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# Assessing Antibiotic Resistance in *Helicobacter pylori*: Comparative Insights from Phenotypic and Genotypic Resistance Testing

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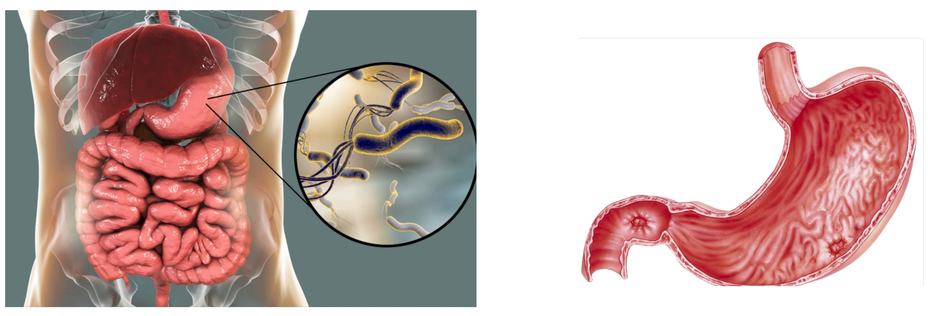
## Introduction

*Helicobacter pylori* is a Gram-negative bacterium recognized as a major causative agent of peptic ulcer disease (Figure 1). Antibiotic resistance in *H. pylori* remains a major challenge in clinical management, particularly for clarithromycin (CLA) and levofloxacin (LVX). While phenotypic methods such as the E-test are widely used in routine practice, genotypic assays targeting resistance-associated mutations provide rapid and precise alternatives. This study aimed to compare phenotypic and genotypic resistance testing in *H. pylori* isolates.

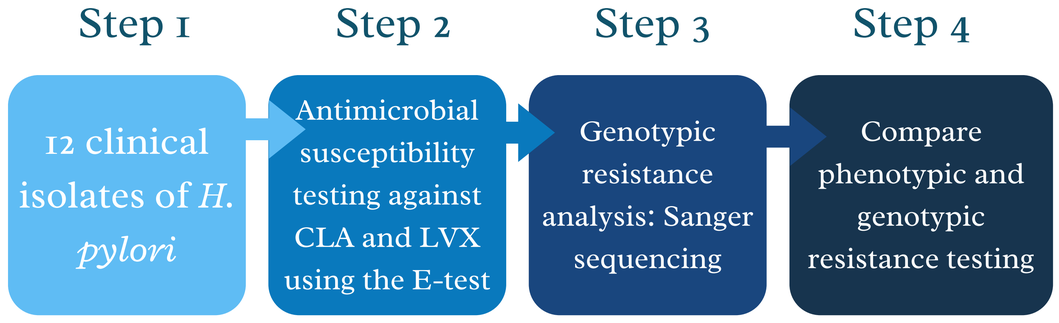
Figure 1: *H. pylori* infection as a major cause of peptic ulcer disease.



## Methodology

Twelve clinical isolates of *H. pylori* were subjected to antimicrobial susceptibility testing against CLA and LVX using the E-test. In parallel, genotypic resistance analysis was performed by Sanger sequencing (Figure 2). Sequencing targeted the *23S rRNA* gene, focusing on mutations A2142G and A2143G associated with CLA resistance, and the *gyrA* gene, focusing on amino acid substitutions at codons N87K and D91N/Y linked to LVX resistance (Figure 3). Results of phenotypic and genotypic testing were compared to assess concordance.

Figure 2. Schematic representation of the methodology employed in this study.




## Results

For clarithromycin, concordance between E-test and genotypic testing was observed in 10 of 12 isolates. Two isolates exhibited phenotypic resistance without detectable *23S rRNA* mutations. For levofloxacin, concordance was also 10 of 12 isolates: one isolate harbored resistance-associated *gyrA* mutations despite phenotypic susceptibility, while another demonstrated phenotypic resistance in the absence of genotypic alterations. Overall, 20 of 24 comparisons (83.3%) were concordant across both antibiotics (Table 1).

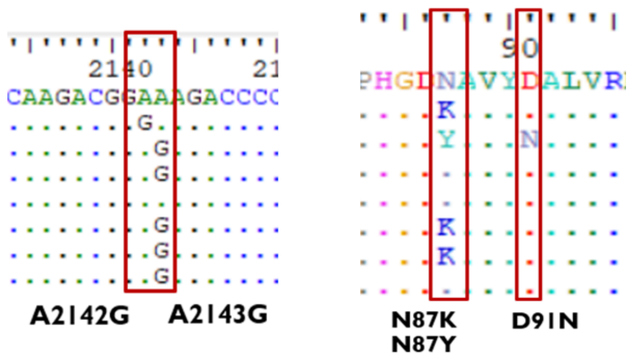



Figure 3: Mutations in *23S rRNA* (left) and *gyrA* (right) genes associated with resistance to clarithromycin (CLA) and levofloxacin (LVX).

Table 1. Comparison of phenotypic and genotypic resistance profiles among *H. pylori* isolates.

| Isolate # | Clarithromycin | | Levofloxacin | |
|---|---|---|---|---|
| | E-test | Genetic testing | E-test | Genetic testing |
| 1 | S | S | S | S |
| 2 | R | R | S | R |
| 3 | R | R | R | R |
| 4 | R | R | R | R |
| 5 | R | S | R | S |
| 6 | R | R | R | R |
| 7 | R | R | R | R |
| 8 | R | R | S | S |
| 9 | R | S | S | S |
| 10 | R | R | R | R |
| 11 | R | R | R | R |
| 12 | R | R | S | S |

## Conclusion

This study demonstrated a high degree of concordance between phenotypic and genotypic methods for detecting clarithromycin and levofloxacin resistance in *H. pylori*. Genotypic testing offers the advantage of rapid, mutation-specific detection, independent of bacterial growth conditions, and can uncover resistance even when phenotypic results are ambiguous. The observed discordances underscore the limitations of relying solely on a single method. Incorporating genotypic assays into routine diagnostics holds the potential to enhance the precision of resistance detection, enabling more effective and individualized antimicrobial therapy for *H. pylori* infection.

## References

- Contreras M, Mujica H, Garcia-Amado M. Molecular tools of antibiotic resistance for *Helicobacter pylori*: an overview in Latin America. Frontiers in Gastroenterology. 2024;3.
- Samanic I, Dadic B, Sanader Marsic Z, Dzelalija M, Maravic A, Kalinic H, et al. Molecular Characterization and Mutational Analysis of Clarithromycin- and Levofloxacin-Resistance Genes in *Helicobacter pylori* from Gastric Biopsies in Southern Croatia. Int J Mol Sci. 2023;24(19).