

Relationship between the severity of *Helicobacter pylori* infection and neutrophil and lymphocyte ratio and mean platelet volume in children

Associate Professor^a, Yasin Sahin, M.D.^a, Ozlem Gubur, M.D.^b and Emine Tekingunduz, M.D.^b

ABSTRACT

Introduction: HP causes local inflammation in the stomach and a systemic humoral immune response. No relationship was found in adult studies investigating the association between HP infection and neutrophil/lymphocyte ratio (NLR) and mean platelet volume (MPV). To our knowledge, there is no study in children. We aimed to evaluate the association between NLR and MPV values with HP infection, severity classification, and pre- and post-treatment status.

Population and Methods: Patients with dyspepsia aged between 2-18 years and without any chronic diseases undergoing gastroduodenoscopy were included in the study. Endoscopic mucosal biopsy was performed, HP presence and infection severity were determined according to Sydney Classification.

Results: 153 patients with HP positivity and 211 patients with HP negativity were included in the study, the mean age of them was 13.3 ± 3.4 years and 13.1 ± 3.5 years, respectively. No statistically significant difference was found between patients with HP positive and negative patients and also between severity subgroups of HP positive patients in terms of NLR and MPV ($p > 0.05$).

Conclusions: There was no association between NLR and MPV values with HP infection, severity classification, or pre- and post-treatment status.

Key words: child, endoscopy, *Helicobacter pylori*, mean platelet volume, neutrophil / lymphocyte ratio.

<http://dx.doi.org/10.5546/aap.2020.eng.e241>

To cite: Sahin Y, Gubur O, Tekingunduz E. Relationship between the severity of *Helicobacter pylori* infection and neutrophil and lymphocyte ratio and mean platelet volume in children. *Arch Argent Pediatr* 2020;118(3):e241-e245.

- a. Clinics of Pediatric Gastroenterology, Mersin City Training and Research Hospital, Mersin, Turkey.
- b. Department of Pathology, Mersin City Training and Research Hospital, Mersin, Turkey.

E-mail address:
Yasin Sahin, M.D.:
ysahin977@gmail.com

Funding:
None.

Conflict of interest:
None.

Received: 6-7-2019
Accepted: 11-25-2019

INTRODUCTION

Helicobacter pylori (HP) is one of the most common chronic bacterial infections in the world. More than half of the world's population has HP colonization.¹ The seroprevalence rate of HP was estimated to be 33 % in 21 meta-analysis studies performed in children between 2011 and 2016. In addition to that, this rate is reported to be approximately 40 % in symptomatic children.²

It is a common microorganism found with a frequency of 80 % in developing countries. HP causes chronic inflammation in the stomach and elicits immune response.³ Most of the patients are usually asymptomatic.

HP tends to settle in the antrum which is less acidic. HP attracts neutrophils and lymphocytes. Some substances released from neutrophils and mononuclear cells cause mucosal inflammation and thus cause gastritis. As a result, in addition to many cytokines, neutrophils, macrophages and lymphocytes infiltrate gastric mucosa and subclinical systemic low grade inflammation occurs.^{3,4}

It is the most common cause of chronic gastritis and has a high morbidity. HP is particularly associated with severe gastric diseases such as gastric cancer, gastric infection, peptic ulcer, and chronic gastritis.⁵

The increase in blood leukocytes and especially neutrophils is commonly associated with inflammation.⁶ Neutrophil lymphocyte ratio (NLR) is obtained from a simple hemogram and is used as an inflammatory marker in various diseases. NLR has been proposed as an independent predictor

in various clinical problems from cardiovascular events to cancer.^{7,8}

Increased NLR has been shown to be a poor prognosis marker in patients undergoing cardiovascular intervention, and it has been shown to be high in acute coronary syndromes with high NLR.⁹

As a result of HP antibodies cross-reacting with platelet glycoproteins, it has been suggested that there is a decrease in platelet count and consequently immune thrombocytopenia (ITP) develops. Positive response in platelet counts is seen in patients with ITP after HP eradication.^{10,11} The patients with low grade inflammation, such as coronary artery disease, and diabetes mellitus has been shown to have higher MPV values.¹²

There is limited study investigating the association between HP infection and NLR and MPV in adults.^{10,13-15} No significant difference was found between patients with HP positivity and negativity in regards to NLR and MPV in adults with HP infection.^{10,14,15} It has been shown that higher NLR values were associated with severity of HP gastritis in adults.¹³

No relationship was found in adult studies investigating the association between HP infection and neutrophil/lymphocyte ratio (NLR) and mean platelet volume (MPV). To our knowledge, there is no study in children. We aimed to evaluate the association between NLR and MPV values with HP infection, severity classification, and pre- and post-treatment status.

POPULATION AND METHODS

The current study was conducted between March 2017 and October 2018.

Inclusion criteria

Patients with dyspepsia aged between 2-18 years and without any chronic diseases undergoing gastroduodenoscopy were included in the study.

Exclusion criteria

Patients with any organic disease, systemic disease, and infection were excluded from the study because any concomitant disease could change neutrophil and lymphocyte numbers. Patients with intestinal metaplasia or atrophy on endoscopic mucosal biopsies were also excluded from the study. Also, non-compliant patients and patients with treatment failure were excluded from the study.

Of the 390 patients, 364 patients with dyspeptic complaints who were followed up at the Clinics of Pediatric Gastroenterology and underwent gastroduodenoscopy, were included in the study.

At least 4 biopsies including 2 antrum and 2 corpus were obtained from patients. The biopsies were evaluated according to Sydney Classification.^{16,17}

All endoscopic mucosal biopsy specimens were stained with hematoxylin and eosin (HE), and HE-stained preparations were evaluated histopathologically and for the presence of *Helicobacter pylori* according to Sydney Classification. Patients with HP positivity were detected and included as patient group in the study. They were divided into three groups as mild, moderate and severe according to Sydney Classification. Patients with gastritis without HP infection, were included as a control group. Hemogram test was performed from peripheral blood before endoscopy. White blood cell count, neutrophil, lymphocyte and platelet counts were obtained from hemogram. Neutrophil / lymphocyte ratio was calculated by dividing absolute neutrophil count to absolute lymphocyte count. Because this study is retrospective, a written informed consent was not obtained. The Ethics Committee of Mersin University approved the current study (03 October 2018-385).

First-line triple therapy (including PPI, amoxicillin and metronidazole) for 14 days were given to the patients according to the latest ESPGHAN / NASPGHAN guidelines.¹⁶

Non-compliance is a very important factor in the treatment failure, so non-compliant patients were excluded from the study.¹⁸

The efficacy of the HP eradication treatment was assessed by a two-step monoclonal stool HP antigen test one month after the end of treatment.

Statistical analysis

Statistical analysis was performed using SPSS software version 13.0 (SPAA Inc, Chicago IL, USA). Frequency, percentage, and mean \pm standard deviation (SD) were used as descriptive statistics. The Mann-Whitney U test was used to compare groups of numerical variables, and the relationships between categorical variables were analyzed by chi-square test. Comparisons among the groups were performed using Kruskal-Wallis one-way analysis of variance test for five independent groups. *P* value was considered statistically significant as a < 0.05 .

RESULTS

During the current study, 840 patients with dyspepsia were followed-up at the Outpatient Clinics of Pediatric Gastroenterology. Gastroduodenoscopy was performed in 390 patients. Of 390 patients, 17 patients did not meet the inclusion criteria: 7 patients with celiac disease, 5 patients with diabetes mellitus, 3 patients with nonsteroidal anti-inflammatory drug gastropathy, one patient with hypothyroidism, and one patient with ataxia telangiectasia. After histopathological

examination, 4 patients with intestinal metaplasia were also excluded from the study. These patients were excluded from the study because any concomitant disease and nonsteroidal anti-inflammatory drug use could change neutrophil and lymphocyte numbers. Of 390 patients, 364 patients were included in the study.

Of the 364 patients, the mean age of the 153 patients (103 girls) in the patient group was 13.3 ± 3.4 years and the mean age of the 211 patients in the control group (157 girls) was 13.1 ± 3.5 years (Table 1). There was no significant

TABLE 1. Demographic and laboratory characteristics of patients and controls

	Patients (n = 153)	Controls (n = 211)	<i>p</i>
Age (yrs) [†]	13.30 ± 3.46	13.12 ± 3.52	0.627
WBC [†]	7.47 ± 2.00	7.67 ± 2.40	0.390
Hemoglobine (g/dL) [†]	12.87 ± 2.37	12.79 ± 1.25	0.702
MPV [†]	7.87 ± 0.73	7.87 ± 0.66	0.995
Neutrophil count [†]	4.24 ± 1.61	4.39 ± 2.09	0.452
Lymphocyte count [†]	2.36 ± 0.71	2.34 ± 0.79	0.812
NLR [‡]	1.76 (1.12)	1.76 (1.22)	0.093

WBC: white blood cells; MV: mean platelet volume; NLR: neutrophil lymphocytes ratio.

[†]Data are presented as mean ± standard deviation.

[‡]Data are presented as median (interquartile range).

TABLE 2. Pre and post-treatment values of patients with HP positivity

	Pre-treatment (n = 88)	Post-treatment (n = 88)	<i>p</i>
WBC [†]	7.74 ± 2.15	7.16 ± 2.08	0.013
MPV [†]	7.81 ± 0.72	7.87 ± 0.79	0.380
Neutrophil count [†]	4.44 ± 1.71	4.01 ± 1.90	0.064
Lymphocyte count [†]	2.40 ± 0.68	2.32 ± 0.61	0.195
NLR [‡]	1.83 (1.26)	1.60 (0.98)	0.342

WBC: white blood cells; MV: mean platelet volume; NLR: neutrophil lymphocytes ratio.

[†]Data are presented as mean ± standard deviation.

[‡]Data are presented as median (interquartile range).

TABLE 3. Comparison of HP subgroups in regards to degree of HP severity

	Mild form (n=73)	Moderate form (n=55)	Severe form (n=25)	<i>p</i>
WBC [†]	7.21 ± 2.19	7.69 ± 1.76	7.73 ± 1.91	0.315
MPV [†]	7.82 ± 0.69	7.95 ± 0.83	7.81 ± 0.64	0.551
Neutrophil count [†]	4.01 ± 1.75	4.46 ± 1.35	4.44 ± 1.69	0.234
Lymphocyte count [†]	2.35 ± 0.74	2.35 ± 0.68	2.42 ± 0.69	0.913
NLR [‡]	1.62 (1.17)	2.00 (0.98)	1.95 (1.17)	0.732

WBC: white blood cells; MV: mean platelet volume; NLR: neutrophil lymphocytes ratio.

[†]Data are presented as mean ± standard deviation.

[‡]Data are presented as median (interquartile range).

difference in terms of age and gender between the two groups ($p > 0.05$). No statistically significant difference was found between patients with HP positive and negative patients in terms of NLR and MPV ($p > 0.05$) (Table 1). When pre and post treatment values of NLR ratio and MPV of 88 patients treated were compared, we did not find any significant difference between them ($p > 0.05$) (Table 2). There was no statistically significant difference between subgroups of HP positive patients (mild, moderate and severe) in terms of MPV and NLR rates ($p > 0.05$) (Table 3).

DISCUSSION

More than half of the world's population is still infected with HP. Approximately 4.4 billion people were reported to be infected worldwide in 2015.¹ The prevalence of HP is high in developing countries and is often associated with socio-economic level and hygiene situation.¹

The leukocytes and its subgroups, and NLR have been shown to be indicators of systemic inflammation in previous study.¹⁹

There is limited study investigating the association between HP infection and NLR and MPV.^{10,13-15}

Guclu M et al.¹⁵ did not find any significant difference between patients with HP positivity and negativity in regards to NLR and MPV. The lymphocyte and thrombocyte values were within the normal range in patients with HP positivity, but were significantly higher than in patients with HP negativity. The reason for this increase is probably the increase in absolute lymphocyte levels. Also, the number of cases with severe HP gastritis was low. Authors suggested that studies with higher number of severe HP gastritis are needed. As compatible with other study, there was no any changes in MPV values in HP positive patients.¹⁰

In a study including 50 HP positive and 50 HP negative patients, the leukocyte, lymphocyte and neutrophil counts were found higher in HP positive patients than HP negative patients.¹³ Higher NLR values were also detected in HP positive patients. In addition, higher NLR values were associated with severity of gastritis and increased symptoms. HP negative patients had significantly lower NLR levels. It has been shown that higher NLR values returned to normal levels after successful treatment and eradication. The authors suggested that NLR can be used in the follow-up of patients after successful treatment. In contrast to this study, although we have 3 times

higher patients than Farah et al.¹³ study, we did not detect higher NLR in patients with HP positive. As compatible with this study, we found a decrease in NLR after HP eradication treatment, but this is not statistically significant. Further studies with more HP infected patients are needed to be confirmed.

In Jakarzadeh et al.¹⁴ study, mean leukocyte count, neutrophil count and NLR were significantly higher in patients with HP positivity and asymptomatic group than control group.¹⁴ The mean leukocyte count, neutrophil count and NLR were significantly different between the asymptomatic group and HP positive group. In addition to that, no difference was found between the 3 groups in terms of lymphocyte counts. The authors suggested that higher leukocyte and neutrophil counts in the asymptomatic group are probably due to subclinical microinflammatory reactions caused by HP.

In the current study, no significant difference was detected between patients with HP positivity and negativity in regards to NLR and MPV. There was no statistically significant difference between subgroups of HP positive patients (mild, moderate and severe) in regards to MPV and NLR rates ($p > 0.05$). When pre and post-treatment values of NLR and MPV of 88 patients who received HP eradication treatment were compared, we also did not find any significant difference between them ($p > 0.05$).

It has been shown a decrease in neutrophil counts after HP eradication in a study conducted in Japan.²⁰ As consistent with this study, we also detected a decrease in neutrophil counts, but these differences were not statistically significant.

As compatible with previous two studies, we found that NLR and MPV values did not correlate with the severity of HP infection in children.^{10,15} The reason why we could not detect any relationship may be that there is a small number of patients with severe HP infection in the current study. Because of that, further studies with more patients with severe HP infection may be needed.

Limitations

First, since the present study is a cross-sectional study, we could not detect a causal relationship between NLR, MPV and HP. Second limitation is that the absence of long-term follow-up after treatment to control treatment response, and the lack of monitoring of this marker over time. NLR and MPV were measured only once after HP eradication treatment.

In conclusion, there was no association between NLR and MPV values with HP infection, severity classification, or pre- and post-treatment status. ■

REFERENCES

- Hooi JKY, Lai WY, NG WK, Suen MMY, et al. Global prevalence of *Helicobacter pylori* infection: systematic review and meta-analysis. *Gastroenterology*. 2017;153(2):420-9.
- Zabala Torres B, Lucero Y, Lagomarcino AJ, Orellana-Manzano A, et al. Review: prevalence and dynamics of *Helicobacter pylori* infection during childhood. *Helicobacter*. 2017;22(5): e12399.
- Ruggiero P. *Helicobacter pylori* and inflammation. *Curr Pharm Des*. 2010;16(38):4225-36.
- Kusters JG, van Vliet AHM, Kuipers EJ. Pathogenesis of *Helicobacter pylori* infection. *Clin Microbiol Rev*. 2006;19(3):449-90.
- Kotilea K, Kalach N, Homan M, Bontemps P. *Helicobacter* infection in pediatric patients: Update on diagnosis and eradication strategies. *Pediatr Drugs*. 2018;20(4):337-51.
- Nagatomi R. The implication of alterations in leukocyte subset counts on immune function. *Exerc Immunol Rev*. 2006;12:54-71.
- Muhammed Suliman MA, Bahnacy Juma AA, Ali Almadhani AA, Pathare AV, et al. Predictive value of neutrophil to lymphocyte ratio in outcomes of patients with acute coronary syndrome. *Arch Med Res*. 2010;41(8):618-22.
- Ubukata H, Motohashi G, Tabuchi T, Nagata H, et al. Evaluations of interferon-gamma / interleukin-4 ratio and neutrophil / lymphocyte ratio as prognostic indicators in gastric cancer patients. *J Surg Oncol*. 2010;102(7):742-7.
- Tamhane UU, Aneja S, Montgomery D, Rogers EK, et al. Association between admission neutrophil to lymphocyte ratio and outcomes in patients with acute coronary syndrome. *Am J Cardiol*. 2008;102(6):653-7.
- Tan HJ, Goh KL. Extragastrintestinal manifestations of *Helicobacter pylori* infection: facts or myth? A critical review. *J Dig Dis*. 2012;13(7):342-9.
- Arnold DM, Bernotas A, Nazi I, Stasi R, et al. Platelet count response to *H. Pylori* treatment in patients with immune thrombocytopenic purpura with and without *H. Pylori* infection: a systemic review. *Haematologica*. 2009;94(6):850-6.
- Shah B, Sha D, Xie D, Mohler ER 3rd, Berger JS. The relationship between diabetes, metabolic syndrome, and platelet activity as measured by mean platelet volume: the National Health and Nutrition Examination Survey, 1999-2004. *Diabetes Care*. 2012;35(5):1074-8.
- Farah R, Khamisy-Farah R. Association of neutrophil to lymphocyte ratio with presence and severity of gastritis due to *Helicobacter pylori* infection. *J Clin Lab Anal*. 2014; 28(3):219-23.
- Jakarzadeh A, Akbarpoor V, Nabizadeh M, Nemati M, Rezayati MT. Total leukocyte counts and neutrophil-lymphocyte count ratios among *Helicobacter pylori*-infected patients with peptic ulcers: independent of bacterial CagA status. *Southeast Asian J Trop Med Public Health*. 2013; 44(1):82-8.
- Guclu M, Faruq Agan A. Association of severity of *Helicobacter Pylori* infection with peripheral blood neutrophil to lymphocyte ratio and mean platelet volume. *Euroasian J Hepatogastroenterol*. 2017;7(1):11-6.
- Jones NL, Koletzko S, Goodman K, Bontemps P, et al. Joint ESPGHAN / NASPGHAN Guidelines for the Management of *Helicobacter pylori* in Children and Adolescents (Update 2016). *J Pediatr Gastroenterol Nutr*. 2017;64(6):991-1003.
- Dixon MF, Genta RM, Yardley JH, Correa P. Classification and grading of gastritis. The updated Sydney System. International Workshop on the Histopathology of Gastritis, Houston 1994. *Am J Surg Pathol*. 1996;20(10):1161-81.
- Koletzko S, Jones NL, Goodman KJ, Gold P, et al. Evidence-based guidelines from ESPGHAN and NASPGHAN for *Helicobacter pylori* infection in children. *J Pediatr Gastroenterol Nutr*. 2011;53(2):230-43.
- Papa A, Emdin M, Passino C, Michelassi C, et al. Predictive value of elevated neutrophil-lymphocyte ratio on cardiac mortality in patients with stable coronary artery disease. *Clin Chim Acta*. 2008;395(1-2):27-31.
- Kondo Y, Joh T, Sasaki M, Oshima T, et al. *Helicobacter pylori* eradication decreases blood neutrophil and monocyte counts. *Aliment Pharmacol Ther*. 2004;20 Suppl 1:74-9.