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Professional paper

AGE AND GENDER AS RISK FACTORS IN *HELICOBACTER PYLORI* INFECTIONS: A RETROSPECTIVE STUDY

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Abstract

Helicobacter pylori infection remains widespread globally, affecting an estimated 44.3% of the world population. Although previous studies have linked infection prevalence with factors such as age, gender, and socioeconomic conditions, the primary risk factors vary significantly between regions. This study aimed to investigate the potential influence of age and gender on *H. pylori* infection rates within a sample from the Polog Region. A total of 225 outpatients, both male and female, were enrolled. Serum IgG levels were measured using a solid-phase chemiluminescent immunometric assay. Findings revealed a notably high infection rate, with roughly one-quarter of participants testing positive. Statistical analysis demonstrated a significant positive association between age and IgG test positivity (coefficient = 0.017, p = 0.027), indicating an increased likelihood of infection among older individuals. While gender alone did not show a significant effect, models incorporating both age and gender yielded improved predictive performance. Nonetheless, the inclusion of additional variables could further enhance risk assessment accuracy.

Keywords: Helicobacter pylori, peptic ulcers, age, gender, chemiluminescent, risk factors.

1. Introduction

Helicobacter pylori is a spiral-shaped, gram-negative bacterium that infects nearly half of the global population, showing a higher prevalence in developing countries. This pathogen is a major cause of chronic or atrophic gastritis, peptic ulcer, gastric lymphoma, and gastric carcinoma. However, these complications are less common in children and adolescents than in adults (Hooi *et al*, 2017). The infection is often asymptomatic, but can cause symptoms such as stomach pain and other gastrointestinal discomfort. Although *H. pylori* infection is known to be influenced by factors such as age, gender, and socioeconomic status, the main risk factors and modes of transmission vary by country. In both developing and developed countries, the high prevalence of this infection appears to be closely associated with adverse socioeconomic conditions (Shi *et al*, 2008). Zamani in his study concluded that in general the infection was more widespread in developing countries (50.8%), compared to developed countries, where it was lower (34.7%) (Zamani *et al*, 2018). Through a systematic literature review, Peleteiro et al (2014) aimed to describe the prevalence of *H. pylori* infection in different countries and time periods. They identified 37 eligible studies, including data for 22 countries. Prevalences were highest in Central/South America and Asia, and at least twice as high in countries with a high incidence of gastric cancer. In most countries that provided data for different time periods, prevalences were usually lower in more recent surveys (Peleteiro *et al*, 2014). The prevalence of infection differs in developing countries compared to developed countries. In Eastern Europe, the prevalence of infection in adults is 70%, while in Western Europe it is 30%. A similar prevalence (30%) of *H. pylori* infection in adults has also been estimated in the United States

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and Canada (Suerbaum *et al*, 2002; Hunt *et al*, 2010). In Albania, in the study conducted by Dura et al. (2024), in which age, gender, clinical risk factors, sociodemographic and hygienic conditions were taken into consideration in the assessment of the positivity of infection, it was found that the prevalence of *H. pylori* was 38.42% (842 out of 2191) (Dura *et al*, 2024). Unfortunately, the exact method by which *H. pylori* is transmitted is unknown. There are reports that it is spread through the fecal-oral and/or oral-to-oral routes. Both drinking water and food contaminated with this pathogen contribute to the spread. Infections can also occur as a result of poor hygiene, inadequate nutrition, and geographical changes, while the development of several virulence factors allows *H. pylori* to survive at a lower pH level. Since the bacterium cannot produce its own acid, the enzyme urease neutralizes gastric acid (Elbehiry *et al*, 2023). The main component of *H. pylori's* resistance to the acidic environment of the stomach is the enzyme urease, which breaks down urea into ammonia and carbamate, which spontaneously converts into another molecule of ammonia and carbon dioxide. The ammonia produced by this reaction increases the pH. Both ammonia and bicarbonate produced by urease are involved in the pathogenesis of *H. pylori* infection, and ammonia is thought to have a cytotoxic effect on gastric epithelial cells (Kusters *et al*, 2006).

H. pylori colonizes the stomach in childhood and persists throughout life, as a result of a perfect adaptation to its location and the ability to evade the immune response. Its spiral shape and flagella enable it to penetrate gastric mucus, and its multiple adhesins enable selective attachment to the epithelium, and *H. pylori* also possesses multiple mechanisms for protection against gastric acid (Atherton *et al*, 2009). *H. pylori* infection and smoking or drinking habits have given conflicting results. According to the study by Hussein et al. (2022), men who smoke and drink alcohol are more likely to test positive for *H. pylori* (Hussein *et al*, 2022), Luzza et al (1998) report that wine consumption is not a protective factor against infection (Luzza *et al*, 1998), while the study by Quartero & Wit (1998) reported that alcohol consumption has a protective effect against *H. pylori* infection (Quartero & Wit 1998). The degree of gastric damage from *H. pylori* infection varies from one subject to another, suggesting the existence of several genetic factors that play an important role in determining *H. pylori* infection (Hishida *et al*, 2010). The increasing prevalence of antibiotic resistance has complicated therapy, and as an infectious disease, therapy should be based on antibiotic susceptibility testing. The use of natural compounds in the treatment of *H. pylori* has gained great popularity due to their few side effects and low toxicity. In vitro and in vivo studies have reported encouraging results (Cardos *et al*, 2021).

This study aims to examine the impact of age and gender as risk factors for the development of *Helicobacter pylori* infection within the population of the Polog Region.

2. Materials and Methods

This retrospective, cross-sectional study was conducted between June and December 2024 and included 225 adult outpatients of both genders. Participants were consecutively selected based on their presentation to gastroenterology outpatient clinics in Tetovo, following predefined inclusion and exclusion criteria. Individuals aged 18 years or older who provided informed consent and had not previously received Helicobacter pylori treatment were eligible for inclusion. Patients who had recently used antibiotics or proton pump inhibitors, or those with severe comorbidities (e.g., cancer, immunodeficiency), were excluded.

Participants were categorized according to age groups and gender. Morning venous blood samples were collected from each subject. After centrifugation, serum was separated and analyzed for the presence of H. pylori-specific IgG antibodies using the Immulite 2000 analyzer (Siemens Healthcare Diagnostics). The assay is a solid-phase, chemiluminescent immunometric method with two incubation periods of 30 minutes each. A result of ≥1.1 U/mL was interpreted

as positive (indicating current or past exposure to H. pylori), while values <0.9 U/mL were considered negative.
All laboratory analyses were performed at the Biochemical Laboratory of the Clinical Hospital in Tetovo, which is equipped with the necessary technology for standardized serological testing. It is important to note that serological testing for IgG antibodies does not distinguish between active and past infections. In this study, no additional confirmatory tests, such as the urea breath test or stool antigen test, were performed, which represents a limitation in diagnostic accuracy. Data on socioeconomic status, hygiene conditions, history of antibiotic use, and lifestyle-related factors (e.g., smoking and alcohol consumption) were not collected and therefore not included in the statistical analysis.
All statistical analyses were conducted using IBM SPSS Statistics, version 20.0. Descriptive statistics were calculated, and group comparisons were performed using appropriate statistical tests. A p-value < 0.05 was considered statistically significant.

3. Results

The study enrolled 225 outpatients, ranging in age from 3 to 87 years, who presented with a variety of gastrointestinal complaints. Detailed results from the collected data are summarized in the tables and figures below.
From the results of testing for *Helicobacter pylori* IgG in a sample of 225 individuals in the Polog region of the Republic of North Macedonia presented below (Figure 1), it results that from the total number of patients included in the study (225 patients), 44.4% (100 out of 225) people tested positive for *Helicobacter pylori*, while 55.6% (125 out of 225) were negative. This result suggests a relatively high prevalence of *Helicobacter pylori* infection in this population sample, indicating that approximately one quarter of the individuals tested are affected by this infection.

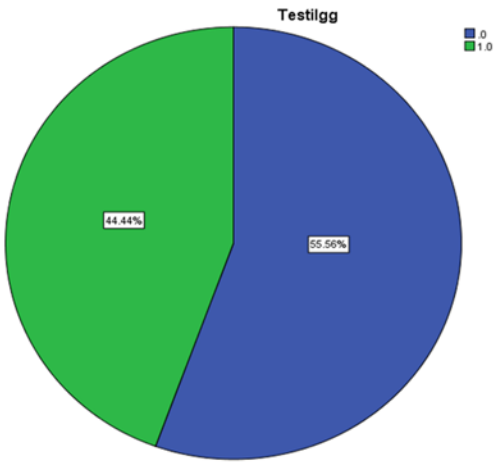

Figure 1. Relative frequency of positive and negative tests for *Helicobacter pylori* through serological testing

Table 1. Distribution of patients by gender tested with the IgG serological test for H. pylori

	Negative	Percentage (%)	Positive	Percentage (%)	Total
Total	125	55.56%	100	44.44%	225
Female gender	79	57.7%	58	42.3%	137
Masculine gender	46	52.3%	42	47.7%	88

From the data in the Table 1, it can be observed that the percentage of females and males with a negative result is higher than the percentage of those with a positive result. Out of a total of 225 individuals, 125 (55.56%) have a negative result, while 100 (44.44%) have a positive result.

In the female group, 79 (57.7%) have a negative result and 58 (42.3%) have a positive result. While in the male group, 46 (52.3%) have a negative result and 42 (47.7%) have a positive result. These data show that the distribution of negative and positive results between genders is relatively similar, however, negative results are more frequent in both groups.

Table 2. Descriptive statistics for IgG test results, as well as patient age

	IgG test	Age
Average	.444	48.12
Mode	.0	50
Standard deviation	.4980	18.125
Range	1.0	84
Maximum	1	87
Minimum	0	3

These data are important for understanding the gender impacts of infection and can help define risk groups. The average age of patients tested for *H. pylori* IgG was 48.12 years, with a standard deviation of 18.13 years, indicating a wide age range among the participants. As can be seen, the youngest patient in the IgG test is 3 years old, while the oldest patient is 87 years old. This information indicates that middle-aged patients are the ones who are most likely to be tested for *Helicobacter pylori*, or that they are most likely to need these types of tests. The mean IgG test result is 0.444, indicating an overall mean result across all patients, with a value that is below the maximum possible (1.0). The mode 0.0 indicates that some patients have a negative result (not infected with *Helicobacter pylori*), and this is the most common result value for the IgG test. The standard deviation, 0.498, indicates the relative variability of the results. This is a significant deviation from the mean, suggesting that there is a wide distribution of test results, from negative to positive, meaning that there is a large spread of true values from the mean value of the variable. This test has a range from 0 to 1, where 1 is the maximum, indicating that there are complete possible results that lie within this range. In terms of maximum and minimum, the highest possible value is 1 and the lowest is 0, suggesting that all patients have been tested within this possible range.

The following model was estimated to see how age and gender affect the likelihood of *Helicobacter pylori* in serum. The model we estimated is an ordinal model which has the following form:

$$P(Y \leq j|age, gender) = \beta_0 + \beta_1 age + \beta_2 gender + \epsilon$$

from where:

-**age** is years and in the estimation is a categorical variable with continuous values;

-**gender** is gender, which in the model is a binary variable with a value of 1 for female and 0 for male;

- **Y** represents the test result and is a binary variable with a value of 0 for negative results and 1 for positive results.

Table 3. Estimated coefficients for model 1 and results from the IgG test sample

		Estimate	Standr. error	Wald	d.f.	p-value	Odds
Threshold	[TestIgG = .0]	.933**	.439	4.513	1	.034	2.542
	Age	.017**	.008	4.897	1	.027	1.017
	[gender1=F]	-.184	.278	.437	1	.509	0.832
	[gender1=M]	0[a]	.	.	0	.	

*Statistically significant at *α=0.1 **α=0.05 ***α=0.01, [a] this parameter is set to 0 because it is redundant*

From the results in Table 3, regarding the influence of age, we can say that the age coefficient is 0.017, which means that for each additional year of age, the likelihood of testing positive for IgG increases by 1.017 times, keeping gender unchanged. This coefficient is statistically significant (p = 0.027, which is less than 0.05 or p<0.05), suggesting that age has a significant impact on IgG test results. This means that older individuals are more likely to test positive for IgG than younger individuals.

Regarding the effect of gender, we can say that the coefficient for female gender is -0.184, which indicates that female gender is 0.832 times less likely to be positive in the IgG test compared to male gender. However, this coefficient is not statistically significant (p = 0.509, which is greater than 0.1), indicating that there is no significant influence of gender on the results of the IgG test.

In conclusion, age has a significant impact on the likelihood of IgG testing, with older individuals more likely to test positive. No statistically significant association was found between gender and the IgG test results. The probability for a 35-year-old man to test positive for IgG is 82.2%, but this is relatively higher than the prevalence of infection in this sample.

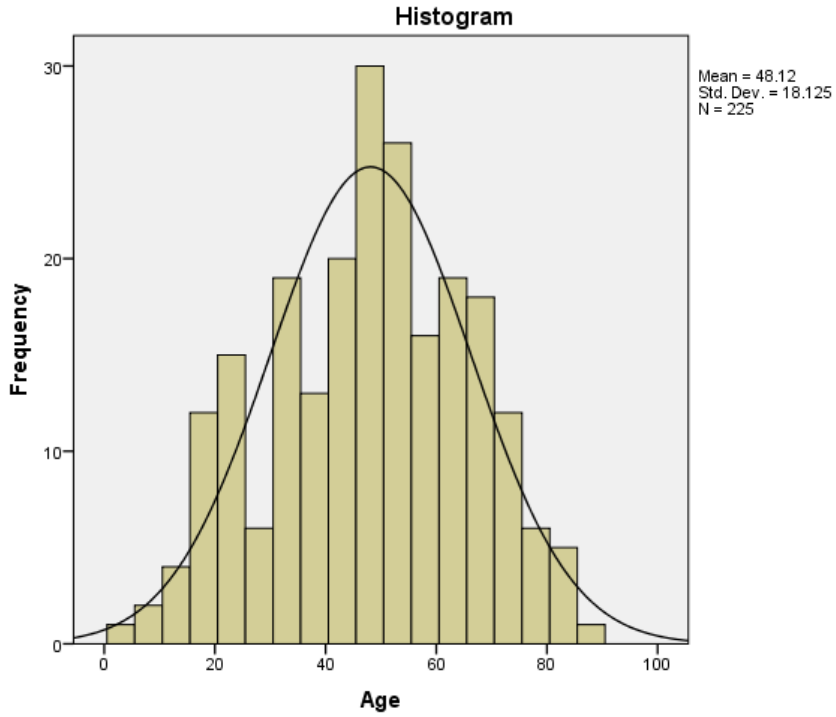

Figure 2. Histogram for patient age for the IgG test

From Figure 2, it is observed that the age distribution is approximately normal, with a slight trend towards older ages, suggesting that older patients have a greater propensity to be tested for *Helicobacter pylori*. The average age is around 48 years with a standard deviation of 18 years, indicating a significant age difference between individuals who underwent the test.
The most represented age is in the range of 40 to 50 years, which indicates that the middle-aged group is the one that dominates for testing. Also, a significant decrease in frequency can be observed at both ends of the graph, indicating that there are fewer very young or very old patients taking this test.
This study has several inherent limitations that should be considered when interpreting the results. The retrospective design restricts the ability to establish causality and limits control over potential confounding variables such as socioeconomic status, lifestyle factors including smoking and alcohol consumption, as well as clinical symptoms that were not recorded. Furthermore, the gender distribution, with a higher proportion of female participants, and the outpatient setting may introduce selection bias, potentially affecting the generalizability of the findings.
The reliance solely on serological IgG testing to diagnose Helicobacter pylori infection is another limitation, as it does not distinguish between active and past infections. Incorporating additional diagnostic methods, such as the urea breath test or stool antigen detection, could improve diagnostic accuracy and better reflect current infection status. Moreover, other influential factors like hygiene practices, living conditions, and prior antibiotic use were not assessed but could significantly impact infection prevalence.
Future research should aim for a prospective study design that allows for better control of confounders and the inclusion of multivariate analyses with a broader set of sociodemographic and behavioral variables. This approach would enhance the understanding of risk factors and the epidemiology of H. pylori infection in the population.

4. Discussion

Diagnosis of *H. pylori* infection in the stomach is commonly established through upper endoscopy. However, serological methods have reached a level of accuracy sufficient to be used as screening tests before performing endoscopy, or for sero-epidemiological studies, as individuals infected with *H. pylori* develop antibodies that are strongly associated with the infection, which is confirmed histologically by *H. pylori* (Kouitcheu *et al*, 2018; Philipp *et al*, 2004). The prevalence of *H. pylori* infection differs significantly across the globe, ranging from under 40% in developed nations to as high as 80-90% in developing countries (Shi et al., 2008). In this study, the prevalence of *Helicobacter pylori* infection was found to be below 50%. Out of a total of 225 patients included, 125 tested negative and 100 tested positive for the infection. In comparison, the study by Vianna et al., which included 227 patients, reported a frequency of 66.5% (151 patients), consistent with frequencies reported in recent years in Brazil and other developing countries (Vianna *et al*, 2019), but higher than that found in our study. The variation in *H. pylori* infection rates worldwide is largely influenced by the socioeconomic status of populations. In developed countries, prevalence tends to be lower (around 30%), which is likely due to better access to improved sanitation, hygiene, and established protocols for disease prevention and treatment. The same study did not reveal any significant difference between gender and *H. pylori* infection (p=0.37), which is also consistent with the results of our study. The largest number of patients tested belonged to the female gender compared to the male gender (60.89% vs. 30.11%), but positive results for *H. pylori*, in higher percentages, were obtained in males compared to females (47.72% vs. 42.33%), however, the influence of gender is not statistically significant in our study. Similar results have been reported in previous studies (Elbehiry *et al*, 2023; Seisebekova *et al*, 2025; Hasings *et al*, 2014; Rothenbacher *et al*, 1998),

however, other studies have shown a higher prevalence of infection in men (Abebaw *et al,* 2014; Murray *et al*, 1997). Gruber et al. in 2008 found no gender differences in the study population, while age, race, and socioeconomic status influenced the prevalence of *H. pylori* positivity (Gruber *et al,* 2008). The results of our study show that *Helicobacter pylori* infection is present in individuals of all age groups and both sexes, a finding that is consistent with Kassid's study, which concludes that *H. pylori* infection is common in the Iranian community, in people of all ages and genders. Several epidemiological and lifestyle factors appear to be crucial in the transmission of the disease. (Kassid *et al,* 2022). The findings of our study are also consistent with those of Vianna et al., that there is no significant difference between gender and *H. pylori* infection (p=0.37), while regarding the relationship between infection and the age of the patients, a statistically significant relationship is observed (p=0.04). The average age of patients with infection was 53.4±13.9 years, with an interval of 20-88 years, where it was observed that the frequency of *H. pylori* infection increases with age (Vianna *et al,* 2019). The study conducted by Bureš et al. (2006) concluded that the prevalence of *H. pylori* infection was not related to gender, but was associated with low socioeconomic conditions and increased with age (Bureš *et al*, 2006).

This study has several limitations that should be acknowledged. The retrospective design restricts the ability to infer causality and does not allow control for potential confounding variables such as socioeconomic status, hygiene practices, lifestyle behaviors (smoking, alcohol consumption), or detailed clinical symptomatology. Furthermore, the use of IgG serology alone may not differentiate between active and past infections, possibly leading to overestimation of current infection prevalence. A more comprehensive diagnostic approach, including urea breath tests or stool antigen testing, would improve accuracy. Additionally, the outpatient population with a predominance of females may introduce selection bias, potentially limiting the generalizability of the findings. Future research should consider prospective study designs and incorporate broader sociodemographic, behavioral, and clinical variables to allow for multivariate analyses and a more nuanced understanding of the determinants of H. pylori infection.

5. Conclusions

The data from this study indicate that age is an important factor influencing serological positivity for *Helicobacter pylori,* with older individuals showing a higher prevalence of infection. Although a higher proportion of positive cases was observed among women compared to men, this difference was not statistically significant. Statistical models suggest that combining age and gender improves predictive accuracy; however, additional clinical and sociodemographic variables may be necessary for a more comprehensive risk assessment. The highest infection rate was identified in the 40–50 age group, reflecting an increased tendency for diagnosis during this stage of life.

While the current study provides valuable insights, the findings should be interpreted with caution due to study limitations. Future studies are recommended to adopt prospective designs, include larger and more diverse samples, and use multiple diagnostic methods for *H. pylori* detection. Incorporating a wider range of sociodemographic, behavioral, and clinical variables will facilitate more robust conclusions and support the development of effective public health strategies and clinical interventions.

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