



# COMPARISON OF RAPID UREASE TEST WITH IMMUNOHISTOCHEMICAL EXAMINATION OF GASTRIC MUCOSA SPECIMEN IN THE DIAGNOSTICS OF *HELICOBACTER PYLORI* INFECTION AMONG OUTPATIENTS

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## Abstract

*Helicobacter pylori* infection affects half of the human population. It is the most common infection in the world. Its presence is associated with numerous complications, including gastric cancer. More than 30 years ago, this bacterium was included by the World Health Organisation in the list of class 1 carcinogens, precisely because of its link to gastric cancer (CHO et al. 2021, LEE et al. 2024, TOBI et al. 2023). Its prevalence varies depending on geographical location, and even within a single country, different incidence rates have been reported (CHO et al. 2021, CODOLO et al. 2022). Infection most often occurs around the age of 10 and is usually asymptomatic. Therefore, it slowly leads to numerous complications. Relatively few patients experience symptoms associated with this condition and seek medical attention. The diagnostic pathway includes non-invasive and invasive tests, depending on the patient's age and the presence of worrying symptoms.

## Introduction

Although the human digestive tract is colonized by many species of bacteria, mainly Firmicutes, as in other mammals, it seems that *Helicobacter pylori* is the most well-known pathogen (BURAIMOH 2023).

*Helicobacter pylori* is a Gram-negative microaerophilic bacterium with numerous features that allow it to survive in the harsh milieu of the stomach. It is characterised by high geographical variability (Figure 1). Its prevalence depends on many socio-economic and demographic factors, antibiotic resistance, and genetic factors (CHO et al. 2021). It is reported that the highest number of infections takes place in Africa, where it affects about 70–80% of the population. However, these data seem to be underestimated, which is also connected to the highest risk of a dangerous, chronic complication of *H. pylori* infection, namely gastric cancer (KOLLI et al. 2021, MACHAJ et al. 2025). The main risk factors for infection are poor hygiene and contaminated water (XIE et al. 2024).

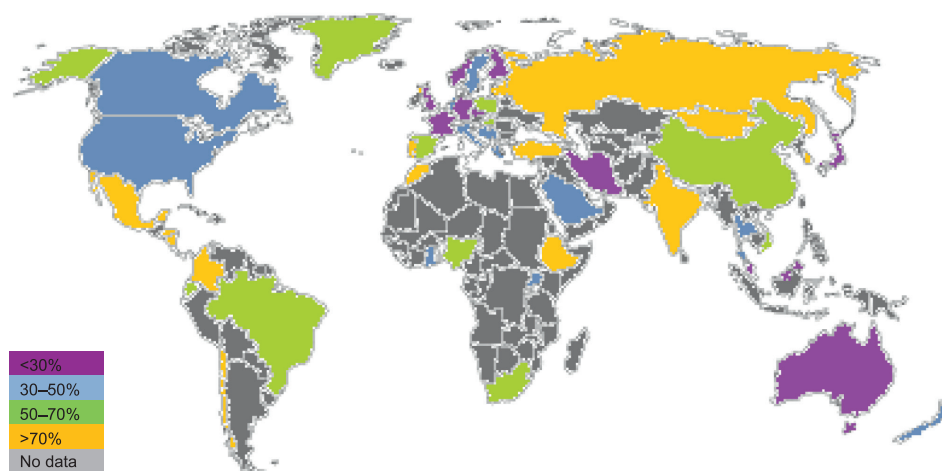


Fig. 1. Global prevalence of *Helicobacter pylori*  
Source: based on HU et al. (2017), CC BY 4.0, modified

This bacterium exhibits many mechanisms that allow it to survive in the stomach and dominate its flora (SOUSA et al. 2022). Its arsenal includes motility, adhesion ability, enzymes that help it create favourable conditions for its existence in the stomach as well as virulence factors. The agents responsible for its virulence are proteins such as CagA, VacA, OpiA, and DupA, which provide its survival, as well. Some of the proteins affect the cytoskeleton of gastric epithelial cells and the host's immune response. They cause chronic inflammation of the gastrointestinal mucosa. According to the Correa pathway, chronic inflammation can lead to ulcers of the gastric or duodenal mucosa. It can also lead to atrophy or metaplasia of the gastric mucosa, i.e. precancerous conditions, and ultimately to carcinogenesis and the development of adenocarcinoma or MALT (marginal zone lymphoma)

(CHO et al. 2021, DE BRITO et al. 2019, FUJIMORI 2021, GUO et al. 2023, IINO et al. 2021). The frequency of individual gastric complications is as follows: chronic gastritis occurs in 100% of infected patients, peptic ulcers occur in 20%, dyspepsia affects 10% and gastric cancer affects 1% of patients (LIANG et al. 2022, SHUKLA et al. 2024). The literature increasingly discusses the extra-gastric consequences of *H. pylori* infection, yet among the complications proven to date are unexplained anaemia due to iron or vitamin B12 deficiency, and immune thrombocytopaenia (CHO et al. 2021, HURKALA et al. 2023). An inevitable difficulty in taking on diagnostics is that only 1–2 out of 10 infected people experience symptoms of infection. This means they present no need to seek medical attention (BARTNIK et al. 2014). According to the majority of gastroenterological societies, *H. pylori* infection is an indication for eradication. Treatment of this infection contributes to the treatment of peptic ulcers and protects against their recurrence. It significantly reduces the risk of gastric cancer and can cure MALT lymphoma (DE BRITO et al. 2019). Therapies used to treat *H. pylori* are multi-drug and complex, but they reduce the risk of gastric cancer by about 40% (CHOI et al. 2025).

### **Diagnostic methods for *Helicobacter pylori* infection**

Methods for detecting *H. pylori* infection include invasive and non-invasive tests. Invasive tests, i.e. those performed during gastroscopy and gastric mucosa biopsy, include the rapid urease test (RUT), culture with 80% sensitivity and 100% specificity, and PCR, for which samples can also be taken from faeces or saliva. PCR tests currently seem to be the field of greatest development in the diagnostics of *H. pylori*, offering enormous potential with high sensitivity (YU et al. 2025).

The rapid urease test possesses high sensitivity and specificity, reaching 90–95%. It uses the enzyme urease, which is produced by *H. pylori*. Urease breaks down urea into ammonia. In turn, ammonia, by binding with hydrogen, causes a pink colouration of the test plate (CHO et al. 2021). Histopathological examination also has high sensitivity and specificity, 95% and 99%, respectively, depending on the bacterial density and staining method used. However, it is expensive and time-consuming (LEE et al. 2015, LU et al. 2025, PARRA-MEDINA et al. 2025). Its sensitivity can be boosted by new techniques from the field of immunohistochemical methods (LOHARAMTAWEEHONG et al. 2025).

Non-invasive tests include the urea breath test, which is recommended by a number of societies. It is a highly specific test with a specificity of 95–100% and a sensitivity of 88 to 95%. The patient consumes a solution of urea

labelled with carbon  $C^{13}$ . Urease breaks down the urea, producing ammonia and carbon dioxide, which when exhaled with the carbon  $C^{13}$  incorporated, indicates a positive test result. Another widely used and easily accessible test is the faecal antigen test, which searches for *H. pylori* antigens excreted in the stool. It is also a high-quality test, with sensitivity and specificity reaching 97%. However, it should be noted that these tests may have varying sensitivity depending on the manufacturer (HORSMA-HEIKKINEN et al. 2025, SZCZEKLIK et al. 2022). Serological blood tests also belong to the so-called non-invasive tests, but due to their low sensitivity and specificity (80%) and the problematic phenomenon of antibodies persisting for years after *H. pylori* eradication, they are not recommended. Apart from serological assay, the above tests are burdened with a raised risk of error due to the use of, among other things, proton pump inhibitors, gastric mucosal atrophy, bleeding or the presence of neoplastic cells (IMPERIAL et al. 2024, SZCZEKLIK et al. 2022). New, increasingly advanced diagnostic methods are constantly being sought.

## Treatment

Despite effective treatment, the annual risk of recurrence reaches 3%. Among the therapeutic strategies analysed by Sun et al. 209 different therapies were taken into account in terms of treatment duration and drugs used. Their research demonstrated how much geographically diverse the problem is, depending on local antibiotic resistance mechanisms. Quadruple therapy with bismuth is most often recommended (SUN et al. 2025).

Curing *H. pylori* infection prevents the development of gastric cancer, influences the resolution of chronic inflammation, the progression of inflammation to atrophic gastritis or its metaplasia. It may also exert a positive impact on the progression of metaplasia and atrophy of the gastric mucosa, although the effects are less pronounced in this case (SHARARA et al. 2025).

It is particularly in these precancerous conditions that eradication is paramount and recommended for all patients (MORGAN et al. 2025). However, there are voices pointing to the negative aspects of the global eradication of *H. pylori*, which are also worthy of considerable attention, namely the growing antibiotic resistance. This is estimated at 15–50%. This is counterbalanced by the possibility of selecting individualised therapy thanks to genetic sequencing, as *H. pylori* strains differ from one another with regard to virulence as well as sensitivity to treatment (UMAR et al. 2025, WRONECKI et al. 2025).

## Material and Methods

88 women (70%) and 38 men (30%) were included in the study, with the mean age of 47 years (the youngest participant was 18, the oldest 82). Gastroscopy was performed with a rapid urease test and biopsy of the gastric mucosa. The biopsy specimen was subjected to immunohistochemical assay, manufactured by Roche, to diagnose the presence of *H. pylori* in the pathomorphological examination. Both diagnostic tests were performed in each patient.

Approval was obtained from the Bioethics Committee at the Faculty of Medicine of the University of Warmia and Mazury in Olsztyn on 21st February 2019, resolution No. 9/2019.

## Results

Among 126 subjects, a positive rapid urease test was found in 51 subjects (40.5%), and a negative test in 75 subjects (59.5%). *H. pylori* infection was confirmed in histopathological examination in 55 patients (43.6%), and not confirmed in 71 patients (56.4%). Our findings are presented in Table 1. A total number of 64 people (50.8%) were confirmed to have *H. pylori* infection by one or both methods.

Results of diagnostic tests for *Helicobacter pylori* (HP)

Table 1

Test <i>n</i> [%]	HP positive	HP negative
Urease test	51 (40.5%)	75 (59.5%)
Biopsy	55 (43.6%)	71 (56.4%)

The concordance of positive results of the rapid urease test and the pathomorphological examination occurred in 42 subjects, i.e. 65.6% of those infected. A positive histopathological examination result was found in 85.9% of the infected population, and a positive rapid urease test result was found in 79.7% of the infected population (Table 2).

Distribution of positive results for *Helicobacter pylori*

Table 2

Test <i>n</i> [%]	Infected <i>n</i> = 64 (50.8%)	All <i>n</i> = 126 (100%)
Positive urease test	51 (79.7%)	51 (40.5%)
Positive biopsy	55 (85.9%)	55 (43.6%)
Positive urease test + positive biopsy	42 (65.6%)	42 (33.3%)

On the grounds of that, the sensitivity of the rapid urease test (13 false negative results) can be determined at 83%, while that of the pathomorphological examination (9 false negative results) at 87.7%.

## Discussion

SUN et al. analysed nine online databases containing guidelines on *H. pylori*. Among the 25 guidelines, the urea breath test is the main recommended tool for the diagnostics of infection. Seven guidelines clearly do not recommend serological blood tests due to the inability to distinguish between past and active infection. The indications for eradication included long-term therapy with aspirin or non-steroidal anti-inflammatory drugs, or a history of peptic ulcer disease. A minority of the guidelines, 40%, recommended getting rid of *H. pylori* as long as it is detected, which is also confirmed by the Kyoto Consensus treating the presence of *H. pylori* as an infectious disease (SUN et al. 2025, SUGANO et al. 2022).

When it comes to verifying the efficacy of eradication, the guidelines mainly recommend the urea breath test, followed by the stool antigen test, while advising against the rapid urease test as a method of confirming effective eradication (SUN et al. 2025). An invasive method of *H. pylori* detection, such as histopathology, is commonly used in Indonesia as the *gold standard*. In their study, MIFTAHUSSURUR et al. (2021) conducted gastroscopy, taking histologic specimens in accordance with the Sydney protocol, then placing the specimens in 10% formalin, paraffin and staining them using the Giemsa method. They compared the effectiveness of histopathological examination in achieving positive results with the urea breath test. Both methods were equally effective, achieving 23.6% positive results. This is considerably less than in our study, where we have demonstrated positive results using histopathology in 43.6% of cases. This is even more striking given the higher prevalence of *H. pylori* in Indonesia, reaching 80–90%, compared to Poland (MIFTAHUSSURUR et al. 2021). On the other hand, MAJALIWA (2024) and his team compared three diagnostic methods: rapid urease test, histopathology and PCR. The rapid urease test was positive in all patients (100%), histopathology in 35% of patients and PCR in 65%. However, when the authors combined all three methods, they achieved 75% positive results for the three tests. In our study, we compared two methods, yet having achieved markedly lower results, with two tests agreeing in 33.3% of our patients. The group studied by MAJALIWA was smaller, consisting of 80 residents of Mozambique, where the expected incidence of *H. pylori* is higher than in the Polish population (MAJALIWA et al. 2024). In other studies, the concordance of three tests was combined: rapid urease test, Gram staining

and serology, which resulted in 94% efficacy in detecting infection, or the rapid urease test was combined with serology, which detected 85% of infections (MUJTABA et al. 2025). The combination of different diagnostic methods is becoming increasingly common. Slightly worse results were reported in Bangladesh, where 32.1% of patients were tested positive by PCR and 32.6% were tested positive by rapid urease test. This is comparable to our results regarding the compatibility of rapid urease test and histopathology (FERDAUS et al. 2025). Similarly, in another study, two diagnostic methods were combined: rapid urease test and histopathology, which allowed *H. pylori* infection to be detected in 43.7% subjects (YADAV et al. 2025). In a different study comparing the rapid urease test with histopathology, 58% of patients presented confirmed *H. pylori* infection in the rapid urease test and 52% in histopathology. That is comparable to our results, although the study only involved 50 patients (EL-NASR et al. 2003). The researchers assume that the varying sensitivity of the tests depends on a number of factors, including the presence of precancerous or cancerous conditions in the stomach (KIM et al. 2025).

## Conclusions

There is an increasing talk of combining two or three diagnostic methods for detecting *H. pylori*, as each of the methods mentioned in the introduction has its weaknesses, which can generate false negative results. This mainly concerns the use of proton pump inhibitors, antibiotics, bleeding, gastric mucosal atrophy, gastric mucosal neoplastic changes, the history of partial gastrectomy, the pathologist's experience, bacterial density in the biopsy specimen, and staining technique. Given the very high prevalence of *H. pylori* infection worldwide and the serious complications that this infection can lead to, combining several methods of diagnosing infection may be a new reasonable diagnostic trend.

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