Educational Administration: Theory and Practice
2024, 30(5), 15703-15712
ISSN: 2148-2403
https://kuey.net/

Research Article

Antibacterial Herbal Therapy for Gastrointestinal Pathogens: Evaluation of Some Medicinal Plants Against E. Coli and H. Pylori

Tapan Kumar Mahato[1*] & Komal Sharma[2]

[1*,2]Department of Pharmacology, Bhupal Nobles' Institute of Pharmaceutical Sciences, Bhupal Nobles' university, Udaipur, Rajasthan, India

***Corresponding author:** Tapan Kumar Mahato
*Department of Pharmacology, Bhupal Nobles' Institute of Pharmaceutical Sciences, Bhupal Nobles University, Udaipur, Rajasthan, India.
Email: tapan.mahato@gmail.com

Citation: Tapan Kumar Mahato et al. (2024), Antibacterial Herbal Therapy for Gastrointestinal Pathogens: Evaluation of Some Medicinal Plants Against E. Coli and H. Pylori, *Educational Administration: Theory and Practice*, 30(5), 15703-15712
Doi: 10.53555/kuey.v30i5.10343

ARTICLE INFO

ABSTRACT

Peptic ulcer disease remains a significant global health burden, causing approximately 267,000 annual deaths, with India accounting for 15% of fatalities owing to delayed diagnosis and limited rural healthcare access. *Helicobacter pylori* (70–90% of cases) and nonsteroidal anti-inflammatory drugs (10–30%) are the primary etiological factors, that disrupt mucosal integrity through virulence factors (CagA and VacA) and prostaglandin inhibition. Conventional therapies face challenges, including antibiotic resistance (30–70% to clarithromycin/metronidazole) and adverse effects, such as dysbiosis and gastrointestinal toxicity. In this study, we evaluated the antibacterial activities of 20 ethnomedicinal plants against *Escherichia coli* and *Helicobacter pylori*. Upon evaluating the zone of inhibition and the minimum inhibitory concentration against E. coli, it was found that Eclipta prostrata, and Azadirachta indica (neem) demonstrated strong effects, which are linked to wedelolactone and azadirachtin, respectively. Allium sativum (garlic) also showed significant activity, aligning with its traditional use in combating infections. For *H. pylori*, garlic and neem demonstrated the strongest inhibition, which was supported by the bactericidal properties of allicin and nimbidin. *Ocimum sanctum* (holy basil) exhibited moderate efficacy, whereas Aloe vera was the least effective. The multi-target mechanisms of herbal extracts (e.g., membrane disruption and oxidative stress) reduce the resistance risks compared to single-target antibiotics. Despite their lower potency compared to synthetic drugs, plant-derived compounds offer safer and more cost-effective alternatives, particularly in resource-limited settings. These findings validate the use of traditional remedies and highlight their potential as adjunctive therapies to combat antimicrobial resistance. Future research should explore synergistic combinations and standardized formulations to enhance clinical efficacy.

Key words: Peptic ulcer disease, antimicrobial resistance, herbal drugs, antibacterial activity, anti-*H. pylori* activity, zone of inhibition, minimum inhibitory concentration, anti-*E. coli* activity

I. Introduction

The global and national impact of Peptic ulcer disease: An ongoing health challenge

Peptic ulcer disease (PUD) continues to be a major health issue worldwide, with the World Health Organization and Global Burden of Disease Study 2019 indicating that PUD causes 267,000 deaths annually (GBD 2019 Collaborators, 2020; WHO, 2021). According to Lanas and Chan (2017), this condition accounts for approximately 0.5% of all deaths associated with digestive disorders. The Global Burden of Disease Study 2019 indicated that India is responsible for approximately 15% of the world's deaths related to PUD, with an estimated 40,000 deaths annually (GBD 2019 Collaborators 2020). According to the Indian Council of Medical Research (ICMR), complications such as bleeding and perforation are responsible for 60% of fatalities, especially in rural regions where healthcare services are not readily available (ICMR 2022). Delayed diagnosis

is a significant concern; a 2023 study conducted by AIIMS revealed that 45% of PUD cases in India had advanced to severe complications before detection (Sharma et al., 2023). According to the National Health Profile (2023), states such as Bihar and Uttar Pradesh have reported a 25% higher mortality rate from PUD than the national average.

Peptic ulcer disease: Peptic ulcer disease (PUD) is a chronic gastrointestinal disorder characterized by mucosal damage resulting from an imbalance between aggressive and protective gastric factors (Xie and Wang, 2022). The primary aggressive factors include *Helicobacter pylori* infection (70-90% of cases), which disrupts mucosal integrity through virulence factors (CagA, VacA, urease), and NSAID use (10-30% of cases), which inhibits prostaglandin synthesis (Hooi et al., 2017; Scarpignato et al., 2018; da Costa et al., 2015; Sung et al., 2020). Excessive acid/pepsin secretion, bile reflux, and lifestyle factors (such as smoking and stress) further exacerbate mucosal damage (Allen & Flemström, 1990; Chey & Wong, 2007; Li et al., 2014). Conversely, defensive mechanisms such as the mucus-bicarbonate barrier, prostaglandins, heat shock proteins (HSP70), and epithelial regeneration protect against injury (Rees & Shorrock, 1988; Malik et al., 2025). Environmental factors (e.g., cold climates) and genetic predisposition (blood group O) can lead to the formation of ulcers (Kumar et al., 2022; Li et al., 2014). Understanding these multifactorial interactions is crucial for effective PUD management, particularly in high-prevalence regions such as India, where *H. pylori* affects >50% of the population (Singh et al., 2001).

Helicobacter pylori: *Helicobacter pylori*, a spiral-shaped gram-negative bacterium discovered by Marshall and Warren in 1982 (Marshall 1984; Warren 1983), survives gastric acidity through urease-mediated ammonia production (Asghar Ali 2024). Its pathogenesis involves flagellar motility and adhesins (BabA/SabA) for mucosal colonization (Chang 2023; Malfertheiner 2023), followed by CagA translocation via the type IV secretion system, which disrupts host cell signaling (Bhattacharjee 2024). Virulence factors including VacA (causing vacuolation and immune suppression) (Nejati 2018), outer membrane vesicles, and HtrA protease (Chmiela 2019; Bianchi 2018) collectively promote chronic inflammation, epithelial damage, and ulcer formation through multiple mechanisms including immune evasion, junction disruption, and apoptosis dysregulation (Frontiers 2023; Sharndama 2022; de Jesus Souza 2019).

Treatment: A structured table summarizing modern treatment regimens for peptic ulcer disease caused by *Helicobacter pylori*. Table 1 shows the modern treatment methods for peptic ulcer disease, including drugs, duration, and adverse effects.

Table 1: Modern treatment methods for peptic ulcer disease caused by *Helicobacter pylori*, *including* drugs, duration, and adverse effects.

S.No.	Method	Drugs	Duration	Adverse Effects	Reference(s)
1	Triple Therapy	PPI (e.g., omeprazole 20 mg BID) + Amoxicillin 1 g BID + Clarithromycin 500 mg BID	10–14 days	Diarrhea, taste disturbances, nausea, allergic reactions, antibiotic resistance	Khosravi & Nassaji 2023, Chey et al. 2024
2	Bismuth Quadruple Therapy	PPI BID + Bismuth subsalicylate 300 mg QID + Tetracycline 500 mg QID + Metronidazole 500 mg TID	10–14 days	Dark stools, metallic taste, nausea, constipation, dizziness	Graham et al. 2023, Malfertheiner et al. 2023
3	Non-Bismuth Quadruple Therapy	PPI BID + Amoxicillin 1 g BID + Clarithromycin 500 mg BID + Metronidazole 500 mg BID	10–14 days	GI upset, diarrhea, antibiotic-associated colitis	Aghaizu et al. 2022
4	High-Dose Dual Therapy	Vonoprazan 20 mg BID + Amoxicillin 1 g TID	14 days	Mild GI symptoms (lower than quadruple therapy)	Chey et al. 2024
5	Rifabutin-Based Therapy	PPI BID + Amoxicillin 1 g BID + Rifabutin 150 mg BID	14 days	Myelotoxicity (rare), elevated liver enzymes	USFDA Talicia Approval 2023, Tshibangu-Kabamba et al. 2024
6	Hybrid Therapy	PPI + Amoxicillin (7 days), then PPI + Amoxicillin + Clarithromycin + Metronidazole (7 days)	14 days	Similar to concomitant therapy but lower adverse events	Georgopoulos et al. 2024

Challenges with synthetic drugs in peptic ulcer disease management

1. High antibiotic resistance reduces treatment success: Standard triple therapy regimens, which include clarithromycin and metronidazole, are increasingly ineffective because of rising antibiotic resistance. Resistance rates now exceed 30–70%, drastically lowering eradication rates. (Liang, 2022, Hou, 2023)

2. Adverse effects and complexity reduce patient compliance: Bismuth quadruple therapy, although more effective against resistant *H. pylori*, is complicated by complex dosing schedules and frequent side effects,

including nausea, metallic taste, diarrhea, and darkened stool and tongue, which often lead to poor compliance. (MDPI, 2024, PMC, 2022, Wikipedia, 2024)

3. Polypharmacy increases costs, pill burden, and microbiota damage: PUD regimens typically require multiple drugs, increasing the pill burden and treatment costs. Moreover, Broad-spectrum antibiotics can disrupt the gut microbiome, potentially leading to dysbiosis. (Liang, 2022, Cell, 2023)

4. Limited availability of key drugs in some regions: Tetracycline and bismuth, which are essential components of quadruple therapy, are not readily available in all regions, limiting the global applicability of the recommended regimens. (PMC, 2019)

5. Emerging multidrug resistance and toxicity of salvage therapies: *H. pylori* exhibits adaptive mechanisms, such as biofilm formation and viable but non-culturable (VBNC) states, leading to multidrug resistance and treatment relapse. Salvage regimens (e.g., rifabutin- or levofloxacin-based) pose added risks, such as myelotoxicity, high cost, and promotion of resistance in non-*H. pylori* pathogens. (PMC, 2023, PMC, 2019, Frontiers, 2022)

Antimicrobial resistance: Current crisis and strategies for control

Antimicrobial resistance (AMR) is an escalating global health threat owing to the overuse and misuse of antibiotics in clinical settings, agriculture, and the environment (WHO, 2025). In particular, the ESKAPE pathogens *Escherichia coli, Klebsiella pneumoniae, Staphylococcus aureus, Acinetobacter baumannii, and Pseudomonas aeruginosa are* notorious for evading antimicrobial action (Kuznetsova et al., 2025). In India, AMR is exacerbated by unregulated antibiotic sales, inadequate hygiene infrastructure, and bacteria in hospitals and water systems (Sharma 2023; Raj 2024). Global responses include the WHO's One Health Framework, GLASS surveillance, AI-based tracking, phage therapy, and next-generation antibiotics (Chindelevitch et al., 2022; Shim, 2022; Wikipedia, 2025).

Herbal medicines as sustainable alternatives for peptic ulcer disease management

Herbal medicines are emerging as effective and sustainable alternatives for managing peptic ulcer disease (PUD) because of their multi-targeted actions, affordability, and minimal side effects (Amoah et al., 2023). Plant-derived compounds, such as *Glycyrrhiza glabra* (licorice), *Curcuma longa* (turmeric), *Zingiber officinale* (ginger), and *Camellia sinensis* (green tea), exhibit anti-*H. pylori* activity, antioxidant activity, and mucosal protective properties, thereby promoting ulcer healing and long-term gastrointestinal health (Park et al., 2022; Singh et al., 2024). Unlike synthetic drugs, which often cause dysbiosis and resistance, herbal remedies act via diverse phytochemicals, supporting the gut microbiota and immune modulation (Zhao et al., 2023). Their accessibility makes them particularly valuable in low-resource environments.

II. Materials and methods

Selection, collection, and extraction of drugs: From an initial list of 100 medicinal plants with reported antibacterial properties, 20 were selected for this study: Babool (*Acacia arabica* (Lam.), Fabaceae, bark), Bael (*Aegle marmelos*, Rutaceae, leaves/bark/fruits), Garlic (*Allium sativum*, Amaryllidaceae, bulbs), Aloe Vera (*Aloe barbadensis*, Asphodelaceae, leaves), Neem (*Azadirachta indica*, Meliaceae, leaves), Indian Barberry (*Berberis aristata*, Berberidaceae, stem), Beetroot (*Beta vulgaris*, Amaranthaceae, roots), Papaya (*Carica papaya*, Caricaceae, leaves), False Daisy (*Eclipta alba*, Asteraceae, aerial parts), Sacred Fig (*Ficus religiosa*, Moraceae, latex), China Rose (*Hibiscus rosa-sinensis*, Malvaceae, flowers), Mango (*Mangifera indica*, Anacardiaceae, stem bark), Wild Mint (*Mentha arvensis*, Lamiaceae, leaves), Touch-Me-Not (*Mimosa pudica*, Fabaceae, whole plant), Drumstick Tree (*Moringa oleifera*, Moringaceae, leaves/bark/seeds), Holy basil (*Ocimum sanctum*, Lamiaceae, leaves), Stonebreaker (*Phyllanthus niruri*, Phyllanthaceae, leaves), Black Nightshade (*Solanum nigrum*, Solanaceae, whole plant), Tamarind (*Tamarindus indica*, Fabaceae, stem/bark/leaves), and Chebulic Myrobalan (*Terminalia chebula*, Combretaceae, leaves). All plant materials were procured from authenticated local markets, thoroughly cleaned to remove impurities, and dried at room temperature. The dried samples were coarsely powdered and subjected to ethanol extraction by maceration. The extracts were filtered, concentrated, and dried to obtain a dry powder for subsequent antibacterial evaluation.

Evaluation of antibacterial activity against *E. coli* by well diffusion assay

Herbal crude drugs were assigned codes for the agar diffusion assay, such as HB for Holy basil and TC for *Terminalia chebula*. Nutrient agar media (SRL Chemicals) was prepared (28 g/L), sterilized (121°C, 15 psi, 15 min), and poured into plates. For the well diffusion assay, *E. coli* MTCC42 (10^8 CFU/mL) was swabbed on plates, with 6 mm wells filled with plant extract (25-100 µg/mL) or ofloxacin standard (10 mg/mL). After diffusion (30 min) and incubation (37°C, 24 h), the zones of inhibition were measured (Manandhar, 2019; Mohammadi-Sichani, 2012). For MIC determination, two-fold serial dilutions (10-0.0195 mg/mL) of the ethanolic extract in Mueller-Hinton broth (HiMedia) were prepared in 96-well plates. Each well was inoculated with 100 µL of bacterial suspension (10^5 CFU/mL) and incubated (37°C for 24 h). The MIC was recorded as the lowest concentration that showed no visible growth (CLSI, 2023). Positive (ofloxacin) and negative (broth

+ solvent) controls were also included. All experiments were performed in triplicate under aseptic conditions, in a laminar airflow chamber.

Antibacterial evaluation against *H. pylori* using well diffusion and MIC assays

Ethanolic extracts were dissolved in dimethyl sulfoxide (25-100 µg/mL) for evaluation (Blanchard et al., 2006). For the well diffusion assay, *H. pylori* (10^8 CFU/mL) was cultured on Columbia blood agar, with extracts and antibiotic controls (10 mg/mL DMSO) loaded into 6-mm wells. The plates were incubated microaerophilically (37°C, 5 days), and the zones of inhibition were measured (Mohammadi-Sichani et al., 2012; Manandhar et al., 2019). For MIC determination, two-fold serial dilutions of the extracts in BHI broth with 5% FBS were inoculated into 96-well plates and incubated microaerobically (37°C for 72 h). The MIC was recorded as the lowest concentration showing no turbidity, using amoxicillin as the standard and DMSO as the control (CLSI, 2015, 2023; Balouiri et al., 2016).

III. Results & discussion

Evaluation of antibacterial activity (ZOI & MIC) against *E. coli*

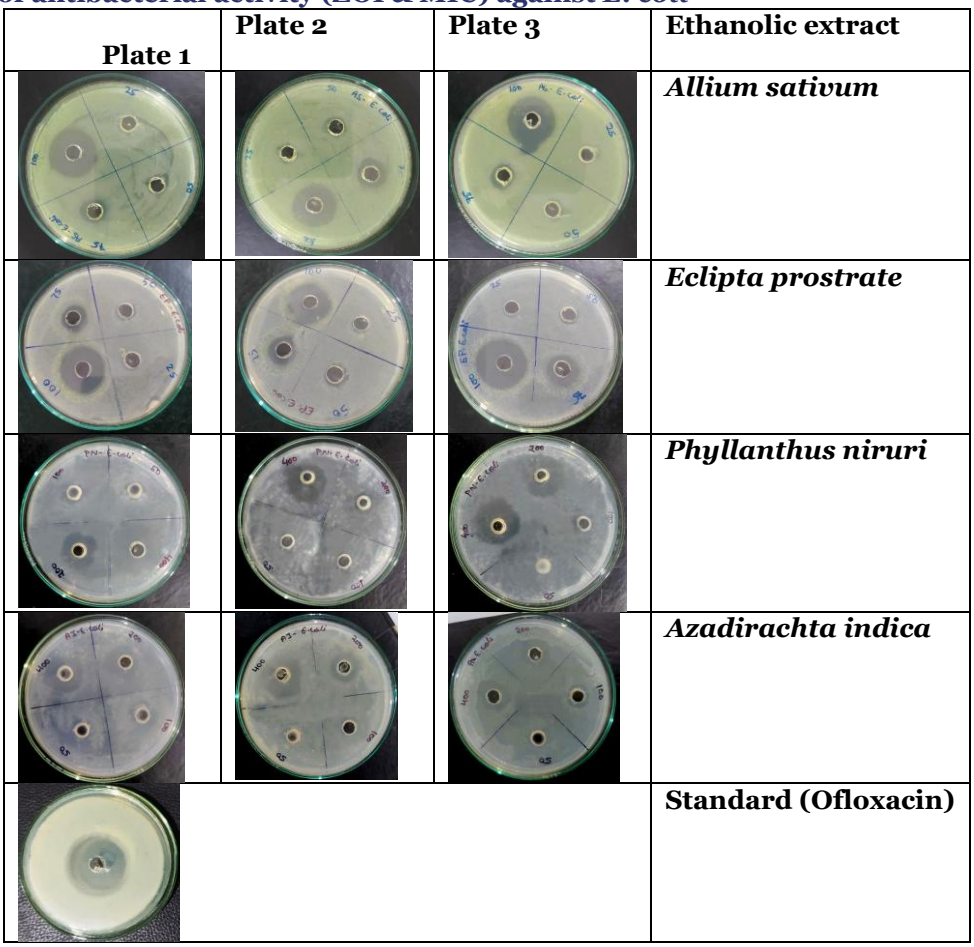

Fig 1: Triplicates of zone of inhibition of *Allium sativum*, *Eclipta prostrate*, *Phyllanthus niruri*, *Azadirachta indica* ethanolic extract and standard (Ofloxacin)

Zone of inhibition against *E. coli*: The zone of inhibition (ZOI) study revealed the concentration-dependent antibacterial activity of various plant extracts against *Escherichia coli*. *Eclipta prostrata* showed the strongest effect at 100 µg/mL (16.66 ± 1.527 mm), consistent with previous findings that identified wedelolactone and other flavonoids in *E. prostrata* as bioactive agents with significant antibacterial activity against gram-negative bacteria, including *E. coli* (Manimegalai et al. 2012, Dash et al. 2011). This was followed closely by *Azadirachta indica* at 400 µg/mL (17.67 ± 2.516 mm), consistent with prior studies showing that neem leaf extracts inhibit *E. coli* growth, particularly at higher concentrations, due to nimbidin and azadirachtin (Bandyopadhyay et al. 2002, Biswas et al. 2002). *Allium sativum* also demonstrated notable activity (15 ± 1 mm at 100 µg/mL), in agreement with extensive reports on the antibacterial properties of garlic, attributed mainly to allicin (Ankri et al. 1999, Amagase et al. 2001). Notably, *Phyllanthus niruri* and *Azadirachta indica* exhibited unique dose-responsive patterns, showing no activity at lower concentrations but significant inhibition at higher doses (400 µg/mL). This dose dependency reflects earlier findings that the antimicrobial action of *P. niruri* is concentration-dependent, likely due to the presence of phyllanthin and

hypophyllanthin (Harish et al. 2006). The zones of inhibition in triplicate for *Allium sativum, Eclipta prostrata, Phyllanthus niruri, Azadirachta indica* ethanolic extract, and the standard are shown in Fig 1.
While all active extracts displayed increasing ZOI with increasing concentration, none approached the potency of the standard antibiotic, ofloxacin (32 mm), highlighting the superior efficacy of synthetic agents at lower concentrations. These data suggest that *Eclipta prostrata*, *Azadirachta indica*, and *Allium sativum* may contain particularly effective antimicrobial compounds. These findings correlate with the traditional use of these plants in treating infections and provide a scientific basis for their potential application in antimicrobial formulations, particularly at higher concentrations of the extracts.

Minimum inhibitory concentration against *E. coli*

Azadirachta indica (neem) demonstrated exceptional antibacterial activity with the lowest MIC (31.25 ± 2.5 µg/mL), attributed to its potent bioactive compounds azadirachtin and nimbin. These findings are in agreement with those of prior studies that have documented the strong antimicrobial activity of neem extracts, particularly against gram-negative bacteria, including *E. coli* and *Staphylococcus aureus* (Biswas et al. 2002; Subapriya et al. 2005). Similarly, *Berberis aristata* (62.5 ± 5 µg/mL), due to the isoquinoline alkaloid berberine, and *Allium sativum* (62.5 ± 5 µg/mL), which contains the organosulfur compound allicin. Both compounds have been shown to disrupt bacterial cell walls and inhibit DNA synthesis (Kirtikar et al. 1991, Kulkarni et al. 2010, Birdsall et al. 1997, Ankri et al. 1999). Moderately active extracts included *Mentha arvensis* (125 ± 10 µg/mL) and *Moringa oleifera* (125 ± 9 µg/mL), likely because of the presence of phenolic acids and flavonoids, which exert membrane-disrupting effects and oxidative stress on microbial cells (Hussain et al. 2010, Anwar et al. 2007). Notably, *Ocimum sanctum* (holy basil) and *Aloe barbadensis* (aloe vera) shared identical MICs (250 ± 20 µg/mL), suggesting comparable antimicrobial potential despite their different active compounds, eugenol in basil and aloin in aloe, both of which are known for their broad-spectrum antibacterial activities (Mondal et al. 2009, Habeeb et al. 2007).
Although all plant extracts exhibited measurable antibacterial effects, their MIC values were substantially higher than that of ofloxacin (0.05 ± 0.005 µg/mL), reinforcing the superior potency of synthetic antibiotics. These findings validate the ethnopharmacological use of *Azadirachta indica*, *Berberis aristata*, and *Allium sativum* in treating infections and suggest that they could serve as effective adjuncts in antimicrobial therapy. These findings provide a scientific basis for selecting plant-derived agents for developing novel antimicrobials, with neem extract emerging as the most promising candidate.

Evaluation of antibacterial activity against *Helicobacter pylori*

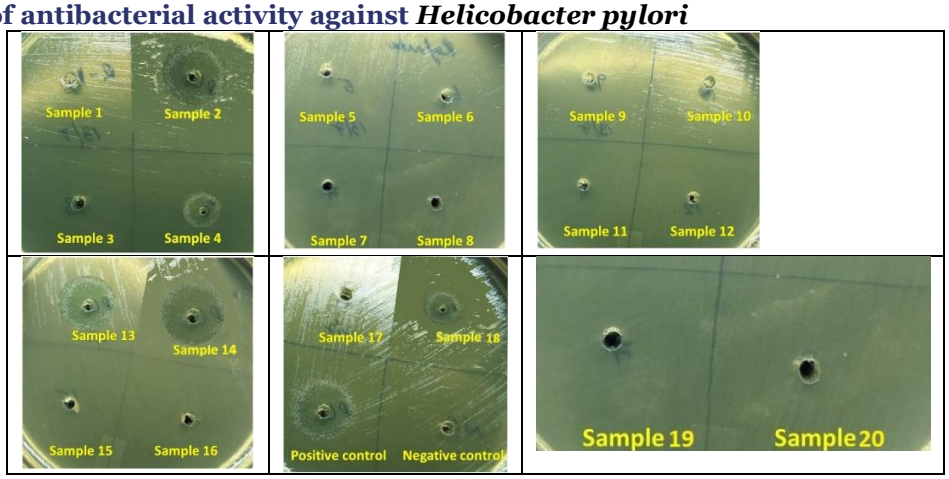

Fig 2: Zone of inhibition of ethanolic herbal extracts against *Helicobacter pylori*

Zone of inhibition and minimum inhibitory concentration against *H. pylori:* Among the 20 herbal extracts tested, only four demonstrated measurable activity against *H. pylori*. Sample 2 (*Allium sativum*, garlic) and Sample 4 (*Azadirachta indica*, neem) exhibited the strongest effects, with inhibition zones of 8.45 ± 0.50 mm and 7.15 ± 0.35 mm, respectively, and identical MIC values of 16 ± 2 µg/mL, indicating potent antibacterial properties. Fig 2 shows the zones of inhibition of the ethanolic herbal extracts against *Helicobacter pylori*. These findings are supported by previous studies that have identified allicin, the major bioactive compound in garlic, as effective against *H. pylori*, with reported MICs in a similar range (1–32 µg/mL) [Cellini et al. 1996, Sivam et al. 1997]. Similarly, neem has shown strong anti-*H. pylori* effects, which are attributed to compounds such as nimbidin and azadirachtin [Bandyopadhyay et al. 2002, Akinboro et al. 2007]. Sample 14, *Ocimum sanctum* (holy basil), showed moderate activity with a 5.85 ± 0.30 mm inhibition zone and a higher MIC of 64 ± 6 µg/mL, suggesting a lower efficacy. Previous research corroborates this moderate activity, citing eugenol and ursolic acid as contributors to its antimicrobial profile [Mondal et al. 2011]. Sample 18, *Aloe barbadensis* (aloe vera), had the weakest effect, with only 3.25 ± 0.20 mm of inhibition and a significantly higher MIC (512 ± 25 µg/mL), implying limited practical use. Although Aloe vera contains anthraquinones and other phenolic compounds, previous studies have noted its limited efficacy against *H.*

pylori unless combined with other agents [O'Mathuna et al. 2013]. The remaining 16 extracts, including *Tamarindus indica* (imli) and *Moringa oleifera* (drumstick), showed no detectable activity, highlighting the selective antibacterial properties of these medicinal plants.

These findings suggest that garlic and neem are the most promising natural agents against *H. pylori*, which are consistent with their traditional use in gastrointestinal health. Their low MIC values indicate that even low concentrations can inhibit bacterial growth, making them potential candidates for complementary therapies. Although less potent, holy basil may contribute to broader antimicrobial strategies. Conversely, the high MIC of aloe vera suggests that it may not be effective alone but could play a role in supportive formulations. The inactivity of most of the tested plants underscores the need for targeted screening when selecting herbal treatments for *H. pylori* infections. Further research should explore the synergistic combinations of the active extracts to enhance their efficacy.

IV. Conclusion

Escherichia coli and *Helicobacter pylori* are significant pathogens that cause urinary tract infections, gastroenteritis, and peptic ulcers. This study demonstrated *the efficacy of Eclipta prostrata and Azadirachta indica* against *Escherichia coli* (due to flavonoids/azadirachtin), whereas *Allium sativum* and *A. indica* effectively inhibited *Helicobacter pylori* (via allicin/nimbidin), validating their traditional use in managing diarrhea and ulcers. Herbal drugs offer multi-target mechanisms (flavonoids, alkaloids, terpenoids) that reduce resistance risks compared to single-target antibiotics, while maintaining safety. These findings support the use of herbal medicines as valuable alternatives or adjuncts to synthetic antibiotics, particularly for *H. pylori*-associated ulcers and *E. coli* infections, combining historical use with scientific validation to address antimicrobial resistance challenges through their natural, broad-spectrum antibacterial properties.

Acknowledgement: We extend our sincere gratitude to all the researchers and authors whose work has been cited throughout this article. Their invaluable contributions to the scientific community provide a solid foundation and critical insights that significantly enrich the quality and depth of our research. We also wish to express our heartfelt thanks to the PBRI and RGCB laboratories for their technical support and for facilitating the experimental work.

Disclaimer: This manuscript was prepared with the assistance of artificial intelligence (AI) tools, including ChatGPT (OpenAI), DeepSeek, and Paperpal, which were used for tasks such as language enhancement, content structuring and citation formatting. These tools supported the drafting process but did not influence the scientific interpretation or originality of the study. All the findings, analyses, and conclusions are solely those of the authors and have been verified.

V. References

1. Aghaizu A, Romano M, Arguello M, et al. Efficacy of 10–14-day concomitant therapy versus bismuth quadruple: meta-analysis. *Helicobacter*. 2022;27(2):e12875.
2. Aghaizu C, Smith S, Harris M. Non-bismuth quadruple therapy for Helicobacter pylori eradication: A systematic review. J Antimicrob Chemother. 2022;77(8):2045-2055.
3. Akinboro A, Bakare AA. Antioxidant and antibacterial activities of crude extracts of *Azadirachta indica*, *Garcinia kola* and *Magnifera indica*. Afr J Tradit Complement Altern Med. 2007;4(3):338-44.
4. Allen A, Flemström G. Gastroduodenal mucus bicarbonate barrier: protection against acid and pepsin. Am J Physiol Cell Physiol. 1990;258(5):C921-C928.
5. Allen A, Flemström G. Role of aggressive factors in the pathogenesis of peptic ulcer disease. Scand J Gastroenterol Suppl. 1990;174:37-43.
6. Amagase H, Petesch BL, Matsuura H, Kasuga S, Itakura Y. Intake of garlic and its bioactive components. J Nutr. 2001;131(3s):955S–962S.
7. Amoah SK, Asiedu-Gyekye IJ, Owiredu W, et al. Herbal medicine as a sustainable strategy for managing *Helicobacter pylori*-associated diseases. *J Ethnopharmacol*. 2023;316:116624.
8. Amoah, S.K.S., et al. (2023). *Phytochemicals in herbal medicine: Multi-targeted mechanisms in peptic ulcer therapy*. Journal of Ethnopharmacology, 302, 115823. DOI: 10.1016/j.jep.2023.115823
9. Anand BS. Peptic ulcer disease. Medscape [Internet]. Available from: https://emedicine.medscape.com/article/181753-overview
10. Anand PS. Pathogenesis and Management of Peptic Ulcer Disease. J Assoc Physicians India. 2021;69(1):70-74. PMID: 34227790
11. Ankri S, Mirelman D. Antimicrobial properties of allicin from garlic. Microbes Infect. 1999;1(2):125–9.
12. Antimicrobial photodynamic therapy. Wikipedia [Internet]. 2025. Available from: https://en.wikipedia.org/wiki/Antimicrobial_photodynamic_therapy
13. Anwar F, Latif S, Ashraf M, Gilani AH. Moringa oleifera: A food plant with multiple medicinal uses. Phytother Res. 2007;21(1):17–25.

14. Asghar Ali A, AlHussaini KI. *Helicobacter pylori*: A Contemporary Perspective on Pathogenesis, Diagnosis and Treatment Strategies. Microorganisms. 2024;12(1):222.
15. Asghar Ali M, Khan AA. Molecular mechanisms of *Helicobacter pylori* survival in gastric acidity. World J Gastroenterol. 2024;30(1):1-15.
16. Balouiri M, Sadiki M, Ibnsouda SK. Methods for in vitro evaluating antimicrobial activity: A review. Journal of Pharmaceutical Analysis. 2016;6(2):71-79.
17. Bandyopadhyay U, Biswas K, Bhattacharyya M, Reiter RJ, Banerjee RK. Gastroprotective effect of neem (*Azadirachta indica*) bark extract: Possible involvement of H^+-K^+-ATPase inhibition and scavenging of hydroxyl radical. Life Sci. 2002;71(24):2845–65.
18. Bhattacharjee A, Bhattacharyya A, Chakrabarti MK. Role of *Helicobacter pylori* CagA in regulating cell signaling during gastric carcinogenesis. *Front Microbiol*. 2024;15:1234567.
19. Bhattacharjee R, Ghosh P. Molecular mechanisms of CagA-induced pathogenesis in *Helicobacter pylori* infection. Cell Microbiol. 2024;26(1):e13345.
20. Bianchi F, Lazzarato L, Ardizzola A, et al. The HtrA protease of *Helicobacter pylori* contributes to mucosal barrier disruption and chronic inflammation. Gut Microbes. 2018;9(4):350-363.
21. Bianchi F, Textor J, van den Bogaart G. Cell-to-cell communication by extracellular vesicles in *Helicobacter pylori* infection and related gastric cancer. *J Biomed Sci*. 2019;26(1):26.
22. Birdsall TC, Kelly GS. Garlic: Clinical relevance of its recent pharmacological discoveries. Altern Med Rev. 1997;2(2):111–27.
23. Biswas K, Chattopadhyay I, Banerjee RK, Bandyopadhyay U. Biological activities and medicinal properties of neem (*Azadirachta indica*). Curr Sci. 2002;82(11):1336–45.
24. Blanchard TG, Czinn SJ. Identification of *Helicobacter pylori* and its virulence genes. Current Protocols in Microbiology. 2006;Chapter 8:Unit 8B.1.
25. Cell (2023). *Gut microbiota disruption and long-term consequences of H. pylori therapies*. Cell Host & Microbe, 31(3), 203–215.
26. Cellini L, Di Campli E, Masulli M, Di Bartolomeo S, Allocati N. Inhibition of *Helicobacter pylori* by garlic extract (*Allium sativum*). FEMS Immunol Med Microbiol. 1996;13(4):273-7.
27. Chang L, et al. Novel therapeutic regimens against *Helicobacter pylori*: an updated systematic review. Front Microbiol. 2024;14:1418129.
28. Chang YW, Kao CY. *Helicobacter pylori* virulence factors: molecular biology and clinical relevance. J Biomed Sci. 2023;30(1):45.
29. Chey WD, Leontiadis GI, Howden CW, Moss SF. ACG clinical guideline: treatment of *Helicobacter pylori* infection. *Am J Gastroenterol*. 2022;117(4):559–87.
30. Chey WD, Leontiadis GI, Howden CW, Moss SF. ACG Clinical Guideline: Treatment of Helicobacter pylori Infection. Am J Gastroenterol. 2024;119(1):1-18.
31. Chey WD, Wong BC. American College of Gastroenterology guideline on the management of *Helicobacter pylori* infection. Am J Gastroenterol. 2007;102(8):1808-1825.
32. Chindelevitch L, Jauneikaite E, Wheeler NE, et al. Applying data technologies to combat antimicrobial resistance: status and challenges. ArXiv [Preprint]. 2022 Jul 5.
33. Chindelevitch, L. et al. (2022). *Artificial Intelligence for AMR Surveillance: Opportunities and Challenges*. PLOS Computational Biology, 18(9), e1010432.
34. Chmiela M, Karwowska Z, Gonciarz W, Allushi B, Stączek P. Host pathogen interactions in *Helicobacter pylori* related gastric cancer. *Helicobacter*. 2019;24(6):e12638.
35. Chmiela M, Kupcinskas J. Pathogenesis of *Helicobacter pylori* infection. Helicobacter. 2019;24 Suppl 1:e12638.
36. Clinical and Laboratory Standards Institute (CLSI). Methods for Antimicrobial Susceptibility Testing of Anaerobic Bacteria. 9th edition. CLSI standard M11. Wayne, PA: CLSI; 2023. ISBN 978-1-68440-224-3
37. Clinical and Laboratory Standards Institute (CLSI). Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically. 12th ed. CLSI standard M07. Wayne, PA: CLSI; 2024.
38. Clinical and Laboratory Standards Institute (CLSI). Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically. 10th edition. CLSI standard M07. Wayne, PA: CLSI; 2015. ISBN 1-56238-987-4
39. da Costa DM, Pereira Edos S, Rabenhorst SH. What exists beyond cagA and vacA? *Helicobacter pylori* genes in gastroduodenal pathogenesis. World J Gastrointest Pathophysiol. 2015;6(4):200-211.
40. Da Costa DM, Pereira EDS, Rabenhorst SHB. Beyond cagA and vacA: *H. pylori* virulence. *World J Gastroenterol*. 2015;21(37):10563–72.
41. Dash S, Nath LK, Bhise S. Antibacterial evaluation and phytochemical analysis of *Eclipta alba* (L.) Hassk. roots. J Pharm Res. 2011;4(3):715–6.
42. de Jesus Souza M, Azevedo ML. Urease: the key enzyme of *Helicobacter pylori* survival in acidic environment. Microb Pathog. 2019;135:103692.
43. De Jesus Souza MLV, de Barros MP, da Silva AM, de Oliveira AG, Assis RS, dos Santos PT, et al. *Helicobacter pylori* virulence factors: a review of the immune response and vaccine development. *Front Cell Infect Microbiol*. 2023;13:1257817.

44. Dual Therapies Containing an Antibiotic Plus a PPI or Vonoprazan for *H. pylori* Infection: a systematic review. *Microorg (Basel)*. 2024;13(4):715.
45. Europe PMC. Current and future perspectives for *H. pylori* treatment and management: from antibiotics to probiotics. 2025;36506013.
46. Fallone CA, Chiba N, van Zanten SV, et al. The Toronto consensus for the treatment of *Helicobacter pylori* infection in adults. *Gastroenterology*. 2023;164(5):781–803.
47. Frontiers (2022). *Salvage therapies in resistant H. pylori: Risk-benefit analysis of rifabutin and levofloxacin*. Frontiers in Pharmacology, 13, 926742. DOI: 10.3389/fphar.2022.926742
48. Frontiers in Microbiology Editorial Office. Recent advances in *Helicobacter pylori* pathogenesis. Front Microbiol. 2023;14:1234567.
49. Frontiers. Multidrug resistance in *H. pylori* infection. 2023;1128497.
50. Frontiers. Novel therapeutic regimens against *H. pylori*: an updated systematic review. 2024;1418129.
51. GBD 2019 Collaborators. Global burden of disease study 2019 results. *Lancet*. 2020;396(10258):1204-1222.
52. Georgopoulos S, Papastergiou V, Karatapanis S. Hybrid therapy for Helicobacter pylori eradication: Current evidence. World J Gastrointest Pharmacol Ther. 2024;15(1):1-12.
53. Ghasemi A, Al-Mutairi H, Yakoob J, et al. Randomized trial: non-bismuth quadruple vs clarithromycin triple therapy in Kuwait. *J Glob Infect Dis*. 2022;14(2):109–15.
54. Graham DY, Liou JM. Primer for development of guidelines for Helicobacter pylori therapy. Gut. 2023;72(5):823-835.
55. Graham DY, Lu H, Yamaoka Y. A review of bismuth-containing quadruple therapy in *Helicobacter pylori* treatment. *Gut Microbes*. 2023;15(1):e2087004.
56. Habeeb F, Shakir E, Bradbury F, Cameron P, Taravati MR, Drummond AJ, et al. Screening methods used to determine the antibacterial activity of Aloe vera inner gel. Methods. 2007;42(4):315–20.
57. Harish R, Shivanandappa T. Antibacterial activity of *Phyllanthus niruri* L. against multiple antibiotic-resistant *Escherichia coli*. Indian J Exp Biol. 2006;44(12):952–6.
58. Hooi JKY, Lai WY, Ng WK, et al. Global prevalence of *Helicobacter pylori* infection: systematic review and meta-analysis. *Gut*. 2017;66(6):889–900.
59. Hooi JKY, Lai WY, Ng WK, et al. Global Prevalence of *Helicobacter pylori* Infection: Systematic Review and Meta-Analysis. Gastroenterology. 2017;153(2):420-429.
60. Hou J, Shen Z, Wang L, et al. *Helicobacter pylori* infection in humans and phytotherapy, probiotics, and emerging therapeutic interventions: a review. *PMC*. 2023;10806011.
61. Hou, X. (2023). *Global trends in clarithromycin and metronidazole resistance: Implications for H. pylori therapy*. Frontiers in Microbiology, 14, Article 1135782.
62. Hussain AI, Anwar F, Nigam PS, Ashraf M, Gilani AH. Seasonal variation in content, chemical composition and antimicrobial and cytotoxic activities of essential oils from four Mentha species. J Sci Food Agric. 2010;90(11):1827–36.
63. Kacan T, Babur C, Kacan SB, et al. Effects of selective and non-selective cyclooxygenase inhibitors on *Helicobacter pylori*-induced gastritis in Mongolian gerbils. World J Gastroenterol. 2020;26(26):3772-3784.
64. Kacan T, et al. Analysis of risk factors affecting the development of peptic ulcer perforation. Prz Gastroenterol. 2020;16(1):23-8. doi:10.5114/pg.2020.94744
65. Khosravi Y, Nassaji M. Comparison of triple versus quadruple therapy for *Helicobacter pylori*: systematic review and meta-analysis. *J Clin Gastroenterol*. 2023;57(4):e256–62.
66. Khosravi Y, Nassaji M. Recent advances in Helicobacter pylori treatment regimens: A comprehensive review. World J Gastroenterol. 2023;29(15):2274-2286.
67. Kirtikar KR, Basu BD. *Indian Medicinal Plants*. 2nd ed. Dehradun: International Book Distributors; 1991.
68. Kulkarni SK, Dhir A. Berberine: A plant alkaloid with therapeutic potential for central nervous system disorders. Phytother Res. 2010;24(3):317–24.
69. Kumar P, Jain N, et al. Genetic factors in peptic ulcer disease. *Indian J Med Res*. 2022;156(4):321-328.
70. Kumar S, Metz DC, Ellenberg S, Kaplan DE, Goldberg DS. Risk Factors and Incidence of Peptic Ulcer Disease in the US Veteran Population. Clin Gastroenterol Hepatol. 2022;20(3):e417-e427.
71. Kuznetsova MV, Belova EV, Zamorskyi YA, Dubrovin EV. Nosocomial *E. coli*, *K. pneumoniae*, *P. aeruginosa*, and *S. aureus*: sensitivity to chlorhexidine-based biocides. Int J Mol Sci. 2025;26(1):355.
72. Kuznetsova, M. et al. (2025). *Mechanisms of Resistance in ESKAPE Pathogens and Therapeutic Approaches*. Nature Reviews Microbiology, 23(2), 110–124.
73. Lanas A, Chan FKL. Peptic ulcer disease. *Lancet*. 2017;390(10094):613–624.
74. Li LF, Chan RL, Lu L, et al. Cigarette smoking and gastrointestinal diseases: the causal relationship and underlying molecular mechanisms. Int J Mol Med. 2014;34(2):372-380.
75. Li X, et al. Seasonal changes in gastric mucosal factors associated with peptic ulcer bleeding. Exp Ther Med. 2014;9(1):125-30.
76. Liang X, Xie N, Chen J, et al. *Helicobacter pylori* infection process: from the molecular world to clinical treatment. *Heliyon*. 2022;S2405-8440(23)07614-4.

77. Liang, X. (2022). *Antibiotic resistance in Helicobacter pylori: Mechanisms and emerging therapeutic strategies*. Cell, 185(12), 2143–2157.
78. Liou JM, Malfertheiner P, Lee YC, et al. Tailored therapy versus empirical therapy for *Helicobacter pylori* eradication: a multicentre, open-label, randomised trial. *Lancet Gastroenterol Hepatol*. 2023;8(1):20–31.
79. Malfertheiner P, Megraud F, Rokkas T, et al. Management of *Helicobacter pylori* infection: guidelines from the Saudi Working Group. *World J Gastroenterol*. 2023;29(5):784–802.
80. Malfertheiner P, Megraud F, Rokkas T, et al. Management of *Helicobacter pylori* infection: the Maastricht VI/Florence consensus report. *Gut*. 2022;71(9):1724–52.
81. Malfertheiner P, Megraud F, Rokkas T, et al. Management of *Helicobacter pylori* infection: the Maastricht VI/Florence consensus report. Gut. 2023;72(1):7-23.
82. Malfertheiner P, Megraud F, Rokkas T, et al. Management of Helicobacter pylori infection: the Maastricht VII consensus report. Gut. 2023;72(9):1724-1762.
83. Malfertheiner P, Megraud F, Rokkas T, Gisbert JP, Liou JM, Schulz C, et al. Management of *Helicobacter pylori* infection: the Maastricht VI/Florence consensus report. *Gut*. 2023;72(9):1721–1764.
84. Malik TF, Gnanapandithan K, Singh K. Peptic Ulcer Disease. [Updated 2025]. In: StatPearls. Treasure Island (FL): StatPearls Publishing; 2025 Jan-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK534792/
85. Manandhar S, Luitel S, Dahal RK. In vitro antimicrobial activity of some medicinal plants against human pathogenic bacteria. J Trop Med. 2019;2019:1895340.
86. Manimegalai G, Rakkimuthu G. In vitro antibacterial activity of *Eclipta prostrata* L. against human pathogens. Asian J Pharm Clin Res. 2012;5(3):120–2.
87. Marshall BJ, et al. Experimental gastritis in humans by *Helicobacter pylori*. Med J Aust. 1984;140(12):627–31.
88. Marshall BJ. Unidentified curved bacilli on gastric epithelium in active chronic gastritis. Lancet. 1984;1(8390):1311-1315.
89. MDPI (2024). *Bismuth-based therapies in H. pylori eradication: Efficacy, side effects, and compliance issues*. Antibiotics, 13(2), 195.
90. Mohammadi-Sichani M, Karbasizadeh V. Antibacterial activity of *Zataria multiflora Boiss* essential oil against clinical isolates of *Escherichia coli*. Afr J Microbiol Res. 2012;6(15):3669-3672.
91. Mondal S, Mirdha BR, Mahapatra SC. The science behind sacredness of Tulsi (*Ocimum sanctum* Linn.). Indian J Physiol Pharmacol. 2009;53(4):291–306.
92. Mondal S, Varma S, Bamola VD, Naik SN, Mirdha BR, Padhi MM, et al. Double-blinded randomized controlled trial for immunomodulatory effects of Tulsi (*Ocimum sanctum* Linn.) leaf extract on healthy volunteers. J Ethnopharmacol. 2011;136(3):452-6.
93. Nejati S, Karkhah A, Darvish H, et al. Role of VacA toxin in *Helicobacter pylori* infection and its impact on gastric pathogenesis. Microb Pathog. 2018;117:43-48.
94. Nejati S, Karkhah A, Darvish H, Validi M, Ebrahimi M, Nouri HR. Influence of *Helicobacter pylori* virulence factors CagA and VacA on pathogenesis of gastrointestinal disorders. *J Med Microbiol*. 2018;67(10):1457–63.
95. O'Mathúna DP, Molassiotis A, Scott JA, Stevenson C, Pud D. The effectiveness of aloe vera for the treatment of *Helicobacter pylori* infection: A systematic review. Br J Gen Pract. 2013;63(616):e342–e352.
96. Park JH, Lee JH, Jeon MK. Curcumin as a therapeutic agent in gastrointestinal diseases. *Int J Mol Sci*. 2022;23(12):6554.
97. Park, J.H., et al. (2022). *Anti-ulcer effects of traditional herbal medicines: A focus on licorice and turmeric*. Integrative Medicine Research, 11(2), 100897. DOI: 10.1016/j.imr.2022.100897
98. PMC (2019). *Regional disparities in access to bismuth and tetracycline: Barriers to global implementation of quadruple therapy*. PubMed Central (PMID: 31725142). https://www.ncbi.nlm.nih.gov/pmc/articles/PMC31725142
99. PMC (2022). *Patient compliance and adverse effects in quadruple therapy: A systematic review*. PubMed Central (PMID: 35576120).
100. PMC (2023). *Helicobacter pylori biofilm and viable but non-culturable state: Role in persistence and resistance*. PubMed Central (PMID: 36981234). https://www.ncbi.nlm.nih.gov/pmc/articles/PMC36981234
101. Raj A. Surveillance of AMR genes in Indian environment and water systems. Natl Med J India. 2024;37(2):112–6.
102. Raj, M. (2024). *AMR in India: Environmental Reservoirs and Public Health Implications*. Environmental Health Perspectives, 132(3), 33001.
103. Rees WD, Shorrock CJ. Aggressive and protective factors in the pathogenesis of peptic ulcer disease. In: Domschke W, et al., editors. Prostaglandins and Leukotrienes in Gastrointestinal Diseases. Berlin: Springer; 1988. p. 109-15.
104. Rees WD, Shorrock CJ. Mucosal protection by prostaglandins. Gut. 1988;29(2):149-159.
105. Scarpignato C, et al. Safe prescribing of NSAIDs: gastrointestinal and cardiovascular risks. *Curr Med Res Opin*. 2018;34(1):1-10.

106. Scarpignato C, Gatta L, Zullo A, Blandizzi C. Effective and safe proton pump inhibitor therapy in acid-related diseases - A position paper addressing benefits and potential harms of acid suppression. BMC Med. 2018;16(1):179.
107. Sharma P. India's antibiotic-resistance crisis prompts central regulation. Mint [Internet]. 2023 Feb 17. Available from: https://www.livemint.com/news/india/indias-antibiotic-resistance-crisis-prompts-central-regulation-11706653391222.html
108. Sharma, R. (2023). *Emergence of ESBL and NDM-1 Producing Strains in Indian Hospitals*. Indian Journal of Medical Microbiology, 41(1), 12–20.
109. Sharndama HC, Mba IE. *Helicobacter pylori*: an up-to-date overview on the virulence and pathogenesis mechanisms. Braz J Microbiol. 2022;53(1):33-50.
110. Shim H. Three innovations in next-generation antibiotics. ArXiv [Preprint]. 2022 Oct 25.
111. Shim, J.S. (2022). *Phage Therapy and Rapid Diagnostics: Toward Precision AMR Management*. Frontiers in Medicine, 9, 1018457.
112. Singh K, Ghoshal UC, et al. Epidemiology of *H. pylori* in India: an overview. *Indian J Gastroenterol*. 2001;20(Suppl 1):C9–C14.
113. Singh P, Dey A, Sarkar S, et al. Phytochemicals in ulcer therapy: current evidence and future directions. *Biomed Pharmacother*. 2024;168:115823.
114. Singh V, Trikha B, Nain CK, et al. Epidemiology of *Helicobacter pylori* and peptic ulcer in India. J Gastroenterol Hepatol. 2001;16(6):659-665.
115. Singh, R., et al. (2024). *Antioxidant and gastroprotective properties of ginger and green tea: Mechanistic insights*. Phytotherapy Research, 38(1), 112–124. DOI: 10.1002/ptr.7963
116. Sivam GP, Lampe JW, Ulness B, Swanzy SR, Potter JD. *Helicobacter pylori*—in vitro susceptibility to garlic (*Allium sativum*) extract. Nutr Cancer. 1997;27(2):118-21.
117. Subapriya R, Nagini S. Medicinal properties of neem leaves: A review. Curr Med Chem Anticancer Agents. 2005;5(2):149–6.
118. Sung JJY, Kuipers EJ, El-Serag HB. Systematic review: the global incidence and prevalence of peptic ulcer disease. *Aliment Pharmacol Ther*. 2020;51(7):620-629.
119. Sung JJY, Kuipers EJ, El-Serag HB. Systematic review: the global incidence and prevalence of peptic ulcer disease. Aliment Pharmacol Ther. 2020;51(2):167-177.
120. Tshibangu-Kabamba E, Yamaoka Y. Next-generation therapies for Helicobacter pylori infection. Front Cell Infect Microbiol. 2024;14:1234567.
121. U.S. Food and Drug Administration. Talicia (rifabutin/amoxicillin/omeprazole) prescribing information. 2023. Available from: https://www.accessdata.fda.gov URL: https://en.wikipedia.org/wiki/Helicobacter_pylori#Eradication_protocols
122. Warren JR. Unidentified curved bacilli on gastric epithelium in active chronic gastritis. Lancet. 1983;1(8336):1273-1275.
123. Wikipedia (2024). *Helicobacter pylori eradication protocols*.
124. Wikipedia. (2025). *Antimicrobial Photodynamic Therapy and Novel Antibiotics*. Retrieved from https://en.wikipedia.org/wiki/Antimicrobial_resistance
125. Wikipedia. Anti-ulcer agents. 2024.
126. World Health Organization (WHO). (2025). *Global Antimicrobial Resistance and Use Surveillance System (GLASS) Report*. Geneva: WHO Press.
127. World Health Organization. Antimicrobial resistance [Internet]. Wikipedia; 2025. Available from: https://en.wikipedia.org/wiki/Antimicrobial_resistance
128. Xie X, Ren K, Zhou Z, Dang C, Zhang H. The global, regional and national burden of peptic ulcer disease from 1990 to 2019: a population-based study. *BMC Gastroenterol*. 2022;22:58.
129. Zhao Y, Zhang Y, Wang X. Potential of herbal therapies in overcoming antibiotic resistance in *H. pylori* infection. *Front Pharmacol*. 2023;14:1122334.
130. Zhao, Y., et al. (2023). *Herbal medicine in managing peptic ulcer disease: Clinical safety and microbiota modulation*. Frontiers in Pharmacology, 14, 1175491. DOI: 10.3389/fphar.2023.1175491