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Detección de *Helicobacter pylori* en adultos con dispepsia en Ibagué (Colombia): evaluación comparativa de métodos

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ABSTRACT

Introduction: *Helicobacter pylori* infection has been associated with various gastrointestinal disorders, including chronic gastritis, peptic ulcers, dyspepsia, and gastric cancer. This study aimed to compare the effectiveness of four invasive diagnostic methods for detecting *H. pylori* in 297 patients with premalignant gastric lesions and gastric cancer. **Method:** In this cross-sectional study, the methods evaluated included Giemsa stain (GST), culture, rapid urease test (RUT), and polymerase chain reaction (PCR). Patients were recruited between 2016 and 2019 in Ibagué – Colombia, and a Case Definition Criteria (CDC) was used for diagnosis. Analysis of operational and epidemiological characteristics was performed to assess result agreement. **Results:** According to this,

it showed that *H. pylori* infection rates of 43% (RUT), 63% (GST), 24% (culture), and 42% (PCR). The Kappa index demonstrated higher values for PCR (0.7704) and RUT (0.7030) compared to other methods. The RUT test displayed the highest Kappa index (0.59) when compared to PCR. Non-parametric tests indicated that PCR (0.779) and RUT (0.708) had the strongest correlation, reducing prediction error by 51.1% and 40.4%, respectively. **Conclusion**: The CDC criteria exhibited enhanced reliability in diagnosing *H. pylori* infection. Notably, PCR and RUT showed significant correlation in diagnostic accuracy for *H. pylori* detection.

Keywords

Helicobacter pylori; diagnostic accuracy; molecular epidemiology; stomach neoplasms; precancerous conditions.

RESUMEN

Introducción: La bacteria *Helicobacter pylori* coloniza la mucosa gástrica e incrementa el riesgo de padecer trastornos digestivos, como gastritis crónica, úlceras pépticas, dispepsia y cáncer gástrico. **Metodología:** Este estudio transversal comparó la eficacia de cuatro métodos invasivos de detección de *H. pylori* en 297 pacientes con lesiones gástricas premalignas y cáncer gástrico. Se emplearon la tinción de Giemsa (GST), cultivo, prueba rápida de ureasa (RUT) y reacción en cadena de la polimerasa (PCR) en pacientes reclutados entre 2016 y 2019 en Ibagué (Colombia), usando un criterio de definición de caso (CDC). En el diagnóstico también se analizaron las características operativas y epidemiológicas para evaluar la concordancia de los resultados. **Resultados:**La identificación de *H. pylori* fue del 43%, 63%, 24% y 42%, mediante RUT, GST, cultivo y PCR. El índice kappa mostró que la PCR y la RUT tuvieron los valores más altos, con lo cual se demostró una correlación significativa en la mayoría de los análisis estadísticos. Las pruebas no paramétricas señalaron una mayor correlación entre la PCR y la RUT, que reduce significativamente el error al predecir la otra variable. **Conclusión:** El criterio de definición de caso mostró ser confiable para el diagnóstico de *H. pylori*, y la PCR y la RUT demostraron ser métodos correlacionados en la detección de esta bacteria.

Palabras clave

Helicobacter pylori; precisión diagnóstica; epidemiología molecular; neoplasias del estómago; condiciones precancerosas.

Introduction

Helicobacter pylori infection is a prevalent and significant health concern, particularly in developing countries with reported rates as high as 85-95% (1). In Colombia, the prevalence of *H. pylori* infection ranges from 69-78%, varying among different regions within the country (2,3). While many individuals infected with *H. pylori* remain asymptomatic, the bacterium's colonization of the gastric mucosa can lead to various gastrointestinal disorders, including chronic gastritis, peptic ulcers affecting up to 10% of infected individuals, and even gastric adenocarcinoma in 1-3% of cases (4,5). Despite extensive research, the precise role of *H. pylori* in the epidemiology and pathogenesis of gastric cancer remains incompletely understood, with genetic, infectious, and environmental factors collectively contributing to disease progression (6).

Diagnosing *H. pylori* infection involves noninvasive tests such as the Urea Breath Test (UBT), serology, and stool Antigen Test (SAT), while invasive methods like the Rapid Urease Test (RUT), histological analysis, culture, and molecular techniques require an endoscopic biopsy. Eradication of *H. pylori* is recommended for treating gastric and duodenal ulcers and for preventing associated diseases, including gastric cancer (7,8). Despite the availability of various diagnostic methods, their efficacy in the Colombian context remains insufficiently explored, emphasizing the need to establish standardized and effective diagnostic protocols for *H. pylori* infection. For instance, the RUT is commonly used for *H. pylori* diagnosis in Colombia; however, it may lack specificity regarding the actual presence of the bacterium in the stomach (9). Polymerase Chain Reaction (PCR) is increasingly adopted due to its high sensitivity, specificity, and rapid turnaround time (10). Similarly, Giemsa Staining (GST) enables histopathological diagnosis, facilitating the detection of *H. pylori* and assessment of gastric pathology (11). Bacterial culturing from gastric biopsies, although technically demanding, provides valuable information and can perform antibiotic susceptibility testing, albeit with variation in sensitivity across laboratories (12).

Given the importance of accurate and standardized diagnostic strategies, this study aims to evaluate and compare the efficacy of four invasive diagnostic methods for detecting *H. pylori* infection in dyspeptic patients.

Materials and methods

Type of study: This is a cross-sectional study of diagnostic test performance and concordance. The methods evaluated included GST, culture, RUT, and PCR.

Patients and sampling: 297 patients with gastric pathologies underwent upper gastrointestinal endoscopy at the Javeriano Medical Center in Ibagué – Colombia, from 2016 to 2019. All selected patients presented dyspeptic symptoms, had not previously undergone endoscopy, had not received treatment for *H. pylori* infection, or were undergoing follow-up for other gastric pathologies. For this study, three gastric biopsies were obtained from the pyloric antrum of each patient: one for RUT and molecular validation by PCR amplification of a 16S rDNA gene fragment, one for culture of bacteria, and one for histology with GST for the identification of the *H. pylori* infection. Finally, the treating physician took three antral biopsies for the histopathology analysis and pathology determination as part of the dyspepsia study of the patient. Biopsies were preserved in appropriate solutions, transported at 4 °C, and stored at −20 °C. All patients provided informed consent, and the study was approved by the University of Tolima Ethics Committee following the Helsinki Declaration.

Rapid Urease Test: Biopsy specimens were analyzed using the Sensibacter Pylori-Test® to detect *H. pylori* urease. This method is both sensitive and specific, providing a reliable and rapid alternative for diagnosis. A color change to red or fuchsia within ten minutes indicates the presence of *H. pylori*, resulting from urease hydrolyzing urea into ammonia and carbon dioxide, which alters the pH and subsequently changes the color (13). To ensure the effectiveness of the test, biopsies were obtained from patients who had not received antibiotic treatments in the last six months, and the sample collection of the patients included in this study was performed by the same gastroenterologist, using a Fujinon EG-590 WR endoscope, ensuring homogeneity in the size of all biopsies (2.8 mm). Additionally, it was verified that each urease test was within its expiration date and had been transported and stored at 4 °C, which was crucial for reducing the bias of false negatives.

Culture of H. pylori: One antral biopsy per patient was placed in 1 ml brucella broth with 10% horse serum, which is a suitable medium of transport that allows the bacteria to remain viable; posteriorly, the samples were placed in ice and processed in the following 4 hours of its collection. The gastric biopsy specimens were grown on blood agar supplemented with sodium carbonate, hydrolyzed casein, tryptone, activated carbon, 10% fresh horse blood serum, and 1% Vitox supplements and *H. pylori* selective supplements (Oxoid, Basingstoke, UK) at 37°C for 3 to 15 days under microaerophilic conditions (5% O_2) before being considered negative.

Molecular identification of H. pylori: DNA extraction was performed using DNeasy Blood and Tissue Kit (QIAGEN, USA) following the manufacturer's instructions. DNA was quantified using a Nanodrop ND™ 2000 UV-Vis spectrophotometer of Thermo Scientific, and quality was evaluated using the absorbance ratio at 260 and 280 nm (A260/A280). To test for the presence of *H. pylori*, a 537-bp fragment of the Sub unit 16 of ribosomal DNA (*16Ss rDNA*) gene was amplified using polymerase chain reaction (PCR) with the primers ACT-1 and ACT-2. To determine the genotype of *H. pylori* virulence genes the samples were subjected to PCR for the *cagA* and *cagE* genes and the signal regions (s_1 and s_2 alleles) and mid-region (m_1 and m_2 alleles) of *vacA* (14). The *H. pylori* NCTC 11638 strain (donated by the National Institute of Cancerology, Bogotá – Colombia) was used as a positive control, ultrapure water as a negative control, and thermocycling conditions were as described by López et al. (15).

Giemsa Staining: Biopsies embedded in paraffin were stained with Giemsa for histopathological diagnosis, determining bacterial load and classifying patients into pathology groups based on diagnostic criteria. For that reason, the bacterial load in 4 groups was determined: negative, low (+), medium (++), and high (+++). Finally, patients were assigned to one of the two groups: (i) chronic non-atrophic gastritis

(NAG), and (ii) preneoplastic lesion (PNL) included patients with chronic and atrophic gastritis, intestinal metaplasia, dysplasia, and GC. For the unified diagnosis, a consensus was reached between the endoscopy report and the pathology results provided by the treating physicians.

Diagnosis of H. pylori *infection:* The *H. pylori* infection was evaluated using the Case Definition Criteria (CDC) as the gold standard to determine the sensitivity and specificity of the diagnostic tests. A patient was considered positive if two or more tests confirmed the infection and negative if only one test was positive or all methods were negative. The choice of the CDC as the gold standard is justified by the need for a reliable standard that accurately distinguishes between healthy and sick individuals. However, it is important to note that this definition may present biases in identifying the infection, especially in patients with low bacterial load. Creating a CDC from the test data not only validates a reliable gold standard but also helps to estimate the biases present in the evaluated tests, allowing for a more accurate interpretation of their results.

Statistical analysis

Operational characteristics of diagnostic tests were evaluated, including sensitivity, specificity, and predictive values. The Kappa index determined agreement among tests and with the CDC, the interpretation of the results was carried out based on the scale proposed by Landis and Koch (16). Epidemiological characteristics were analyzed using descriptive statistics across seven features: age, sex, pathology diagnosis, bacterial load, bacterial genotype, CDC, and diagnostic tests. Chi-square independence tests assessed associations among these characteristics, with the Cramer V test and Uncertainty coefficient measuring the degree of association and error reduction, respectively. Descriptive statistics, independence tests, and Odds ratios were calculated for epidemiological characteristics and diagnostic outcomes, analyzing associations and differences in diagnostic test results and pathology diagnoses. Data analysis was performed using IBM SPSS Statistics software version 25 with a significance level set at $p<0.05$.

Results

Sample characteristics

Age and sex distribution varied with more cases in the 31-50 and 51-70 age groups, predominantly 67% female and 33% male (Table 1). *H. pylori* infection was detected in 43.1% (RUT), 63.3% (GST), 23.6% (culture), and 41.7% (PCR) of patients (Table 2). According to the CDC definition, 49.16% (146/297) of patients demonstrated *H. pylori* infection status (Table 1).

Table 1.
Characteristics of the sample according to the infection status by H. pylori

Variable		Sample (n [%])	Infection status* by *H. pylori*	
			Negative (n [%])	Positive (n [%])
Sex	Female	199 (67.00)	104 (68.87)	95 (65.06)
	Male	98 (33.00)	47 (31.12)	51 (34.93)
	Total	297 (100)	151 (50.84)	146 (49.16)
Age (years)	11–30	40 (13.46)	19 (12.58)	21 (14.38)
	31–50	93 (31.31)	35 (23.17)	58 (39.72)
	51–70	139 (46.80)	78 (51.65)	61 (41.78)
	71–90	25 (8.41)	19 (12.58)	6 (4.10)
	Total	297 (100)	151 (50.84)	146 (49.16)
Gastric pathology	NAG	242 (81.48)	125 (82.78)	117 (80.13)
	PNL	55 (18.51)	26 (17.21)	29 (19.86)
	Total	297 (100)	151 (50.84)	146 (49.16)

n (%)number of observations and percentage in each category; NAG: Non atrophic gastritis; PNL: Preneoplastic lesion.
*The infection status was determined according to the case definition.

The unified diagnosis revealed a majority in NAG. Culture, PCR, and RUT showed higher negative cases than GST. A slight correlation

was found between genotype frequencies and bacterial load concerning the CDC.

H. pylori detection using the CDC

Sensitivities, specificities, predictive values, and interrater reliability are presented in Table 2. GST exhibited the highest sensitivity (89.04%), while culture had the lowest (46.57%). Specificity was highest in culture (98.67%). PCR demonstrated the highest Kappa index value (0.7704). The Kappa index showed associations between different tests, with moderate co-occurrence between RUT and PCR.

Table 2.
Characteristics of the H. pylori infection diagnostic methods evaluated using the CDC as a reference

Diagnostic test	Sample (n [%])	Sensitivity (% [CI]	Specificity (% [CI])	PPV (% [CI])	NPV (% [CI])	Kappa CDC
RUT						
Positive	128 (43.1)	78.77 (71.07–84.92)	91.39 (85.43–95.15)	89.84 (82.94–94.26)	81.65 (74.82–87.02)	0.7030
Negative	169 (56.9)					
GST						
Positive	188 (63.3)	89.04 (82.54–93.41)	61.59 (53.30–69.28)	69.15 (61.94–75.56)	85.32 (76.96–91.12)	0.5039
Negative	109 (36.7)					
Culture						
Positive	70 (23.6)	46.58 (38.35–54.99)	98.68 (94.80–99.77)	97.14 (89.14–99.50)	65.64 (59.02–71.72)	0.4564
Negative	227 (76.4)					
PCR						
Positive	124 (41.7)	80.82 (73.30–86.68)	96.03 (91.17–98.37)	95.16 (89.32–98.02)	83.82 (77.28–88.80)	0.7704
Negative	173 (58.3)					

n (%)Number of observations and percentage in each category.
CI: 95% confidence intervals; PPV: Positive predictive value; NPV: Negative predictive value; PCR: Polymerase Chain Reaction; RUT: Rapid Urease Test; GST: Giemsa Staining.

Comparison

- Diagnostic tests and CDC correlations were confirmed through Chi-squared tests, with Cramer's V and uncertainty coefficient indicating significant associations and reduced error prediction rates (Table 3). Significant correlations were also observed between bacterial load and epidemiological characteristics (Table 4).
- Diagnostic tests: It showed that the samples were correlated with a moderate degree of association between the test, with RUT-PCR being the ones with the highest association (0.587) (Table 3).
- Diagnostic tests and CDC: The association values for Cramer's V were highest in PCR (0.779) and RUT (0.708), and the uncertainty coefficient shows that the error when forecasting the other variable decreases by 51.1% and 40.4%, respectively.
- Bacterial Load and Epidemiological Characteristics: CDC and bacterial load showed moderate to high association (Cramer's V = 0.625) with an uncertainty coefficient of 33.6%. Endoscopy, culture, and PCR showed moderate associations (average Cramer's V = 0.48). Sex, age, and pathology showed lower Cramer's V and UC values, despite significant Chi-squared results.
- Genotypes and Epidemiological Characteristics: Chi-squared test showed significant correlation between diagnostic tests, CDC, and genotypes, with high association (Cramer's V = 0.780) and a 52.6% reduction in prediction error for CDC. CDC predicted the presence of bacteria but did not vary between genotypes.

Table 3.
Association measures between diagnostic tests and CDC

Cramer's V	Uncertainty coefficient				
	RUT	GST	Culture	PCR	CDC
RUT	---	0.055	0.059	0.267	0.404
GST	0.268	---	0.103	0.087	0.215
Culture	0.270	0.324	---	0.143	0.246
PCR	0.587	0.333	0.415	---	0.511
CDC	0.708	0.525	0.533	0.779	---

CDC: Case Definition Criteria; PCR: Polymerase Chain Reaction; RUT: Rapid Urease Test; GST: Giemsa Staining.

Table 4.
Association measures between the bacterial load and the epidemiological characteristics of the study

Characteristics	Chi-squared test	Cramer's V	UC
Sex	0.301	0.111	0.007
Age	0.013	0.153	0.029
Pathology	0.226	0.121	0.009
CDC	0.000	0.625	0.336
Endoscopy	0.000	0.463	0.165
Culture	0.000	0.428	0.17
PCR	0.000	0.551	0.172

CDC: Case Definition Criteria; PCR: Polymerase Chain Reaction; UC: Uncertainty coefficient.

Risk analysis

No significant difference was found between *H. pylori* infection and clinical diagnosis across tests. Cramer's V indicated a slight correlation with unified pathology diagnosis, and the low odds ratio (1.192) suggested that there is no significant increase in risk related to CDC diagnosis. No notable association between infection status in diagnostic tests and predisposition to preneoplastic lesions was detected.

Discussion

H. pylori infection is implicated in various gastrointestinal disorders such as gastritis, gastroduodenal ulcers, and gastric cancers, leading to the development of multiple diagnostic methods to identify the infection (17). Each method has its advantages and disadvantages regarding the processing procedure, length of time for results, limitations, sensitivity, specificity, and cost (18).

GST and RUT are preferred for diagnosing *H. pylori* infection due to their low cost, simplicity, and reliability. Although effective, methods like culture and PCR are more expensive, take longer to produce results, and require specialized equipment and trained personnel, which are often unavailable in the country's public endoscopy units or hospitals, limiting their widespread use in clinical practice (17). The variety of tests that can be used to diagnose this infection is significant, and it can be confusing to define which test, or combination, can be considered a highly efficient method to improve the detection of this pathogen (19). In Colombia, the diagnosis is based mainly on detecting the microorganism in histological preparations and on the RUT in biopsy specimens, and bacterial culturing is recommended when a therapeutic failure has occurred (20).

In the present study, the comparative analysis between GST and CDC exhibited the highest sensitivity (89%) and NPV (85.32%), aligning with existing literature (21). This higher sensitivity may be attributed to a lower number of false-positive cases, resulting in an increased number of true-positive cases. However, the values of Cramer's V and UC indicated that these results had a less accurate true/false negative ratio. Previous studies have reported GST specificity values ranging from 83%–90% (21,22).

In terms of specificity, our study reported the lowest value compared to other tests, with previous studies reporting values between 90%–100% (21,23). The sensitivity and specificity of histological examination in diagnosing *H. pylori* are often influenced by variables such as the number, location, and size of collected biopsies, leading to variability in results (11,22).

The updated Sydney system recommends taking biopsies from five different sites for accurate diagnosis of gastritis and *H. pylori*: two from the antrum, two from the body, and one from the incisura. A higher number of samples reduces false negatives due to sampling errors or inadequate bacterial distribution, ultimately increasing the sensitivity and specificity of the tests.

Our study found that the analysis between culture and CDC had the highest PPV (97.14%) and specificity (98.67%) despite having a lower KI, indicating a higher number of actual negative cases but a less accurate true/false-positive ratio, consistent with previous studies (10,21). Culturing *H. pylori* remains a debate due to its growth requirements, which can make isolating the bacterium challenging, such as specific temperature, microaerophilic conditions, and nutritional needs, resulting in slow growth rates (18).

The scarcity of materials and supplies needed to cultivate *H. pylori* in diagnostic labs in developing countries adds to the challenge (8). Although the culture is an expensive, complicated, and time-consuming test for *H. pylori* detection, it is essential in clinical practice. Culture provides insights into bacterial growth characteristics, genetic diversity, and epidemiology and is the recommended gold standard for antibiotic susceptibility testing (17).

However, negative culture results do not definitively exclude the possibility of *H. pylori* infection. This limitation highlights the necessity of assessing the concordance between GST and culture, which can yield a lower agreement, as evidenced by the Kappa Index (KI) of 0.232 reported in this study. Several factors contribute to the observed discordance, including the technician's skill and experience, the quality of the sample collected, exposure to aerobic conditions, and the transport conditions of the samples (17).

RUT demonstrated a specificity of 91.39% and sensitivity of 78.76%, which was lower than reported by other studies (24). While RUT sensitivity is typically high, above 90%, and specificity values rarely drop below 95% (25), various factors like biopsy condition and disease type can impact RUT results (25). False positive and false negative outcomes are possible, with rare occurrences of false positives due to other urease-active microorganisms besides *H. pylori* such as *Proteus mirabilis, Klebsiella pneumoniae, Citrobacter freundii, Enterobacter cloacae, Staphylococcus aureus, Streptococcus salivarius, Staphylococcus capitis urealiticum* (10,26). False negatives can result from factors that reduce bacterial load, though taking multiple biopsies, especially from different stomach areas, can enhance diagnostic accuracy (21).

Our study reported high PPV and NPV values for RUT, indicating reliable results for both positive and negative test outcomes. However, some authors have reported higher PPV values above 95% for RUT, while NPV values can vary from 50% to 90%, influenced by factors like reduced bacterial density from antibiotics and bismuth-containing compounds (5,21).

Several studies have highlighted PCR as a highly sensitive and specific test for *H. pylori* detection (12,21). In our study, PCR demonstrated a sensitivity of 80.82% and specificity of 96.02%, consistent with previous literature citing high sensitivity and specificity values (>95%) (17,19). Moreover, PCR and other molecular methods have been reported to offer high sensitivity and specificity, around 80% and 100% respectively (12), corroborating our study results.

Additionally, high PPV and NPV values (95% and 82% respectively) were obtained in our study, comparable to findings by Kismat et al. (23). The validation with PCR in our study proved essential for accurate diagnosis, with PCR and RUT showing strong agreement in KI, highest Cramer association values, and reduced UC

values, standing out against CDC as a primary recommendation. Real-time PCR testing for *H. pylori* has been shown to accurately identify infected patients, preventing false negatives from other tests (27).

Even though PCR can detect DNA pieces of dead bacteria, resulting in false positives, its effectiveness also depends on the need for special skills, an accurate primer design, and a proper gene selection, which are critical for a successful PCR reaction (11). PCR-based diagnosis, according to Ansari and Yamaoka (11), can be considered a reference method by designing specific primers targeting multiple *H. pylori* genes, allowing for the detection of virulence factors and genetic variability critical for understanding clinical differences among *H. pylori* strains associated with virulence. For instance, the detection of virulence factors by PCR helps evaluate the genetic variation within this pathogen's virulence factors. It gives more information to understand the clinical disparities between patients infected with different strains of *H. pylori* (12).

Our study identified specific virulence factor genotypes, such as *cagA, cagE*, and *vacA*, linked to severe gastric inflammation and diseases like peptic ulcers and gastric cancer (5). The genotype showed the highest association of the data in women with NAG and negative classification, followed by low pathogenicity (*cagA - cagE - vacA s2m2*) between 51–70 and 31–50 years, respectively. In Colombia, molecular methods are not commonly used in routine practice due to their difficulty in determining the biological viability of the bacteria, in addition to their limited availability (28).

Even though in our study, the precision of PCR in identifying *H. pylori* by sequencing essential bacterial genes related to colonization and pathogenicity has been discussed, we should be conscious that in daily routine, this might increase diagnostic challenges due not only to its higher costs and reagents difficulting and limiting other resources but time-consuming, the need of skilled expertise to interpret results accurately and careful sample collection. According to current guidelines in Colombia, endoscopy samples from the antrum and gastric body should undergo histology or rapid urease test (29,30); however, based on our results, for *H. pylori* identification, we strongly recommend undergoing the samples to both tests to ensure a higher precision in the diagnostic before to start an antibiotics treatment.

The genotypic analysis compared with CDC indicated a strong correlation between CDC predictions of bacterial presence or absence, regardless of genotype classification. To comprehensively understand genotype variations and their relationships with pathological conditions, thorough systematic analyses are warranted. Our study's findings on the association of genotypes with age groups and sexes offer insights that complement previous studies (31), suggesting varied risks for conditions like cancer and dysplasia based on genotype and demographic factors. Notably, considering bacterial load values, while variables like sex and pathology showed random distribution, CDC values indicated a higher probability of negative cases with increased bacterial load, emphasizing the importance of sufficient bacterial load for reliable diagnostic outcomes.

Our study highlighted the complex nature of *H. pylori* pathogenicity and the need for a more comprehensive understanding of its infective mechanisms. While diagnostic techniques like RUT and PCR are valuable in reducing false positive results and unnecessary interventions, we should also focus on implementing preventive measures by gaining deeper insights into *H. With pylori's* pathogenic processes, we can develop more effective strategies for prevention and focus more on reducing infection rates associated with health risks. Collaborative use of multiple tests can help clinicians and patients establish appropriate diagnostic routines, mitigating biases in clinical decision-making. While no single method can serve as a definitive reference standard, further research is crucial to explore innovative preventive strategies, such as targeted interventions in high-risk populations, improvement of sanitation practices, and public health education programs,

which could significantly impact the prevalence and management of *H. pylori* infection.

Conclusions

Our study underscores the necessity of integrating multiple diagnostic methods to achieve accurate detection of *H. pylori*, given its complexity. The combination of PCR and RUT has shown significant promise, particularly in relation to clinical outcomes across various demographics. We emphasize the need to refine diagnostic strategies in Colombia, focusing on optimizing detection methods that consider cost-effectiveness, availability, and accuracy within diverse healthcare settings.

Additionally, enhancing public health education programs will be critical in improving the management and prevention of *H. pylori* infections. When conditions are optimal, we recommend using both PCR and RUT tests for *H. pylori* identification. However, in less ideal circumstances, we strongly advocate for conducting both RUT and GST on samples to ensure greater diagnostic precision before initiating antibiotic treatment.

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Conflicts of interest

The authors declare no conflicts of interest.

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