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*Original article*

**The efficacy of culture-guided *versus* empirical therapy with high-dose proton pump inhibitor as third-line treatment of *Helicobacter pylori* infection — A real-world clinical experience**

**Abbreviations:** ACG, American College of Gastroenterology; CI, confidence interval; ITT, Intention-to-treat; *H. pylori*, *Helicobacter pylori*; homEM, homogeneous extensive metabolizer; MALT, mucosa-associated lymphoid tissue; MIC, minimal inhibitory concentrations; NBQT, non-bismuth-based quadruple therapy; NA, not applicable; PP, per-protocol; PPIs, proton pump inhibitors

**Abstract**

**Background:** Most consensuses recommend culture-guided therapy as third-line *H. pylori* treatment. This study aimed to investigate the efficacies of culture-guided therapy and empirical therapy with high-dose proton pump inhibitor (PPI) in the *H. pylori* third-line treatment.

**Materials and Methods:** Between August 2012 and October 2021, *H. pylori*-infected patients with at least two failed eradication attempts received anti-*H. pylori* therapy according to the results of antimicrobial sensitivity tests plus high-dose rabeprazole and/or bismuth. They were categorized into three groups: (1) patients who had positive results of culture with equal to or more than three susceptible antibiotics were treated by culture-guided non-bismuth quadruple therapy, (2) patients who had positive results

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of culture with one or two susceptible antibiotics were treated by culture-guided bismuth-containing therapy, and (3) patients who had a negative result of culture were treated by an empirical therapy with high-dose rabeprazole plus amoxicillin, tetracycline and levofloxacin. A post-treatment assessment was conducted at week 8.

**Results:** We recruited 126 patients. The eradication rates of culture-guided non-bismuth quadruple therapy (*n*=50), culture-guided bismuth-containing therapy (*n*=46) and empirical therapy (*n*=30) were 84.0%, 87.0% and 66.7% (95% confidence interval: 73.8%-94.2%, 77.3%-96.7% and 49.8%-83.6%), respectively. Overall, culture-guided therapy achieved a higher eradication rate than empirical therapy (85.4% *vs.* 66.7%; 95% confidence interval, 0.4% to 37.0%, $P = 0.022$).

**Conclusions:** Culture-guided therapy with high-dose PPI achieves a higher eradication rate than empirical therapy with high-dose PPI in the third-line treatment of *H. pylori* infection. The eradication rate of rescue therapy with bismuth plus two susceptible antibiotics is not inferior to that with three susceptible antibiotics.

**Keywords:** antibiotic resistance; *Helicobacter pylori*; high-dose proton pump inhibitor; susceptibility-guided therapy

## INTRODUCTION

*Helicobacter pylori* (*H. pylori*), one of the most prevalent pathogens in the world, is an important pathogen that colonizes the human gastric mucosa and affects more than half of the world's population.[1] Although most infected individuals will be asymptomatic throughout their lives, *H. pylori* infection confers a 1–10% risk of developing gastric or duodenal ulcers, 0.1–3% risk of developing gastric adenocarcinoma, and < 0.01% of developing mucosa-associated lymphoid tissue (MALT) lymphoma.[2]

Nevertheless, eradicating *H. pylori* has become a rising challenge for physicians worldwide owing to the growing prevalence of global antibiotic resistance.[3] The practice guidelines, including the Maastricht V/Florence Consensus and the American College of Gastroenterology (ACG), recommend that patients who suffer from multiple treatment failures should undergo *H. pylori* culture and antimicrobial susceptibility testing to individualize the following course of treatment.[4,5] The Taiwan Consensus also suggests *H. pylori* culture with susceptibility testing or molecular determination of genotype resistance as third-line rescue therapy.[6] However, there is limited evidence regarding the effectiveness of culture-guided therapy for rescue therapy. Furthermore, most hospitals do not have facilities to perform *H. pylori* cultures and antimicrobial susceptibility tests, and/or the high cost of endoscopically obtaining gastric biopsy specimens for *H. pylori* cultures restricts the clinical application of culture-guided therapy in many countries. Therefore, empirical rescue treatments were chosen as alternative therapies in some clinical conditions.

Previous studies have demonstrated high rates of resistance to clarithromycin and

metronidazole in patients who have undergone *H. pylori* eradication therapy.[7-9] Several studies have revealed that tetracycline, amoxicillin, and levofloxacin are good candidates for antibiotics used in the rescue treatment of *H. pylori* infection.[7-11] Because bismuth is not available in many hospitals, non-bismuth-based quadruple therapy (NBQT) is often used to salvage treatment of *H. pylori* infection.[12] However, variable eradication rates of NBQT ranging from 72% to 90% for rescue treatment have been reported in different studies.[9,12,13]

Proton pump inhibitors (PPIs) play an important role in *H. pylori* treatment. They possess anti-*H. pylori* activity and increase the bioavailability and activity of some antibiotics by reducing gastric acid secretion.[14] The minimal inhibitory concentrations (MIC) of most antibiotics against *H. pylori* are dependent on the pH of the environment. Therefore, PPIs are an important component of anti-*H. pylori* regimen because they increase the pH of the stomach, allowing better antimicrobial activity.[15,16] They are metabolized by the hepatic cytochrome P450 system, particularly S-mephenytoin 40-hydroxylase (*CYP2C19*). Previous studies have reported that the *CYP2C19* homogeneous extensive metabolizer (homEM) genotype is an independent factor for failure of first-line *H. pylori* eradication therapy.[17,18] High-dose dual therapy consists of high-dose PPI and amoxicillin, which maintain the intragastric pH at a value higher than 6.5 regardless of the *CYP2C19* genotype and maintains a steady plasma concentration of amoxicillin above the MIC for *H. pylori*.[19,20] The efficacy of the new therapy was significantly higher than that of standard triple therapy.[21]

Currently, it is unclear whether culture-guided therapy is superior to empirical therapy as third-line treatment of *H. pylori* infection. A systematic review reported an

unsatisfactory eradication rate of less than 80% with culture-guided therapy as a third-line treatment.[22] Whether the suboptimal eradication of culture-guided therapy as the third-line treatment of *H. pylori* infection is related to the rapid metabolism of PPIs in individuals with *CYP2C19* homEMs remains unknown. Therefore, we designed this study to investigate the efficacy of culture-guided and empirical therapies as the third-line treatment of *H. pylori* infection. To maintain a steady plasma concentration of antibiotics regardless of the *CYP2C19* genotype, high-dose PPI was administered in both rescue therapies.

## MATERIALS AND METHODS

### *Study population*

The study, a prospective, multicenter, open-label clinical trial was conducted at Kaohsiung Medical University Hospital, Kaohsiung Veterans General Hospital, and Tainan Municipal An Nan Hospital in Taiwan. *H. pylori*-infected adult patients of at least 20 years of age and failure of first-line (standard triple, non-bismuth quadruple, and bismuth quadruple therapies) and second-line eradication treatments (bismuth quadruple, fluoroquinolone-containing triple/quadruple therapies) were recruited for this study. Written informed consent was obtained from all the patients prior to enrollment. Subjects with any of the following criteria were excluded from this trial: (a) known allergy to any antibiotic or PPIs in our study or, (b) previous gastric surgery, (c) pregnant or lactating women, and (d) the use of antibiotics within the prior 4 weeks. This trial was approved by the Institutional Review Board of Kaohsiung Medical University Hospital (KMUHIRB-E(II)-20150091), Kaohsiung Veterans General

Hospital (VGHKS13-CT12-17), and Tainan Municipal An Nan Hospital (TMANH110-REC015).

***Procedures***

Before enrollment, the status of *H. pylori* infection was determined by (i) positive results of both rapid urease test and histology, or (ii) positive culture results. Patients with positive results in one of these tests were eligible for enrollment. Complete medical history and demographic data of all patients were acquired, including age, sex, medical history, history of smoking, and alcohol, coffee, and tea consumption, and a standard questionnaire was completed by each patient on enrollment.

All eligible patients underwent esophagogastroduodenoscopy, and two gastric specimens were obtained from the lesser curvature of the antrum and the corpus by endoscopic biopsy.[23] The biopsy specimens were then cultured on Brucella chocolate agar with 7% sheep blood, incubated for 7 days under microaerobic conditions, and the *H. pylori* strains were tested for antibiotic susceptibility using the E-test (AB Biodisk, Solna, Sweden). *H. pylori* strains with MIC > 0.5 μg/mL, > 1 μg/mL, > 1 μg/mL, > 4 μg/mL, and > 8 μg/mL were considered to be resistant to amoxicillin, clarithromycin and levofloxacin, tetracycline, and metronidazole, respectively.[21]

Eligible patients received anti-*H. pylori* therapy based on the results of culture and antimicrobial sensitivity tests. In patients with a positive result of culture and *H. pylori* strains susceptible to three or more antibiotics, a non-bismuth regimen containing a high-dose PPI plus three susceptible antibiotics was used to treat *H. pylori* infection. In those with a positive result of culture and *H. pylori* strains susceptible to only one or two antibiotics, a bismuth-containing regimen containing a high-dose PPI, bismuth and

susceptible antibiotics was applied as rescue treatment. In patients with a negative culture results, an empirical therapy containing a high-dose PPI plus amoxicillin, tetracycline and levofloxacin was administered to treat *H. pylori* infection. Therefore, the treatments were categorized into three groups: (1) 14-day culture-guided non-bismuth quadruple therapy (rabeprazole [Pariet E.C.; Bushu Pharm, Saitama-ken, Japan] 20 mg q.d.s. and three effective antibiotics) for patients with a positive culture result *H. pylori* strains susceptible to at least three antibiotics; (2) 14-day culture-guided bismuth-containing therapy (rabeprazole 20 mg q.d.s., tripotassium dicitrato bismuthate [KCB; Swiss Pharm, Tainan, Taiwan] 300 mg q.d.s., and all the effective antibiotics) for patients with a positive result of culture and *H. pylori* strains susceptible to only one or two antibiotics; and (3) 14-day empirical therapy (rabeprazole 20 mg q.d.s, amoxicillin [amoxicillin trihydrate; Yung Shin Pharm, Taichung, Taiwan] 500 mg q.d.s., tetracycline [tetracycline HCl; Taiwan Veterans Pharm, Chungli, Taiwan] 500 mg q.d.s. and levofloxacin [Cravit; Sanofi-Aventis, Taoyuan, Taiwan] 500 mg o.d.) for patients with a negative result of culture.

The patients were requested to return in the second week to assess drug adherence and adverse effects. Adverse events were assessed by a research assistant according to the defined criteria. The patients were informed of the common side effects of the study drugs and were asked to record these symptoms in the provided diaries. Adverse events were assessed according to a four-point scale: none, mild (discomfort annoying but not interfering with daily life), moderate (discomfort sufficient to interfere with daily life), and severe (discomfort resulting in discontinuation of eradication therapy).[24] Drug adherence was assessed by pill counts after treatment. Adherence was graded as good

if the patients took equal to or more than 80% of the total medication, or as poor otherwise.[24]

Post-treatment *H. pylori* status was determined by $^{13}$C-urea breath tests using an infrared spectrometer at the end of week 8. All patients were asked to stop treatment with PPI and histamine-2 blockers for at least 2 weeks before undergoing esophagogastroduodenoscopy or urea breath tests. $^{13}$C-urea breath tests were performed after an overnight fast using the Proto Pylori kit (Isodiagnostika, Montreal, Canada). The 75 mg $^{13}$C-urea manufactured by the Institute of Nuclear Energy Research in Taiwan, was dissolved in water. Baseline and 30 min breath samples were assayed with an infrared spectrometer that produced computer-generated results at Kaohsiung Medical University Hospital and Kaohsiung Veterans General Hospital. Positive results were defined as a computer-generated $\delta^{13}CO_2$ value $\geq$ 4 units and negative results as < 2.5 units.[10] Successful eradication was defined as a negative result of the $^{13}$C-urea breath test. Patients with intermediate delta values (2.5-4 units) underwent another $^{13}$C-urea breath test at least 4 weeks later until the results became conclusive.

***Statistical analysis***

The primary endpoint of the study was *H. pylori* eradication rate. Intention-to-treat (ITT) and per-protocol (PP) analyses were conducted. The ITT population included all randomized subjects who received at least one dose of the eradication drugs. Patients whose infection status was unknown following treatment were considered to have treatment failures in the ITT analysis. The PP analysis excluded patients with unknown *H. pylori* status following therapy and those with poor drug adherence. The secondary

endpoints were the frequency of adverse events and drug adherence. A chi-square test with or without the Yates correction for continuity and Fisher's exact test were used where appropriate to compare the major outcomes between the groups. Statistical significance was set at $P < 0.05$. SPSS (version 25 for Microsoft Windows) was used for all statistical analyses.

## RESULTS

### *Characteristics of the patients*

Between August 2012 and October 2021, 137 patients with failure of first-and second-line eradication were initially assessed for eligibility, and 126 of them were recruited in our study. Figure 1 demonstrates the patient composition. Ninety-six patients with the positive culture results underwent culture-guided non-bismuth quadruple therapy ($n = 50$) or culture-guided bismuth-containing therapy ($n = 46$). The other patients ($n = 30$) with negative culture results were treated with empirical therapy.

The clinical characteristics of the patients administered culture-guided or empirical therapy are summarized in Table 1. The previous failed eradication regimens of all patients were listed in Supplemental Table 1. The two patient groups had comparable characteristics in terms of age, sex, and history of peptic ulcer disease, and alcohol, coffee, and tea consumption. The resistance rates for clarithromycin, metronidazole, amoxicillin, tetracycline, and levofloxacin in culture-guided therapy group were 79.2%, 65.6%, 0%, 0%, and 81.3%, respectively.

### *Eradication rate, drug adherence and adverse events of culture-guided and empiric*

***therapies in the third-line treatment of H. pylori infection***

Table 2 illustrates the major outcomes of culture-guided and empiric therapies in the third-line treatment of *H. pylori* infection. The eradication rates of culture-guided therapy with high-dose rabeprazole and empirical therapy with high-dose rabeprazole were 85.4% (95% confidence interval [CI]: 80.4%-92.6%) and 66.7% (95% CI: 49.8%-83.6%), respectively. Overall, culture-guided therapy achieved a higher eradication rate than empirical therapy (Difference, 18.7%; 95% CI: 0.4% to 37.0%, $P$ = 0.022). PP analysis yielded similar results (88.2% *vs.* 68.0%; $P$ = 0.015). All patients were included in the ITT analysis for adverse events and adherence. Patients administered culture-guided therapy had better drug adherence than those administered empiric therapy (95.8% *vs.* 83.3%, respectively; $P$ = 0.035). The two therapies exhibited similar frequencies of overall adverse events (31.3% *vs.* 43.3%; $P$ = 0.122). Table 3 lists the profiles of adverse events of the two eradication treatments. The empirical therapy group had a higher frequency of nausea than the culture-guided therapy group (40.0% *vs.* 17.7%; $P$ = 0.011). No significant differences were observed between the groups in terms of the frequencies of other adverse events.

***Intra-group analysis for eradication rate, drug adherence and adverse events in culture-guided therapy patients***

Table 2 also displays the intra-group comparison of the major outcomes of patients administered NBQT or bismuth-containing therapy in the culture-guided therapy group. The detail eradication rates of each specific therapy of NBQT and bismuth-containing therapies were listed in the Table 4 and Table 5, respectively. The most commonly

prescribed antibiotics combination in NBQT was amoxicillin plus tetracycline and metronidazole (31/50), and that in bismuth-containing therapies was amoxicillin plus tetracycline (35/46). Overall, NBQT subgroup and bismuth-containing subgroup had comparable eradication rates in ITT (84.0% *vs.* 87.0%; $P = 0.682$) and PP analysis (87.5% *vs.* 88.9%). However, the NBQT subgroup had a higher frequency of adverse events than the bismuth-containing subgroup (42.0% *vs.* 19.6%; $P = 0.018$). There were no significant differences in drug adherence between the two patient groups (96.0% *vs.* 97.8%; $P > 0.999$).

## DISCUSSION

In the current study, we conducted a prospective trial to investigate the efficacy of culture-guided and empirical therapy with high-dose PPI as the third-line treatment of *H. pylori* infection. The results demonstrate several novel findings. First, culture-guided therapy with high-dose PPI achieved a higher eradication rate than empirical therapy with high-dose PPI as the third-line treatment of *H. pylori* infection. Second, rescue therapy with bismuth plus two or three antibiotics, to which *H. pylori* was sensitive, had comparable eradication rates in the treatment of refractory *H. pylori* infection, but the bismuth with two antibiotics exhibited a lower frequency of adverse events than the bismuth with three antibiotics (19.6% *vs.* 42.0%; $P = 0.018$). Third, a significant number (15%) of infected subjects failed to eradicate *H. pylori* infection, although the antibiotics in their rescue regimens were effective *in vitro*, and high-dose PPI was administered to increase antimicrobial activity.

Currently, most national and international consensus recommend culture-guided

therapy as third-line treatment of *H. pylori* infection.[4-6] However, whether culture-guided therapy is superior to empirical therapy as third-line treatment of *H. pylori* infection remains unclear. Yu et al. conducted a prospective trial of 200 patients and demonstrated that culture-guided therapy could prove highly efficacious (overall eradication rate: 94.5%) in multidrug-resistant *H. pylori* infection.[25] However, a systematic review and meta-analysis showed that the cure rates of susceptibility-based treatment as a third-line therapy were not superior to empirical therapies, and the unsatisfactory mean cure rate of susceptibility-based treatment was reported to be 72%.[26] Possible explanations for the discrepancies in the efficacy of culture-guided therapy included divergent patient compliance, different treatment durations, and variable frequencies of *CYP2C19* genotypes.[26,27]

The MIC of most antibiotics against *H. pylori* are dependent on the pH of the environment. PPIs possess anti-*H. pylori* activity and can increase the bioavailability and activity of antibiotics by reducing gastric acid secretion. In this study, we applied high-dose rabeprazole to maintain the intragastric pH at a value higher than 6.5, regardless of the *CYP2C19* genotype. The data of this prospective trial showed that culture-guided therapy with high-dose PPI had a markedly higher eradication rate than empirical therapy with high-dose PPI, in both ITT (85.4% *vs.* 66.7%) and PP analysis (88.2% *vs.* 68.0%). Additionally, the patients who underwent culture-guided therapy had better drug adherence than those who underwent empirical therapy (95.8% *vs.* 83.3%; $P = 0.035$). These novel findings in this study support the recommendations made, concerning the third-line treatment of *H. pylori* infection, by most national and international consensus guidelines.[4-6]

In this study, the patient groups receiving regimens containing three antibiotics (the empirical therapy group and the non-bismuth subgroup in the culture-guided group) had higher frequencies of adverse effects than the patient group receiving regimens containing only two antibiotics (the bismuth containing subgroup) (43.3% & 42.0% *vs.* 19.6%). The drug adherence in the culture-guided group was higher than that in the empirical group, probably due to the fewer adverse events of the bismuth containing subgroup.

In the current study, the frequencies of antibiotic resistance to clarithromycin, metronidazole, amoxicillin, tetracycline, and levofloxacin in *H. pylori* strains isolated from patients with failure of at least two anti-*H. pylori* therapies were 79.2%, 65.6%, 0%, 0%, and 81.3%, respectively. The results were similar to the antibiotic resistant data of a multicentre study from Taiwan.[28] In that study, the resistant rates of *H. pylori* to amoxicillin, levofloxacin and tetracycline in the third-line treatment from 2017 to 2019 were 3%, 84% and 1%, respectively. Another randomized controlled trial from Taiwan showed that the resistant rates of *H. pylori* to amoxicillin, levofloxacin and tetracycline in the third-line treatment were 14%, 59% and 6%, respectively.[29] In general, the resistant frequencies of amoxicillin and tetracycline of *H. pylori* strains in third-line eradication therapy are less than 15% in Taiwan, and the resistant frequency of levofloxacin are higher than 50%.

In this study, only 50 (52.1%) of the 96 *H. pylori* strains with culture data had equal to or more than three susceptible antibiotics. Because bismuth salts have a synergistic effect with antibiotics by destroying bacteria like an antiseptic,[30,31] we applied tripotassium dicitrato bismuthate to increase the effects of antibiotics in patients who

had only two susceptible antibiotics. The results demonstrated that the eradication rate of rescue therapy with bismuth plus two antibiotics was not inferior to that with three antibiotics to which *H. pylori* was susceptible (87.0% *vs.* 84.0%). Additionally, rescue therapy with bismuth plus two antibiotics, to which *H. pylori* was susceptible, had a lower frequency of adverse events than rescue therapy with three antibiotics as the third-line treatment of *H. pylori* infection (19.6% *vs.* 42.0%). The data indicate that a salvage regimen consisting of a PPI, bismuth salt, and two antibiotics to which *H. pylori* is susceptible is a good choice for the treatment of refractory *H. pylori* infection.

This study has some limitations. First, the current study was not a double-blind placebo-controlled trial in which selection bias would be minimized. Patients who underwent culture-guided therapy and empirical therapy might have different antibiotic susceptibility profiles. However, this work provided real-world data on culture-guided and empirical therapies as the third-line treatment of *H. pylori* infection. Second, the current study was conducted in a single country, so the results need to be validated in different countries where there are distinct patterns of antibiotic resistance and *CYP2C19* genotypic polymorphism. Third, a significant number (15%) of infected subjects failed to eradicate *H. pylori* infection, although the antibiotics in their rescue regimens were effective *in vitro*, and high-dose PPI was administered to increase antimicrobial activity. The causes of eradication failure in patients administered antibiotics to which *H. pylori* is susceptible remain to be identified. Fourth, the culture-guided group and empirical therapy group might have different antibiotic susceptibility profiles. However, antibiotic resistance data are unavailable sometimes in clinical practice if patients refuse to receive endoscopy for culture or the result of *H. pylori*

culture is negative. The current study clearly show that the eradication rate of an empirical therapy with a PPI, amoxicillin, tetracycline and levofloxacin in patients with refractory *H. pylori* infection and unavailable antibiotic resistance data was lower than that of culture-guided therapy in patients with refractory *H. pylori* infection and available antibiotic resistance data. It is therefore important to for physicians to get antibiotic susceptibility data in the third-line therapy for *H. pylori* infection. Fifth, it was underpowered to make an adequate analysis to investigate the association between eradication outcome and previous eradication regimens in each treatment arm because the recruited patients in this study received more than 10 kinds of treatment combinations of first-line and second-line therapies (Supplementary Table 1).

In conclusion, culture-guided therapy with high-dose PPI achieved a higher eradication rate than empirical therapy with high-dose PPI as the third-line treatment of *H. pylori* infection. The eradication rate of rescue therapy with bismuth plus two antibiotics to which *H. pylori* was susceptible was not inferior to that with three antibiotics. Additionally, rescue therapy with bismuth plus two antibiotics exhibited a lower frequency of adverse events than rescue therapy with bismuth plus three antibiotics.

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**Table 1. Demographic data of patients**

| Characteristics | Culture-guided group | | | Empiric group | *P* value* |
|---|---|---|---|---|---|
| | Non-bismuth regimen (*n* = 50) | Bismuth-containing regimen (*n* = 46) | All (*n* = 96) | (*n* = 30) | |
| Age (years) (mean ± SD) | 54.3 ± 10.6 | 53.3 ± 9.1 | 53.8 ± 9.8 | 57.5 ± 10.8 | 0.086 |
| Gender (male/female) | 26/24 | 12/34 | 38/58 | 13/17 | 0.715 |
| Smoking | 8 (16.0%) | 6 (13.0%) | 14 (14.6%) | 0 (0.0%) | 0.022 |
| Alcohol consumption | 3 (6.0%) | 2 (4.3%) | 5 (5.2%) | 0 (0.0%) | 0.337 |
| Ingestion of coffee | 24 (48.0%) | 13 (28.3%) | 37 (38.5%) | 7 (23.3%) | 0.127 |
| Ingestion of tea | 16 (32.0%) | 12 (26.1) | 28 (29.2%) | 6 (20.0%) | 0.323 |
| Peptic ulcer disease | 22 (44.0%) | 18 (39.1%) | 40 (41.7%) | 10 (33.3%) | 0.415 |
| Antibiotic resistance | | | | | |
| Clarithromycin | 33 (66.0%) | 43 (93.5) | 76 (79.2%) | - | - |
| Metronidazole | 18 (36.0%) | 45 (97.8%) | 63 (65.6%) | - | - |
| Amoxicillin | 0 (0.0%) | 0 (0.0%) | 0 (0.0%) | - | - |
| Tetracycline | 0 (0.0%) | 0 (0.0%) | 0 (0.0%) | - | - |
| Levofloxacin | 37 (74.0%) | 41 (89.1) | 78 (81.3%) | - | - |

* Comparison between culture-guided therapy and empirical therapy

**Table 2. Outcomes of culture-guided and empirical rescue therapies as the third-line treatment of *H. pylori* infection**

| Outcome | Culture-guided therapy | | | Empiric therapy | *P* value* |
|---|---|---|---|---|---|
| | Non-bismuth regimen (*n* = 50) | Bismuth-containing regimen (*n* = 46) | All (*n* = 96) | (*n* = 30) | |
| Eradication rate | | | | | |
| Intention-to-treat | 84.0% (42/50) (73.8%–94.2%) | 87.0% (40/46) (77.3%–96.7%) | 85.4% (82/96) (80.4%–92.6%) | 66.7% (20/30) (49.8%–83.6%) | 0.022 |
| Per-protocol | 87.5% (42/48) (80.8%–93.8%) | 88.9% (40/45) (82.4%–94.5%) | 88.2% (82/93) (83.5%–94.0%) | 68.0% (17/25) (49.7%–86.3%) | 0.015 |
| Drug adherence | 96.0% (48/50) (92.1%–98.1%) | 97.8% (45/46) (94.7%–98.9%) | 95.8% (92/96) (91.8%–99.8%) | 83.3% (25/30) (70.0%–96.6%) | 0.035 |
| Adverse events | 42.0% (21/50) (28.3%–55.7%) | 19.6% (9/46)† (8.13%–31.1%) | 31.3% (30/96) (17.9%–44.7%) | 43.3% (13/30) (28.8%–64.6%) | 0.122 |

* Comparison between culture-guided therapy and empirical therapy

† Bismuth containing subgroup had a lower frequency of adverse events than non-bismuth subgroup ($P = 0.018$)

**Table 3. Adverse events of culture-guided and empirical rescue therapies as the third-line treatment of *H. pylori* infection**

| | Culture-guided group | | | | |
|---|---|---|---|---|---|
| | Non-bismuth regimen ($n$ = 50) | Bismuth-containing regimen ($n$ = 46) | All ($n$ = 96) | Empiric group ($n$ = 30) | $P$ value* |
| Abdominal pain | 6 (12.0%) | 0 (0.0%) | 6 (6.25%) | 1 (3.33%) | > 0.999 |
| Diarrhea | 4 (8.0%) | 0 (0.0%) | 4 (4.17%) | 1 (3.33%) | > 0.999 |
| Constipation | 1 (2.0%) | 0 (0.0%) | 1 (1.04%) | 0 (0.00%) | > 0.999 |
| Anorexia | 0 (0.0%) | 0 (0.0%) | 0 (0.00%) | 0 (0.00%) | NA |
| Nausea | 12 (24.0%) | 5 (10.9%) | 17 (17.71%) | 12 (40.00%) | 0.011 |
| Vomiting | 3 (6.0%) | 2 (4.3%) | 5 (5.21%) | 2 (6.67%) | 0.671 |
| Skin rash | 0 (0.0%) | 0 (0.0%) | 0 (0.00%) | 0 (0.00%) | NA |
| Headache | 3 (6.0%) | 0 (0.0%) | 3 (3.13%) | 1 (3.33%) | > 0.99 |
| Dizziness | 3 (6.0%) | 2 (4.3%) | 5 (5.21%) | 2 (6.67%) | 0.671 |
| Bad taste | 3 (6.0%) | 0 (0.0%) | 3 (3.13%) | 1 (3.33%) | > 0.99 |
| Fatigue | 3 (6.0%) | 1 (2.2%) | 4 (4.17%) | 3 (10.00%) | 0.356 |

* Comparison between culture-guided therapy and empirical therapy

Abbreviation: NA, not applicable

**Table 4. The eradication rate of each specific therapy in patients receiving non-bismuth quadruple therapies for *H. pylori* infection**

| | Non-bismuth quadruple therapy | | | |
|---|---|---|---|---|
| | RATM ($n$ = 31) | RATL ($n$ = 7) | RACT ($n$ = 9) | RACM ($n$ = 3) |
| Eradication rate | | | | |
| Intention-to-treat | 96.8% (30/31) | 85.7% (6/7) | 55.6% (5/9) | 66.7% (2/3) |
| Per-protocol | 96.7% (29/30) | 85.7% (6/7) | 62.5% (5/8) | 66.7% (2/3) |

Abbreviation: RATM = rabeprazole + amoxicillin + tetracycline + metronidazole, RATL = rabeprazole + amoxicillin + tetracycline + levofloxacin, RACT = rabeprazole + amoxicillin + clarithromycin + tetracycline, RACM = rabeprazole + amoxicillin + clarithromycin + metronidazole

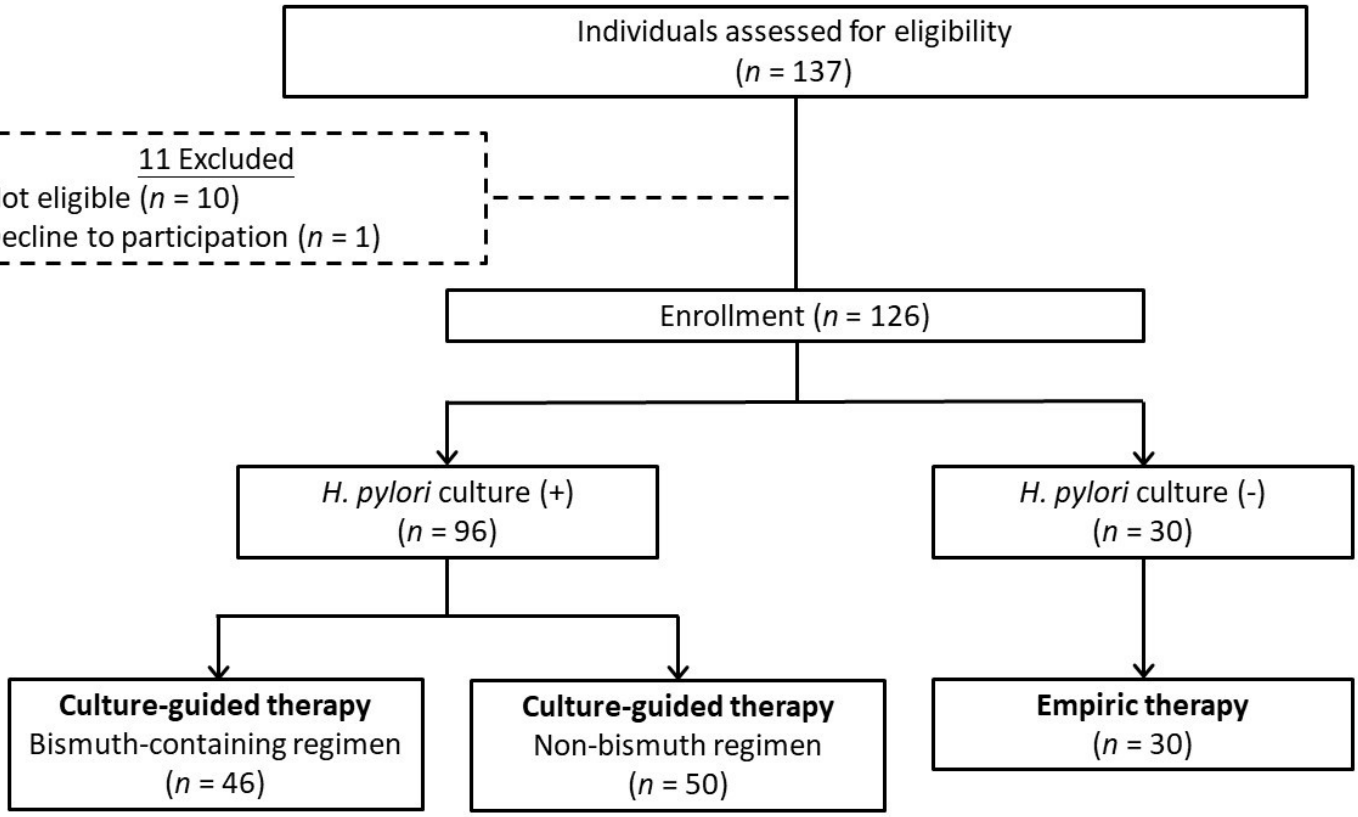
**Table 5. The eradication rate of each specific therapy in patients receiving bismuth-containing third-line treatment for *H. pylori* infection**

| | Bismuth-containing therapies | | | | | |
|---|---|---|---|---|---|---|
| | RBTM ($n$ = 1) | RBTL ($n$ = 4) | RBAT ($n$ = 35) | RBAC ($n$ = 4) | RBAM ($n$ = 1) | RBAL ($n$ = 1) |
| Eradication rate | | | | | | |
| Intention-to-treat | 100% (1/1) | 100% (4/4) | 82.9% (29/35) | 100% (4/4) | 100% (1/1) | 100% (1/1) |
| Per-protocol | 100% (1/1) | 100% (4/4) | 85.3% (29/34) | 100% (4/4) | 100% (1/1) | 100% (1/1) |

Abbreviation: RBTM = rabeprazole + tripotassium dicitrato bismuthate + tetracycline + metronidazole, RBTL = rabeprazole + tripotassium dicitrato bismuthate + tetracycline + levofloxacin, RBAT = rabeprazole + tripotassium dicitrato bismuthate + amoxicillin + tetracycline, RBAC = rabeprazole + tripotassium dicitrato bismuthate + amoxicillin + clarithromycin, RBAM = rabeprazole + tripotassium dicitrato bismuthate + amoxicillin + metronidazole, RBAL = rabeprazole + tripotassium dicitrato bismuthate + amoxicillin + levofloxacin

## FIGURE LEGENDS

**Figure 1. Flow diagram of patient allocation**

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