are low in Russia, Portugal (https://www.worldometers.info/coronavirus/) whereas higher in these regions (Russia 78.5%, Portugal 86.4%). Therefore, it is assumed that H. pylori may play a role in preventing serious infections in COVID-19 infection (9,10).

Conflicting results have been reported between non-COVID viral infections and H. pylori positivity in the literature (11-13).

In our literature searches, we did not find any studies investigating COVID-19 and H. pylori infection together. To our knowledge, we have investigated for the first time the determination of the prevalence of H. pylori in patients with confirmed COVID-19 disease and whether it determines the prognosis of COVID-19 disease.

MATERIAL and METHODS

Study population and sample collection

A total of 117 patients, whose diagnosis of COVID-19 pneumonia was confirmed by nasopharyngeal (NP) PCR-RT swab and thorax computed tomography between 15 March 2020 and July 2020, were included in the study. Stool H. pylori antigen test was investigated in all patients. Patients with a history of H. pylori eradication therapy, a history of gastric malignancy and NP PCR-RT negative patients were excluded from the study. PCR results of the cases, H. pylori test results, demographic parameters, other laboratory parameters and prognosis indicators were recorded. The effect of the presence of H. pylori in patients followed-up for COVID-19 infection was statistically analyzed.

H. pylori antigen test in faeces

Stool samples from patients with confirmed diagnosis of COVID-19 disease were taken into special stool containers and transferred to the laboratory. H. pylori antigen test [H.pylori Antigen Rapid Test Device (feces), China] was studied in stool samples by immunochromatographic method. After all the reagents and stool samples reached room temperature, the applicator stick coming out of the kit was dipped into three different areas of each stool sample and approximately 50mg sample was collected. After placing the applicator stick in the diluent tube and closing its cap, it was shaked. The results were interpreted after 10 minutes of dropping 3 drops of the immunochromatographic cassette from the liquid sample in the tube to the sample section of the test. The formation of a colored line, even faint, was considered positive in patients with a control line showing the validity of the test. The whole procedure has been done in line with the manufacturer's recommendations.

It has been reported that the use of H. pylori antigen test in stool is reliable as a non-invasive test (14-16).

Nucleic acid isolation and SARS-CoV 2 RT-PCR

Combined nasopharynx and oropharynx swab samples were collected by dacron swab and placed in Viral transport medium immediately, and delivered to the laboratory by keeping them at 2-8 °C. The samples were delivered to the laboratory in accordance with the rules of cold chain with the triple transport system, complying the infection prevention and control procedures.

After taking the samples in microbiology laboratory, samples were taken to a negative pressure chamber with 3rd level bio-safety. Bio-Speedy® Viral Nucleic Acid Isolation Kit (Bioeksen, Turkey) was used for total nucleic acid isolation from the specimens. The isolation procedure was carried out according to the recommendations of the manufacturer.

Bio-Speedy® COVID-19 RT-qPCR Detection Kit (Bioeksen, Turkey) was used for the RT-PCR assays. The PCR amplification and evaluation of the results were carried out according to the recommendations of the manufacturer.

Statistical Analysis

Data analysis was performed by using IBM SPSS Statistics version 21.0 software (IBM Corporation, Armonk, NY, USA). Whether the distributions of continuous variables were normally or not being determined by visual (histograms, probability plots) and analytical methods (Kolmogorov-Smirnov and Shapiro Wilk tests). Descriptive analyses were presented using means and standard deviations for normally distributed variables; using medians and and 1st-3rd quarter for the non-normally distributed variables. Categorical variables are specified as numbers and percentages. Independent-Samples T test, Mann-Whitney U test, Chi Square test and Fisher's Exact test were used to analyze the data. The significance level for all of the statistical tests was set at $p<0.05$.

RESULTS

The mean age of 117 patients with COVID-19 pneumonia was 49.68 ± 14.62 years and 48.7% were male. H. pylori was detected in 32 (27.35%) of the patients. In terms of COVID-19 PCR-RT positivity, there was no significant difference between the H.pylori positive group (Group 1) and negative group (Group 2) ($p = 0.769$).

Demographic characteristics of both groups were similar (Table 1). During the follow-up period, no patient died in Group-1, while 4 (4.7%) patients in group-2 died, but there was no significant difference between the two groups ($p = 0.574$). Concerning prognostic markers levels such as white blood count (WBC), neutrophil, lymphocyte, thrombocyte, D-Dimer, ferritin, albumin, lactate dehydrogenase (LDH), procalcitonin and CRP were similar in both groups ($P> 0.05$) (Table 2).

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Table 1: Distribution of demographic and clinical characteristics according to H. pylori status in patients with COVID-19

Parameters	All patient (n=117)	Stool H. pylori + (n=32) (%27.35)	Stool H. pylori – (n=85) (%72,65)	p
PCR + n(%)	78 (66.7)	22 (68.8)	56 (65.9)	0.769*
Age (Years) Mean ± SD Median(1st-3rd quarter)	49.68±14.62 47.00 (40.00-60.00)	46.78±13.80 45.50 (37.00-57.00)	50.78±14.85 48.00 (40.50-60.50)	0.189
Sex n (%)				
Male	57 (48.7)	13 (40.6)	47 (55.3)	0.157*
Female	60 (51.3)	19 (59.4)	38 (44.7)	
Having a Chronic illness + n (%)	64 (54.7)	15 (46.9)	49 (57.6)	0.297*
No chronic disease n(%)	53 (45.3)	17 (53.1)	36 (42.4)	
One chronic disease n(%)	45 (38.5)	11 (34.4)	34 (40.0)	
Two chronic disease n(%)	16 (13.7)	4 (12.5)	12 (14.1)	
Three or more chronic disease n(%)	3 (2.6)	-	3 (3.5)	
Diabetes Mellitus n (%)	17 (14.5)	4 (12.5)	13 (15.3)	1.000**
Hypertension n (%)	46 (39.3)	9 (28.1)	37 (43.5)	0.128*
Coronary artery disease n (%)	4 (3.4)	1 (3.1)	3 (3.5)	1.000**
Chronic obstructive Pulmonary disease n(%)	3 (2.6)	-	3 (3.5)	0.561**
Asthma n (%)	7 (6.0)	4 (12.5)	3 (3.5)	0.088**
Blood groups				
A Rh(+)	22 (38.6)	7 (38.9)	15 (38.5)	
B Rh (+)	8 (14.0)	2 (11.1)	6 (15.4)	-
O Rh (+)	20 (35.1)	7 (38.9)	13 (33.3)	
AB Rh (+)	3 (5.3)	-	3 (7.7)	
A Rh (-)	3 (5.3)	2 (11.1)	1 (2.6)	
O Rh (-)	1 (1.8)	-	1 (2.6)	
Oncet of complaintsn (%)				
Cough n (%)	90 (76.9)	23 (71.9)	67 (78.8)	0.427*
Fever n (%)	39 (33.3)	8 (25.0)	31 (36.5)	0.241*
Shortness of breath n (%)	36 (30.8)	9 (28.1)	27 (31.8)	0.704*
Throat ache (%)	31 (26.5)	8 (25.0)	23 (27.1)	0.822*
Diarrhea n (%)	10 (8.5)	5 (15.6)	5 (5.9)	0.134**
Vomiting n (%)	3 (2.6)	3 (9.4)	-	**0.019****
Anosmia n (%)	2 (1.7)	1 (3.1)	1 (1.2)	0.474**
Headache n (%)	2 (1.7)	1 (3.1)	1 (1.2)	0.474**
Support in/Antiviral treatments n (%)				
Hydroxyqloroqine	117 (100)	32 (100)	85 (100)	-
Oseltamivir	117 (100)	32 (100)	85 (100)	-
Favipiravir	19 (16.2)	2 (6.3)	17 (20.0)	0.072*
Azithromycin	110 (94.0)	29 (90.6)	81 (95.3)	0.390**
Antibacterials	25 (21.4)	6 (18.8)	19 (22.4)	0.672*
Radiolojic involvement n (%)				
Bilateral	97 (83.6)	25 (78.1)	72 (84.7)	0.601*
Unilateral	19 (16.4)	6 (18.8)	13 (15.3)	
Mortality n(%)	4 (3.4)	-	4 (4.7)	0.574**

* Chi Square test, ** Fisher's Exact test, IQR =Interquartile range

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Table 2. Distribution of laboratory values according to H. pylori status in Patients with COVID-19

Parameters	All patient	H. pylori +	H. pylori -	p
WBC (10^3*µl) Median(1st-3rd quarter)	6.64 (4.47-8.59)	6.99 (4.71-8.57)	6.32 (4.44-8.68)	0.523*
Lymphocyte (10^3*µl) (Mean±SD)	1.68±0.79	1.71±0.88)	1.67±0.75	0.808**
Neutrophil (10^3*µl) Median(1st-3rd quarter)	3.89 (2.69-5.69)	4.00 (2.80-6.46)	3.89 (2.64-5.69)	0.709*
Platelet (10^3*µl) Median(1st-3rd quarter)	172.00(142.00-225.50)	172.00 (139.25-224.25)	172.00 (145.00-225.50)	0.976*
D-Dimer (ng/mL) Median(1st-3rd quarter)	429.00(283.50-987.75)	378.00 (280.00-714.00)	482.00 (280.00-1047.00)	0.315*
Ferritine (10^3*µl) Median(1st-3rd quarter)	221.00(100.50-543.50)	128.50 (66.70-542.00)	237.00 (125.00-543.50)	0.082*
Albumin (g/L) Median(1st-3rd quarter)	34.25 (32.68-37.65)	35.50 (32.20-37.83)	34.05 (32.68-37.50)	0.551*
LDH (U/L) Median(1st-3rd quarter)	269.00(212.50-354.50)	249.00 (195.50-332.25)	274.00 (219.50-359.00)	0.253*
Procalcitonine (ng/mL) Median(1st-3rd quarter)	0.05 (0.02-0.26)	0.06 (0.02-0.47)	0.05 (0.02-0.20)	0.762*
CRP (mg/L) Median(1st-3rd quarter)	23.30 (4.70-76.90)	20.25 (3.12-78.63)	25.80 (5.31-76.90)	0.446*

* Mann-Whitney U test, ** Independent-Samples T test, IQR =İnterquartile range, WBC: White blood cell. LDH: lactate dehydrogenase. CRP: C reactive protein.

DISCUSSION

To our knowledge, there is no study investigated the frequency of H. pylori and the prognostic significance of H. pylori positivity in patients with COVID-19 disease. In the present study, we determined a 27.35% of H.pylori positivity in patients with COVID-19. Our outcomes are much lower than the rates in the normal and non-COVID viral infectious population. The prevalence of H.pylori, which has lower rates in studies conducted in European countries, varies between 7-87% worldwide (**17**). In a recently published systematic meta-analysis, the mean positive frequency of H.pylori antigen in stool was found to be 49.4% 9. In a study conducted in our country, a prevalence rate of 25.2% was reported in stool H.pylori antigen test studies in the normal population (18).

Studies on the frequency of H. pylori in various non-COVİD-19 viral infections have been identified. For instance; information from epidemiologic works suggest that the frequency of H. pylori infection is clearly lower in HIV-positive compared with the HIV-negative population and that it further declines with the progression of immunodeficiency in HIV-infected patients. In a study of 1095 HIV-positive and 107 HIV-negative patients using the stool antigen test, the prevalence of H. pylori was significantly higher in the HIV-negative group (51.5% vs 88%, respectively, p< 0.05) (11).

In a meta-analysis evaluated 29 studies; all studies, except one study, reported a higher rate of H. pylori infection in HIV negative subjects (**12**). In contrast, another study found a higher prevalence of H. pylori resistant strain in HIV-positive patients than in HIV-negative patients (**13**). In addition, a meta-analysis including 2977 chronic hepatitis B and 1668 control patients, the H. pylori prevalence was found to be higher in patients with chronic hepatitis B positive. Also, the incidence of H. pylori has been shown to be positively correlated with HBV-associated hepatocellular carcinoma (**19**). However, the H. pylori prevalence in 235 asymptomatic HBV carriers, 573 alcohol users and 1637 non-alcoholic individuals was similar as 38.67%, 26.98%, and 35.94%, respectively (**20**).

Recently, the importance of gastrointestinal microbiota as a decisive for the systemic immune response has been recognized, and a number of extraintestinal, immune-related efficacious of H. pylori positivity have been reported (**21-22**).

With respect to the association between the immune system hypoactivation and H. pylori coinfection, it is known that H. pylori decrease markers (HLA DR, CD38, CD4) of the immune activation system by decreasing T-cell activation in HIV-positive and in HIV-negative individuals. This finding might explain the association of H. pylori infection in the intestine with favorable parameters of HIV disease progression (**23**). The immune response to H. pylori infection is predominantly T-cell mediated. It has been shown that the H. pylori vacuolating toxin directly inhibits T-cell activation by interfering with the maturation of antigen present cells and dendritic cells (**24**). It is known that lymphopenia is frequently reported in patients with COVID-19 and is considered to be a determinant of the prognosis of the disease. However, in our study, with respect to lymphopenia, no significant relationship was shown between the H.pylori positive patient group and the negative group. Various poor prognostic markers such as WBC, neutrophil, D-Dimer, ferritin, LDH, procalcitonin, high CRP, thrombocytopenia, and hypoalbuminemia have been demonstrated in COVID-19 infection (**25**). In our study, no significant difference was found in terms of these prognostic markers between groups with and without H. pylori positive. The most important limitation of the present study was that the immune T lymphocyte activation parameters were not detected in H. pylori positive COVID-19 patients.

CONCLUSION

In conclusion, according to the results of this study, H. pylori positivity or negativity neither determined the severity of the COVID-19 disease nor the poor prognostic indicators of the disease. However, larger and controlled studies are needed to confirm these findings.

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REFERENCES

1. Phelan AL, Katz R, Gostin LO. The Novel Coronavirus Originating in Wuhan, China: Challenges for Global Health Governance. JAMA: the journal of the American Medical Association [Internet] 2020;Available from: http://dx.doi.org/10.1001/jama.2020.1097

2. Li Y, Hu Y, Yu Y et al. Positive result of Sars-Cov-2 in faeces and sputum from discharged patients with COVID-19 in Yiwu, China. Journal of medical virology [Internet] 2020;Available from: http://dx.doi.org/10.1002/jmv.25905

3. Guan W-J, Ni Z-Y, Hu Y et al. Clinical Characteristics of Coronavirus Disease 2019 in China. The New England journal of medicine 2020; 382: 1708.

4. Szymczak WA, Yitzchak Goldstein D, Orner EP et al. Utility of Stool PCR for the Diagnosis of COVID-19: Comparison of Two Commercial Platforms [Internet]. Journal of Clinical Microbiology2020; 58. Available from: http://dx.doi.org/10.1128/jcm.01369-20

5. Mesoraca A, Margiotti K, Viola A, Cima A, Sparacino D, Giorlandino C. Evaluation of SARS-CoV-2 viral RNA in fecal samples. Virology journal 2020; 17: 86.

6. van Doorn AS, Meijer B, Frampton CMA, Barclay ML, de Boer NKH. Systematic review with meta-analysis: SARS-CoV-2 stool testing and the potential for faecal-oral transmission. Alimentary pharmacology & therapeutics [Internet] 2020;Available from: http://dx.doi.org/10.1111/apt.16036

7. Parasa S, Desai M, Thoguluva Chandrasekar V et al. Prevalence of Gastrointestinal Symptoms and Fecal Viral Shedding in Patients With Coronavirus Disease 2019: A Systematic Review and Meta-analysis. JAMA network open 2020; 3: e2011335.

8. Tariq R, Saha S, Furqan F, Hassett L, Pardi D, Khanna S. Prevalence and Mortality of COVID-19 Patients With Gastrointestinal Symptoms: A Systematic Review and Meta-analysis. Mayo Clinic proceedings. Mayo Clinic 2020; 95: 1632.

9. Hooi JKY, Lai WY, Ng WK et al. Global Prevalence of Helicobacter pylori Infection: Systematic Review and Meta-Analysis. Gastroenterology 2017; 153: 420.

10. Jordan RE, Adab P, Cheng KK. Covid-19: risk factors for severe disease and death. BMJ 2020; 368: m1198.

11. Sarfo FS, Eberhardt KA, Dompreh A et al. Helicobacter pylori Infection Is Associated with Higher CD4 T Cell Counts and Lower HIV-1 Viral Loads in ART-Naïve HIV-Positive Patients in Ghana. PloS one 2015; 10: e0143388.

12. Nevin DT, Morgan CJ, Graham DY, Genta RM. Helicobacter pylori gastritis in HIV-infected patients: a review. Helicobacter 2014; 19: 323.

13. Nkuize M, De Wit S, Muls V et al. HIV-Helicobacter pylori Co-Infection: Antibiotic Resistance, Prevalence, and Risk Factors. PloS one 2015; 10: e0145119.

14. Okuda M, Osaki T, Kikuchi S et al. Evaluation of a stool antigen test using a mAb for native catalase for diagnosis of Helicobacter pylori infection in children and adults. Journal of medical microbiology 2014; 63: 1621.

15. Makristathis A, Barousch W, Pasching E et al. Two Enzyme Immunoassays and PCR for Detection ofHelicobacter pylori in Stool Specimens from Pediatric Patients before and after Eradication Therapy [Internet]. Journal of Clinical Microbiology2000; 38: 3710. Available from: http://dx.doi.org/10.1128/jcm.38.10.3710-3714.2000

16. Erzin Y, Altun S, Dobrucali A et al. Comparison of two different stool antigen tests for the primary diagnosis of Helicobacter pylori infection in turkish patients with dyspepsia. Helicobacter 2004; 9: 657.

17. Ford AC, Axon ATR. Epidemiology of Helicobacter pylori infection and Public Health Implications [Internet]. Helicobacter2010; 15: 1. Available from: http://dx.doi.org/10.1111/j.1523-5378.2010.00779.x

18. Demir T, Turan M, Tekin A. Kırşehir bölgesindeki dispeptik hastalarda Helicobacter pylori antijen prevalansı, Dicle Tıp Derg 2011;38(1):44-8).

19. Wang J, Chen R-C, Zheng Y-X et al. Helicobacter pylori infection may increase the risk of progression of chronic hepatitis B disease among the Chinese population: a meta-analysis. International journal of infectious diseases: IJID: official publication of the International Society for Infectious Diseases 2016; 50: 30.

20. Wang M-Y, Yue J-Y, Zhang Y-X, Liu X-D, Gao X-Z. Helicobacter pylori infection in asymptomatic HBV carriers, alcohol users and normal adult population in Shandong Province, China. Clinics and research in hepatology and gastroenterology 2011; 35: 560.

21. Wong F, Rayner-Hartley E, Byrne MF. Extraintestinal manifestations of Helicobacter pylori: a concise review. World journal of gastroenterology: WJG 2014; 20: 11950.

22. Alarcón T, Llorca L, Perez-Perez G. Impact of the Microbiota and Gastric Disease Development by Helicobacter pylori. Current topics in microbiology and immunology 2017; 400: 253.

23. Eberhardt KA, Sarfo FS, Dompreh A et al. Helicobacter pylori Coinfection Is Associated With Decreased Markers of Immune Activation in ART-Naive HIV-Positive and in HIV-Negative Individuals in Ghana. Clinical infectious diseases: an official publication of the Infectious Diseases Society of America 2015; 61: 1615.

24. Alzahrani S, Lina TT, Gonzalez J, Pinchuk IV, Beswick EJ, Reyes VE. Effect of Helicobacter pylori on gastric epithelial cells. World journal of gastroenterology: WJG 2014; 20: 12767.

25. Henry BM, de Oliveira MHS, Benoit S, Plebani M, Lippi G. Hematologic, biochemical and immune biomarker abnormalities associated with severe illness and mortality in coronavirus disease 2019 (COVID-19): a meta-analysis. Clinical chemistry and laboratory medicine: CCLM / FESCC 2020; 58: 1021.