

Empiric Versus Clarithromycin Resistance–Guided Therapy for *Helicobacter pylori* Based on Polymerase Chain Reaction Results in Patients With Gastric Neoplasms or Gastric Mucosa-Associated Lymphoid Tissue Lymphoma: A Randomized Controlled Trial

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INTRODUCTION: We investigated to compare the effect of empirical therapy vs clarithromycin resistance–guided tailored therapy (tailored therapy) for eradication of *Helicobacter pylori*.

METHODS: In this prospective, single center, open-label randomized controlled trial, we enrolled 72 patients with *H. pylori* infection from January 2019 through June 2019 in Korea. The patients were randomly assigned to both groups received empirical (n = 36) or tailored therapy (n = 36). Empirical therapy was defined as triple therapy with esomeprazole, amoxicillin, and clarithromycin for 10 days irrespective of clarithromycin resistance. Tailored therapy was triple or quadruple therapy with esomeprazole, metronidazole, tetracycline, and bismuth for 10 days based on genotype markers of resistance determined by gastric biopsy. Resistance-associated mutations in 23S rRNA were confirmed by multiplex polymerase chain reaction. Eradication status was assessed by ¹³C-urea breath test, and the primary outcome was eradication rates.

RESULTS: *H. pylori* was eradicated in 27 patients (75.0%), given empirical therapy and 32 patients (88.9%) treated with tailored therapy ($P = 0.136$) in intention-to-treat analysis. In per protocol analysis, the eradication rate was 97.0% and 81.8% in tailored vs empirical groups ($P = 0.046$). Although clarithromycin-resistant *H. pylori* was eradicated in 3/9 (33.3%) with empirical therapy, it was treated in 11/12 (91.7%) with tailored therapy ($P = 0.009$). There was no difference in compliance between 2 groups. The rate of adverse events of the tailored group was higher than that of the empirical group ($P = 0.036$) because quadruple therapy had more side effects than those of triple therapy ($P = 0.001$).

DISCUSSION: Tailored therapy based on polymerase chain reaction is a good alternative to increase eradication rates in a region of high prevalence of clarithromycin resistance (see Visual Abstract, Supplementary Digital Content 1, <http://links.lww.com/CTG/A342>).

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INTRODUCTION

Helicobacter pylori is associated with upper gastrointestinal diseases, including gastritis, peptic ulcer disease, mucosa-associated lymphoid tissue (MALT) lymphoma, and gastric cancer (1). The indications for *H. pylori* treatment in the United States and Korea include peptic ulcer disease, low-grade MALT lymphoma, and the endoscopic resection of early gastric cancer (EGC), with *H.*

pylori infection (2,3). In Japan, reversible early stage of preneoplastic change and *Helicobacter*-related gastritis were added to the indications for *H. pylori* treatment starting in February 2013 (4,5).

In the current Korean guidelines for the treatment of *H. pylori* eradication, published in 2013, triple therapy, including a proton-pump inhibitor (PPI), clarithromycin, and amoxicillin, has been recommended for primary eradication therapy (3). However,

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the Maastricht V guidelines recommended quadruple therapy because the primary treatment in areas with higher than 15% clarithromycin resistance (low level, grade of recommendation; strong), which was applied to the revised *H. pylori* Clinical Practice Guideline Amendment. One recent multicenter, randomized trial and a meta-analysis suggested that 10-day sequential therapy could represent a primary treatment option (6,7). However, the standard triple therapy treatment protocol remains the recommended primary treatment for *H. pylori* eradication in Korea (8).

Antibiotic resistance in *H. pylori*, especially clarithromycin resistance, is a major cause of eradication failure, and the clarithromycin-resistance rate in Korea has recently been reported to have increased to 17.8%, based on bacterial cultures (minimum inhibitory concentration [MIC] of >1 µg/mL) (9). The concordance between bacterial cultures and polymerase chain reaction (PCR)-based susceptibility findings was approximately 95% (10,11). Furthermore, based on the identification of 23S rRNA point mutations, the clarithromycin-resistance rate has increased drastically, with up to 32% of the Korean population presenting clarithromycin-resistance (12).

The reported eradication rate for standard triple therapy in Korea has been increasingly unsatisfactory, as the antibiotic resistance rate of *H. pylori* increases. The ideal eradication rate for *H. pylori* is approximately 90% (13); however, in a recently reported randomized, multicenter study examining primary eradication, the overall eradication rate was 63.9%, in the intention-to-treat (ITT) analysis, and 71.4%, in the per protocol (PP) analysis (6). In addition, one meta-analysis suggested that clarithromycin resistance reduces the efficacy of triple therapy by 66% (95% confidence interval: 58.2–74.2) (14). Another systemic review and meta-analysis reported that clarithromycin-susceptible strains showed a 90.2% eradication rate, whereas clarithromycin-resistant strains demonstrated a 22.2% eradication rate in response to triple therapy in the ITT analysis (15).

To detect bacterial resistance, bacterial *H. pylori* cultures are required for the agar dilution test or E-test. Performing antimicrobial susceptibility testing on *H. pylori* cultures is important for predicting antibiotic treatment outcomes and guiding clinicians in their choice of therapy. However, the cultivation of *H. pylori* to determine the MIC values of antimicrobial substances can be very difficult and time consuming (9,16) because of the characteristics of microaerophilic bacteria. In addition, the culture yields are generally low, and this method is quite costly. Recently, tailored treatments based on clarithromycin susceptibility have been proposed, using the results of a dual-priming oligonucleotide-based multiplex (DPO)-PCR test (17–20). This method uses PCR to verify the presence of mutations, especially A2142G and A2143G, which are 23S ribosomal RNA point mutations known to be highly related to clarithromycin resistance (20). This test can only be performed by gastric biopsy; however, the examination time can be as short as several hours. DPO-PCR to identify *H. pylori* was shown to have sensitivity, specificity, and concordance rates of over 87%, 83%, and 90%, respectively, compared with bacterial cultures or the ¹³C-urea breath tests (¹³C-UBTs) (10,21).

This study aimed to examine the effectiveness *H. pylori* infections treated using clarithromycin resistance-guided tailored therapy (tailored therapy) compared with empirical therapy in a randomized, controlled trial.

METHODS

Trial design and patients

This study was a single center, open-label, parallel, randomized trial. The Institutional Review Board of the Seoul National University Hospital and clinicaltrials.gov approved this study (IRB number: 1811-029-983, clinicaltrials.gov ID: NCT04006340). We performed the trial following the Declaration of Helsinki regarding human experimentation.

Patients with *H. pylori* who were expected to undergo endoscopic resection for gastric neoplasms or diagnosed with gastric MALT lymphoma were eligible. The gastric neoplasms included EGCs or gastric adenomas on endoscopy. Exclusion criteria were as follows: (i) history of gastrectomy, (ii) patients aged younger than 20 years or older than 80 years, (iii) history of *H. pylori* eradication therapies or other antibiotics therapy within a month, (iv) contraindication or previous allergic reaction to the treatment drugs (amoxicillin, clarithromycin, metronidazole, tetracycline, and esomeprazole), and (v) pregnant or lactating women. All patients had written informed consents before enrollment. Demographic data were obtained from interview or medical records, including age, sex, body mass index, comorbidity, history of cigarette smoking or alcohol drinking, and tumor findings; pathological data of gastric neoplasm were achieved from medical records (gross type, Lauren type, differentiation, depth of invasion, vertical location, size, en-bloc resection, and resection margin). Complete resection was defined as an en-bloc resection with tumor-negative margin in specimen after endoscopic resection.

Determination of *H. pylori* infection and clarithromycin resistance

Histologic evaluation was conducted by gastric biopsy specimen from the antrum and body and stained with hematoxylin and eosin and Giemsa stains. *H. pylori* status was assessed by the Sydney system. The rapid urease test (RUT) (CLOtest; Delta West, Bentley, Australia) was also conducted by biopsy samples at gastric antrum. Because the eligible patients in our study had gastric neoplasm or gastric MALT lymphoma, 2 endoscopies were needed at the time of diagnosis and further examination (endoscopic ultrasound) or treatment (endoscopic submucosal dissection). Therefore, we evaluated the status of *H. pylori* infection by both histology and RUT at the first endoscopy, and the clarithromycin resistance by DPO-PCR test at the second endoscopy.

DPO-PCR test (Seeplex ClaR-H. pylori ACE Detection; Seegene Institute of Life Science, Seoul, Korea) was performed on gastric biopsy specimens if histology or RUT was positive. Eligible *H. pylori* status for enrollment was judged as positive if DPO-PCR test was positive. DPO-PCR tests were conducted to identify genetic point mutations according to the manufacturer's recommendations. An initial incubation was performed at 94 °C for 15 minutes, and the next processes allowed 40 amplification cycles in a GeneAmp PCR system 9700 (Applied Biosystems, Foster City, CA) at 94 °C for 30 seconds, 65 °C for 30 seconds, and 72 °C for 1 minute. The last extension was proceeded at 72 °C for 10 minutes.

The amplified DNA products were identified using an ultraviolet transilluminator in electrophoresis. The detection kit including 3 primer pairs with a DPO structure was used for the amplification of the *H. pylori* 23S rDNA (621 bp). A single 621-bp

DNA product was considered belonging to wild-type *H. pylori*. The presence of the A2142G and A2143G mutations resulted in DNA bands at 475-bp and 194-bp, respectively. The kit includes a primer pair for internal control.

Randomization and intervention

Eligible patients were randomly assigned into one of the following treatment groups: (A) empirical therapy or (B) clarithromycin resistance-guided tailored therapy using a computer-generated list that had block randomization with a block size of 4, 6 in a 1:1 ratio (https://www.e-ciencecentral.org/articles/pubreader/SC000026629#_fn_header_idm5766816, openepi.com).

The empirical therapy group (empirical group) received the standard triple regimen and clarithromycin resistance-guided tailored therapy group (tailored group) taken the triple therapy or quadruple therapy according to clarithromycin resistance determined by the DPO-PCR test. Patients were treated with triple therapy including esomeprazole 40 mg, amoxicillin 1 g, and clarithromycin 500 mg twice daily for 10 days, or quadruple therapy containing esomeprazole 40 mg and bismuth 300 mg twice daily, tetracycline 500 mg 4 times daily, and metronidazole 500 mg 3 times daily for 10 days.

Outcome assessment

The eradication status of *H. pylori* was identified by ¹³C-UBT at least 4 weeks after therapy. Before ¹³C-UBT, study patients discontinued histamine 2 blocker or PPI during at least 2 weeks. We asked for all subjects to report any adverse event throughout this trial. The adverse effects and compliance at the completion of treatment were investigated by a standardized interview. Poor compliance was defined as the case where the pill taken below 90%. Patients who did not have ¹³C-UBT after the end of therapy were excluded from the PP analysis. Incremental cost-effectiveness ratios were calculated for the tailored group, and the outcome was measured as the eradication rate. The average costs were defined as the combined costs of endoscopy, RUT, biopsy/histology, medication, ¹³C-UBT, and/or DPO-PCR test. The incremental cost-effectiveness ratio value was defined as the difference between the average total costs for the tailored group and those for the empirical group divided by the average difference in the eradication rate (22):

$$\text{ICER} = \frac{\text{Cost}_{\text{tailored group}} - \text{Cost}_{\text{empirical group}}}{\text{Eradication rate}_{\text{tailored group}} - \text{Eradication rate}_{\text{empirical group}}}$$

Statistical analysis

The eradication rate of clarithromycin resistance-guided tailored group was assumed to be 95.7% based on our pilot study. The calculated sample size was at least 36 in each group to detect a 30% difference to give a statistical power of 80% at a 5% significance level on a 2-sided test with a follow-up loss rate of 10% (Fleiss JL. Statistical Methods for Rates and Proportions: John Wiley & Sons; 1981). All randomized patients were included in the intention-to-treat (ITT) analysis. The primary end point was the eradication rates according to ITT and PP analyses. The secondary end points were the compliance and the adverse events.

Clarithromycin resistance-guided tailored therapy vs empirical therapy groups were compared for demographic and clinicopathological data using the Pearson χ^2 tests, Fisher exact tests, Mann-Whitney *U* tests, Student *t*-tests, and logistic regression model. Pearson χ^2 tests and Fisher exact tests were used for the

analysis of categorical variables, whereas Mann-Whitney *U* tests and Student *t*-tests for the analysis of continuous variables. Factors affecting the eradication rates were analyzed by the logistic regression model and subgroup analysis. A *P* value of <0.05 was considered statistically significant. All statistics were analyzed using the Statistical Package for the Social Sciences, version 19.0 (SPSS, Chicago, IL).

RESULTS

Study population and baseline characteristics

A total of 72 patients underwent randomization of the 99 patients who were screened from January 2019 through June 2019 (Figure 1). Of these patients, 6 patients were excluded because 4 subjects were referred to surgery without endoscopic resection and 2 cases did not receive a trial medication. Finally, 66 were included in the PP population: 33 in the empirical therapy group and 33 in the clarithromycin resistance-guided tailored therapy group. Most of the demographic and tumor characteristics were similar between both groups; empirical group had more obese patients and bigger size of neoplasm (Table 1).

Eradication rates of *H. pylori*

The eradication rates of intention-to-treat analysis were 27 of 36 (75.0%) in the empirical therapy group and 32 of 36 (88.9%) in the tailored therapy group (*P* = 0.136). In PP analysis, the eradication rates of the tailored therapy and empirical therapy groups were 32 of 33 (97.0%) and 27 of 33 (81.8%), respectively (*P* = 0.046, Table 2). The factor affecting eradication failure was clarithromycin resistance (odds ratio, 0.48; 95% confidence interval: 0.007–0.346; *P* = 0.003) when adjusted for other variables including age, sex, body mass index, and regimen (Table 3). In the subgroup analysis for those with clarithromycin-resistant *H. pylori*, the eradication rate was much higher in the tailored group than that in the empirical group (91.7% vs 33.3%; *P* = 0.009). After the eradication rate of the tailored group was similar irrespective of clarithromycin resistance, that of the empirical group was significantly different the clarithromycin susceptible and resistant groups (*P* = 0.001, 88.9% vs 33.3%, Table 4). Although clarithromycin-resistant cases that were treated with quadruple therapy achieved total eradication in 10 of 10 patients (100%), eradication was achieved in 4/9 patients (44.4%) treated with triple therapy (*P* < 0.001). In addition, we analyzed the eradication rate according to specific disease, there were no difference among MALT lymphoma and gastric adenoma including benign disease and gastric cancer groups (*P* = 0.457, 100.0%, 90.3%, and 73.7%, respectively). Although *H. pylori* is one of etiologic factors of gastric cancer, gastric adenoma, and MALT lymphoma, these underlying diseases are not associated with the eradication rate of *H. pylori*.

Adverse effect and compliance

Compliance in both groups was similar and relatively good (Table 2). The rate of adverse effects of the tailored group was higher than that of the empirical group (*P* = 0.036) because the quadruple therapy group had more side effects than those of the triple therapy group (*P* = 0.001). One patient of the quadruple therapy group experienced paroxysmal supraventricular tachycardia that was less likely to be related to quadruple therapy. The patient was followed up with medication as an outpatient after being diagnosed at the emergency room.

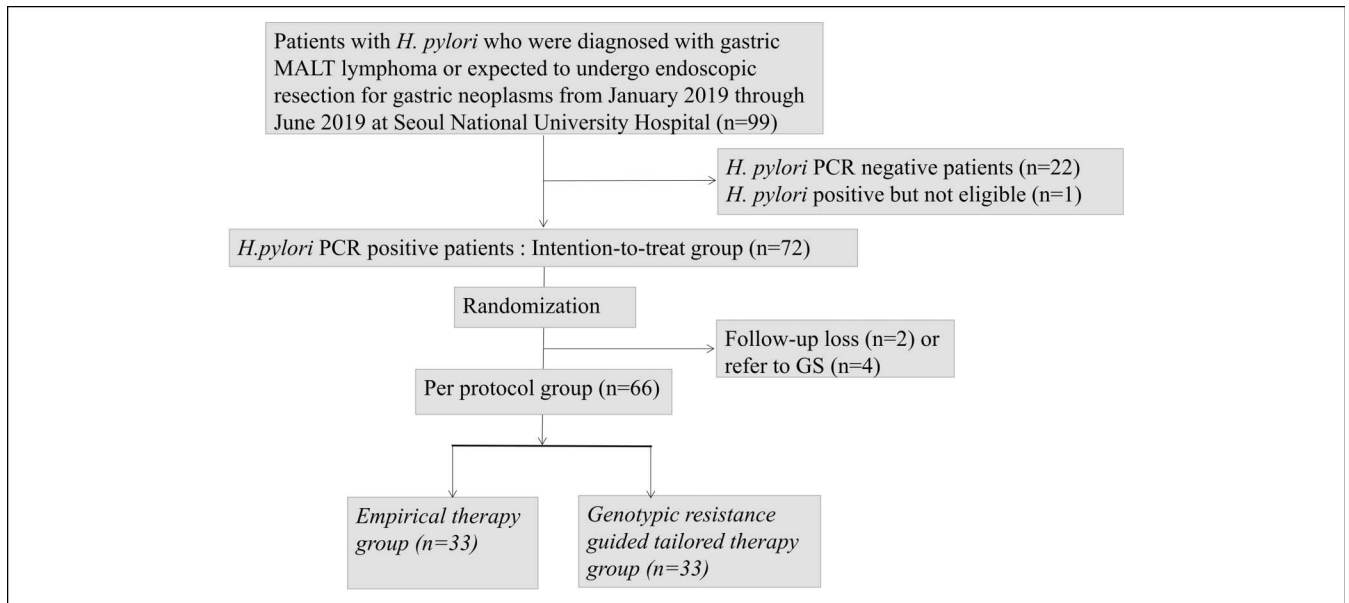


Figure 1. Enrollment, randomization, and follow-up. GS, general surgery; MALT, mucosa-associated lymphoid tissue; PCR, polymerase chain reaction.

Cost analysis

We compared the mean direct costs associated with empirical therapy with those for tailored therapy. The cost of 10-day standard triple therapy was calculated as 48.40 US Dollars (USD), whereas the cost of 10-day quadruple therapy was 34.67 USD, and the cost of genetic testing using DPO-PCR was 47.57 USD. A patient in the tailored therapy group paid an average of 250.86 USD, whereas a patient in the empirical therapy group paid an average of 217.86 USD. We included the prices of endoscopy, biopsy/histology, RUT, medication, and ^{13}C -UBT in the cost estimation. The costs for the tailored group were only 33 USD higher than those for the empirical group, and the cost per additional percentage increase of eradication rate was 2.17 USD (Table 5).

DISCUSSION

This study represents the first randomized, controlled trial to compare empirical triple therapy outcomes with those of tailored therapy based on the results of the DPO-PCR test. The present study demonstrated that the DPO-PCR-based eradication strategy could represent a promising alternative to increase eradication efficacy compared with the outcomes associated with empirical therapy. Clarithromycin resistance was a major factor affecting eradication failure.

The World Health Organization classified *H. pylori* as a group I carcinogen for gastric cancer in 1994 (23). The concept of “point of no return” suggested that new gastric cancers with advanced histologic changes could develop, even after the eradication of *H. pylori* (24,25). However, some randomized, controlled trials have suggested that *H. pylori* treatment improves gastric preneoplastic lesions and reduces the incidence of metachronous gastric cancers after the endoscopic resection of EGC (26–28). Because the necessity of *H. pylori* eradication has become clinically accepted, the national health insurance in South Korea has been amended to allow the treatment of all *H. pylori*-positive patients who wish to receive eradication treatment, starting in 2018.

In a meta-analysis of randomized, controlled trials, the cure rate for triple therapy was reported to be the unacceptable level of 68.9% in the ITT analysis (29). A recent Korean, randomized, multicenter analysis reported that the overall primary triple eradication rate was 63.9% in the ITT analysis, and 71.4% in the PP analysis (6). Compared with the ideal eradication rate, which is generally accepted as above 90% and preferably above 95% (30), the total eradication rate of clarithromycin-guided tailored therapy in the same study was 97.0% in the PP analysis and 88.9% in the ITT analysis, which approached the ideal rate.

Although our pilot study demonstrated a 30% difference between the eradication rates for the empirical group and the tailored group, this study reported only a 15.2% difference in the eradication rates because the eradication rate for the empirical group was higher than expected. The clarithromycin mutation rate was lower and the compliance rate was higher than expected. Although 47.8% of the empirical group and 45.8% of the tailored group presented with clarithromycin resistance in the pilot study, the ratio of clarithromycin resistance in this study was 25.0% for the empirical group and 33.3% for the tailored group. In addition, because compliance was extremely high, above 90%, the eradication rate may have increased in the present trial. These factors all contributed to the higher eradication rate for the control group than we expected.

Although this study did not include the prevalence of amoxicillin and metronidazole resistance, a recent multicenter study (9) showed that the resistance rates against clarithromycin, amoxicillin, and metronidazole, based on bacterial culture analysis, were 17.8%, 29.5%, and 9.5%, retrospectively (9). However, clarithromycin resistance was a more impactful factor for the eradication rate than resistance to the other antibiotics (12,31), which was consistent with the results of this study. As the clarithromycin-resistance rate of *H. pylori* increases, the eradication rate of primary triple therapy in Korea decreases. In this trial, the clarithromycin-resistance rate was relatively high, at 29.2%, which was similar to the 32% resistance rate reported in a previous study (12). The cure rate for clarithromycin-susceptible strains was significantly higher

Table 1. Baseline characteristics of the patients in the intention-to-treat population

Variables	Empirical group (n = 36)	Tailored group (n = 36)	P-value
Age, yr, mean ± SD	67.5 ± 9.7	64.1 ± 8.6	0.075
Sex			
Male, n (%)	24 (66.7)	17 (47.2)	0.096
Female, n (%)	12 (33.3)	19 (52.8)	
BMI, kg/m ² , mean ± SD	25.9 ± 3.4	24.4 ± 2.5	0.045
Obesity, n (%) ^a	23 (65.7)	10 (29.4)	0.002
Comorbidity			
Hypertension, n (%)	23 (63.9)	14 (38.9)	0.034
Diabetes mellitus, n (%)	12 (33.3)	9 (25.0)	0.437
Thyroid disease, n (%)	5 (13.9)	1 (2.8)	0.088
Other malignancy, n (%)	2 (5.6)	1 (2.8)	0.555
Cigarette smoking, n (%)	13 (36.1)	7 (19.4)	0.387
Amount of smoking, PY, mean ± SD	11.1 (17.4)	5.2 (11.2)	0.082
Alcohol drinking, n (%)	16 (44.4)	11 (31.4)	0.259
Amount of alcohol drinking, kg, mean ± SD ^b	97.6 ± 206.1	67.2 ± 174.4	0.2
Clarithromycin resistance			
Total, n (%)	9 (25.0)	12 (33.3)	0.546
A2142G and A2143G, n (%)	1 (2.8)	0 (0)	
A2142G, n (%)	2 (5.6)	4 (11.1)	
A2143G, n (%)	6 (16.7)	8 (22.2)	
Regimen			
Triple therapy, n (%)	33 (91.7)	23 (63.9)	0.003
Quadruple therapy, n (%)	0 (0)	10 (27.8)	
Treatment modality			
Endoscopic resection, n (%)	33 (91.7)	27 (75.0)	0.139
Surgery, n (%)	2 (5.6)	3 (8.3)	
Argon plasma coagulation, n (%)	1 (2.8)	2 (5.6)	
Biopsy only or medical treatment, n (%)	0 (0)	4 (11.1)	
Classification of gastric lesions			
Peptic ulcer, n (%)	0 (0)	0 (0)	NA
MALT lymphoma, n (%)	0 (0)	3 (8.3)	0.197
Gastric neoplasm			
Gastric adenoma and benign disease, n (%)	13 (36.1)	18 (50.0)	0.06
Gastric cancer, n (%)	23 (63.9)	15 (41.7)	
Shape of lesion			
Elevated or flat, n (%)	19 (52.8)	20 (55.6)	0.813

Table 1. (continued)

Variables	Empirical group (n = 36)	Tailored group (n = 36)	P-value
Depressed or ulcerative, n (%)	17 (47.2)	16 (44.4)	
Location of lesion			
Upper third, n (%)	4 (11.1)	6 (16.7)	0.388
Middle third, n (%)	5 (13.9)	9 (25.0)	
Lower third, n (%)	27 (75.0)	21 (58.4)	
Size of neoplasm, mm, mean ± SD	18.2 ± 9.5	12.5 ± 8.7	0.016
Depth of cancer			
T1m, n (%)	21 (58.3)	11 (30.5)	0.227
T1sm1, n (%)	1 (2.8)	0 (0)	
T1sm2, n (%)	1 (2.8)	4 (11.1)	
Histology of gastric dysplasia or cancer			
Low grade dysplasia, n (%)	9 (25.0)	11 (30.6)	0.095
High grade dysplasia, n (%)	3 (8.3)	2 (5.6)	
Differentiated, n (%)	23 (63.9)	15 (39.5)	
Undifferentiated, n (%)	0 (0)	0 (0)	
Complete resection ^c			
Yes, n (%)	32 (88.9)	27 (75.0)	0.202
No, n (%)	1 (2.8)	0 (0)	

BMI, body mass index; m, mucosal cancer; MALT, mucosa-associated lymphoid tissue; NA, not applicable; sm, submucosal cancer.
^aObesity was defined as BMI 25 or higher.
^bOne bottle of soju (17', 360 mL) was calculated as 49 g of alcohol.
^cComplete resection was assessed among the cases of endoscopic resection.

than that for clarithromycin-resistant strains (88.2% vs 66.7%, $P = 0.031$, in the ITT analysis; 95.7% vs 73.7%, $P = 0.008$, in the PP analysis). The eradication rate for triple therapy was 91.5% in clarithromycin-susceptible patients and 44.4% in clarithromycin-resistant patients. Therefore, the development of improved treatments for clarithromycin-resistant patients remains important for increasing the eradication rate.

The Maastricht V guideline does not recommend standard triple therapy without previous susceptibility testing in regions where the clarithromycin-resistance rate is above 15% (8). Standard culture studies, based on MIC, can be difficult to perform in a clinical setting because they are time consuming, inconvenient, and not standardized. Recently, DPO-based multiplex PCR was introduced for the detection of *H. pylori* and clarithromycin susceptibility (11). DPO-based multiplex PCR is a strategy for the accurate detection of the most common point mutations that occur in the 23S rRNA gene of *H. pylori*, based on gastric biopsy samples, and the results can be obtained within a few hours. DPO-PCR has been reported to have a sensitivity of 97.7% and a specificity of 83.1% when using the MIC cultures as the reference test, and the concordance with phenotypic susceptibility testing was 95.3% (10).

Table 2. Eradication rate, adverse effects, and compliance

Variables	Empirical group (n = 36)	Tailored group (n = 36)	P Value
Eradication rate			
Eradication rate (ITT analysis), n (%)	27/36 (75.0)	32/36 (88.9)	0.136
Eradication rate (PP analysis), n (%)	27/33 (81.8)	32/33 (97.0)	0.046
Adverse effects and compliance			
Any adverse effect, n (%)	1 (3.1)	6 (20.0)	0.036
Skin rash, n (%)	0 (0)	1 (3.1)	
Taste distortion, n (%)	1 (3.1)	0 (0)	
Bloating, n (%)	0 (0)	0 (0)	
Abdominal pain, n (%)	0 (0)	1 (3.1)	
Nausea, n (%)	0 (0)	1 (3.1)	
Vomiting, n (%)	0 (0)	0 (0)	
Diarrhea, n (%)	0 (0)	1 (3.1)	
Constipation, n (%)	0 (0)	1 (3.1)	
Cardiac arrhythmia, n (%)	0 (0)	1 (3.1)	
Poor compliance, n (%)	0 (0)	0 (0)	NA
Variables	Triple group (n = 56)	Quadruple group (n = 10)	P Value
Adverse effects			
Any adverse effect, n (%)	3 (5.4)	4 (40.0)	0.001
Skin rash, n (%)	1 (1.8)	0 (0)	
Taste distortion, n (%)	1 (1.8)	0 (0)	
Bloating, n (%)	0 (0)	0 (0)	
Abdominal pain, n (%)	0 (0)	1 (10.0)	
Nausea, n (%)	0 (0)	1 (10.0)	
Vomiting, n (%)	0 (0)	0 (0)	
Diarrhea, n (%)	1 (1.8)	0 (0)	
Constipation, n (%)	0 (0)	1 (10.0)	
Cardiac arrhythmia, n (%)	0 (0)	1 (10.0)	

ITT, intention-to-treat; NA, not applicable; PP, per-protocol.

When compared with the ¹³C-UBT, *H. pylori* detection by DPO-PCR had a sensitivity of 87.5%, a specificity of 91.3%, and an accuracy of 90.0% (21). In the present study, the eradication rate of the tailored group was superior to that of the empirical group (97%, in the PP analysis, $P = 0.046$ vs 88.9%, in the ITT, $P = 0.136$). Therefore, the assessment of clarithromycin resistance using the DPO-based multiplex PCR method in advance of treatment is recommended, as is the administration of quadruple therapy to patients with demonstrated clarithromycin resistance.

The Maastricht V guideline also recommended quadruple therapy as an alternative primary treatment method in areas with high levels of clarithromycin resistance (8). The efficacy rates between tailored therapy and quadruple therapy are predicted to be similar in regions with a high prevalence of clarithromycin resistance. However, the cure rate of clarithromycin-susceptible strains treated with triple therapy reached the ideal value of 91.5% in this trial and in previous studies (91%–96%) (12,31). Although clarithromycin-susceptible patients are likely to achieve the ideal treatment outcome, regardless of whether they receive triple or quadruple therapy as the first-line treatment, adverse events were reported more frequently for the quadruple therapy treatment group than for the triple therapy group in our study (40% vs 5.4%, $P = 0.001$). Therefore, triple therapy continues to be advantageous for clarithromycin-susceptible patients. However, clarithromycin-resistant strains require the use of quadruple therapy as the primary treatment because triple therapy in these strains only achieved an eradication rate of 44.4%, whereas quadruple therapy in these patients achieved a cure rate of 100%.

In a recent, randomized, multicenter study, 10-day sequential therapy was recommended as an alternative primary treatment method for *H. pylori* infections in a country with clarithromycin resistance higher than 15% (6). The eradication rate of the 10-day sequential therapy was superior to that of 7-day triple therapy. The eradication rates were 76.3% in the ITT analyses and 85.0% in the PP analyses. One meta-analysis suggested that sequential therapy was able to eradicate 72.8% of strains resistant to clarithromycin (7). When compared with triple therapy that lasted 7 and 10 days, the 10-day sequential therapy had better eradication rates of 86.5% and 84.3%, respectively. However, the present study showed that tailored therapy achieved superior results compared with those for sequential therapy, achieving eradication rates of 88.9% in the ITT analysis and 97.0% in the PP analysis.

Previous studies (32) have examined tailored therapy, using various treatment regimens and have reported various eradication rates. The efficacy of the tailored treatment varies. The efficacies (eradication rates of tailored therapy [in the ITT analysis]) have been reported as 71.9%, 72%, 80%, 86%, 91%, 94.5%, 94.6%, and 96%. However, these population results are not transferable to other geographical areas with different patterns of resistance. The most important strength of our study was the development of a method that was able to determine whether to apply tailored therapy to specific patients in a region with a high incidence of clarithromycin resistance and an intermediate risk of metronidazole resistance.

Many regimens associated with tailored therapies that have been reported in previous studies included PPI, amoxicillin, and metronidazole when clarithromycin-resistant strains were detected. A meta-analysis (33) showed that the eradication rate of triple therapy including PPI, amoxicillin, and metronidazole was lower than that for triple therapy including PPI, amoxicillin, and clarithromycin. In settings with high levels of clarithromycin resistance, the choice of therapy should be based on the frequency of metronidazole and dual clarithromycin and metronidazole resistance. In geographical areas where metronidazole resistance is almost negligible (e.g., Japan), replacing clarithromycin with metronidazole during triple therapy (i.e., PPI-metronidazole-amoxicillin) continues to demonstrate excellent cure rates (34). Metronidazole resistance in Korea has been reported to range from 9% to 30.3% (9,35). The Maastricht V/Florence Consensus guideline (8) recommended that in areas with high dual

Table 3. Factors affecting eradication rates (intention-to-treat analysis)

Eradication rate in subgroups	Total (n = 72)	Empirical group (n = 36)	Tailored group (n = 36)	Multivariate analysis	
				OR (95% CI)	P Value
Sex					
Male, n (%)	41/72 (56.9)	17/24 (70.8)	16/17 (94.1)	Reference	
Female, n (%)	31/72 (43.1)	10/12 (83.3)	16/19 (84.2)	0.898 (0.104–7.741)	0.922
Age, yr, mean (SD)					
<65 yr old, n (%)	25/28 (89.3)	10/13 (76.9)	17/20 (85.0)	Reference	
≥65 yr old, n (%)	33/37 (89.2)	17/23 (73.9)	15/16 (93.8)	1.048 (0.131–8.358)	0.965
BMI, kg/m ² , mean (SD)					
<25 kg/m ² , n (%)	31/34 (91.2)	8/13 (61.5)	23/26 (88.5)	Reference	
≥25 kg/m ² , n (%)	27/31 (87.1)	19/23 (82.6)	9/10 (90.0)	1.027 (0.110–9.585)	0.981
Clarithromycin resistance					
Clarithromycin susceptible, n (%)	51/72 (70.8)	24/27 (88.9)	21/24 (87.5)	Reference	
Clarithromycin resistant, n (%)	21/72 (29.2)	3/9 (33.3)	11/12 (91.7)	0.48 (0.007–0.346)	0.003
Regimen					
Triple therapy, n (%)	56/72 (77.8)	27/33 (81.1)	22/23 (95.7)	Reference	
Quadruple therapy, n (%)	10/72 (13.9)	NA	10/10 (100)	NA	NA

BMI, body mass index; CI, confidence interval; OR, odds ratio; NA, not applicable.

clarithromycin and metronidazole resistance, bismuth quadruple therapy is the recommended first-line treatment. Therefore, based on the presence of clarithromycin resistance, choosing regimens consisting of PPI, amoxicillin, and metronidazole is not reasonable in Korea. Many previous studies have adopted the combination of PPI, amoxicillin, and metronidazole for tailored regimens, and various eradication rates have been reported for tailored therapy groups; however, this combination is not a good choice in Korea. Our study was a prospective, randomized, controlled trial, in that we chose the therapeutic medications for the tailored group in advance, based on clarithromycin

sensitivity, and investigated eradication rates according to clarithromycin susceptibility. Tailored therapy using a bismuth quadruple therapy regimen to treat the presence of *H. pylori* in Korea is a reasonable option to adopt in the future.

Tailored therapy based on DPO-PCR results also showed cost benefits when the eradication rate of the 14-day empirical therapy declined below 80% (19). When we analyzed the mean direct prices of both types of therapy, a patient in tailored therapy using DPO-PCR would pay approximately 250.86 USD, including costs associated with endoscopy, biopsy/histology, RUT, DPO-PCR test, medication, and ¹³C-UBT. Most

Table 4. Eradication rates according to clarithromycin resistance and group (intention-to-treat analysis)

Eradication rate in subgroups	Clarithromycin susceptible (n = 51)	Clarithromycin resistant (n = 21)	χ^2 test P Value	Logistic regression analysis	
				OR (95% CI)	P Value
Eradication rate					
Total	45/51 (88.2)	14/21 (66.7)	0.031		
Empirical group	24/27 (88.9)	3/9 (33.3)	0.001	Reference	
Tailored group	21/24 (87.5)	11/12 (91.7)	0.773	7.111 (0.805–62.786)	0.078
Eradication rate by regimen					
Eradication rate in subgroups	Triple therapy (n = 56)	Quadruple therapy (n = 10)	χ^2 test P Value	Logistic regression analysis	
Eradication rate					
Total	47/56 (83.9)	10/10 (100)	<0.001		
Clarithromycin susceptible	43/47 (91.5)	NA	NA	Reference	
Clarithromycin resistant	4/9 (44.4)	10/10 (100)	<0.001	0.124 (0.022–0.713)	0.019

CI, confidence interval; OR, odds ratio; NA, not applicable.

Table 5. Cost estimates

Measures	Cost estimates	Calculation
Endoscopy	95.13 USD	
Biopsy/histology	42.58 USD	
RUT	10.88 USD	
Quadruple therapy for 10 d	34.67 USD	
Standard triple therapy for 10 d	48.4 USD	
Genetic testing (DPO-PCR)	47.57 USD	
Tailored therapy	250.86 USD	$Cost_{endoscopy} + Cost_{histology} + Cost_{RUT} + Cost_{DPO_PCR\ test} + Cost_{medication} + Cost_{^{13}C_UBT}$
Standard therapy	217.86 USD	$Cost_{endoscopy} + Cost_{histology} + Cost_{RUT} + Cost_{medication} + Cost_{^{13}C_UBT}$
ICERs	2.17 USD per 1% eradication rate gained	$\frac{Cost_{tailored\ group} - Cost_{empirical\ group}}{Eradication\ rate_{tailored\ group} - Eradication\ rate_{empirical\ group}}$

The ICER is defined as the difference in average total costs for the tailored group minus the empirical group divided by the average difference in the eradication rate. C-UBT, C-urea breath test; DPO-PCR, dual-priming oligonucleotide-based multiplex-polymerase chain reaction test; ICER, incremental cost-effectiveness ratio; RUT, rapid urease test; USD, United States dollars.

patients in the tailored therapy group were successfully treated with one remedy (97.0% in our PP analysis). By contrast, a patient treated with empirical triple therapy would pay 217.86 USD, including the costs associated with endoscopy, biopsy/histology, RUT, medication, and ^{13}C -UBT. Considering the cost per additional percentage increase of eradication rate of 2.17 USD, the expenses associated with the use of second-line medications, additional ^{13}C -UBT evaluations, outpatient clinic registration, and revisits because of treatment failure, the costs of tailored therapy using DPO-PCR are unlikely to be higher than those for empirical therapy. Therefore, clarithromycin-guided tailored therapy allows physicians to choose triple or quadruple therapy to treat *H. pylori* infection in regions with a high prevalence of clarithromycin resistance.

This study has several limitations. First, it was conducted in a tertiary single center because the eligible patients were referred for the treatment of gastric MALT lymphoma, gastric adenoma, and EGC. However, we attempted to maintain consistency during the diagnostic procedures, the establishment of a treatment plan, and treatment because endoscopic resections are sophisticated and technically demanding procedures. Second, ethical issues may apply to our trial design because of the use of an empirical group. However, in South Korea, standard triple therapy has generally remained the primary therapy option in clinical practice and was able to eradicate *H. pylori* in up to 75% of the empirical group population in this study. Our study suggests a practical method for increasing the eradication rate of *H. pylori* infections in South Korea.

In conclusion, clarithromycin resistance-guided tailored therapy, based on DPO-multiplex PCR, was superior to empirical therapy for first-line eradication treatment in a region with a high prevalence of clarithromycin resistance.

CONFLICTS OF INTEREST

Guarantor of the article: Soo-Jeong Cho, MD, PhD.

Specific author contributions: J.L.K., S.-J.C.: planning and/or conducting the study. J.L.K., S.-J.C., S.J.C., A.L., J.C., H.C.: collecting and/

or interpreting data. J.L.K., S.-J.C., S.G.K.: drafting/revision of the manuscript. J.L.K., S.-J.C., S.J.C., A.L., J.C., H.C., S.G.K.: approval of the final draft of the submitted manuscript.

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Potential competing interests: None to report.

Clinical trial registration: NCT04006340.

Study Highlights

WHAT IS KNOWN

- ✓ Eradication rate of triple therapy is unsatisfactory as antibiotic resistance rate of *H. pylori* increases.
- ✓ Clarithromycin resistance of *H. pylori* based on 23S rRNA point mutations is a major cause of eradication failure.
- ✓ DPO-PCR can detect *H. pylori*, which had a good sensitivity, specificity, and concordance rate.

WHAT IS NEW HERE

- ✓ DPO-PCR based eradication strategy had superior eradication efficacy to empirical therapy in a region of high prevalence of clarithromycin resistance.
- ✓ Tailored group had more side effects, which were mild degree not to affect compliance.

TRANSLATIONAL IMPACT

- ✓ PCR-based tailored therapy increases eradication rate of *H. pylori* infection.

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