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Helicobacter pylori Eradication Rate in Clarithromycin-resistant Strains By Pantoprazole - Amoxicillin - Clarithromycin Regimen

***Phadet Noophun, M.D.* ***
Varocha Mahachai, M.D.*
Duangporn Thong-ngam, M.D.[+]
Ratha-korn Vilaichone, M.D.,Ph.D.**
Somying Tumwasorn, Ph.D.[#]
Pinit Kullavanijaya, M.B.chB.*

ABSTRACT

Background: Clarithromycin resistance is an increasing problem in this part of the world and it potentially has an impact on the eradication rate of first line triple therapy (proton pump inhibitor- Amoxicillin-Clarithromycin) for *Helicobacter pylori (H. pylori)* infection.

Objective: This study was designed to determine the effect of clarithromycin resistance on the efficacy of Pantoprazole-Amoxicillin-Clarithromycin for *H. pylori* eradication in Thai patients with non-ulcer dyspepsia (NUD).

Patients and Methods: A total of 69 patients with NUD who had undergone upper endoscopy for dyspeptic symptom and had *H. pylori* infection as determined by positive urease test and positive culture for *H. pylori* were enrolled in this study. Minimal inhibitory concentrations (MICs) of Clarithromycin were identified using Epsilometer test (E-test). The value of MICs cutpoint for Clarithromycin resistant was >1 microgram/ml. There were 16 patients who had Clarithromycin resistant *H. pylori* and 53 patients with Clarithromycin sensitive *H. pylori*. Both groups of patients received Pantoprazole (Controloc®) 40 mg b.i.d., Amoxicillin 500 mg b.i.d. and Clarithromycin (Klacid MR®) 500 mg 2 tablets daily for 1 week. *H. pylori* eradication was evaluated using ^{14}C urease test (PY-test®) one month after triple therapy was discontinued.

Results: Primary *H. pylori* resistance to Clarithromycin was observed in 16 of 69 patients (23.2%). The eradication rate were 90.6% (48/53) and 56.3% (9/16) in patients with Clarithromycin sensitive and resistant *H. pylori*, respectively. The difference in eradication rate between sensitive and resistant strains was statistically significant (p = 0.002).

Conclusions: Clarithromycin resistant *H. pylori* is increasing in this part of the world. This finding may have a significant impact on the outcome of *H. pylori* eradication using regimen containing Clarithromycin.

Key words : *Helicobacter pylori,* eradication, Clarithromycin-resistant

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* *Gastroenterology Unit, Department of Internal Medicine,* [+] *Department of Physiology,*
\# *Department of Microbiology, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand*
** *Gastroenterology Unit, Department of Internal Medicine, Thammasat University Hospital, Bangkok, Thailand*

BACKGROUND

Helicobacter pylori (H. pylori) was first described by Warren and Marshall in 1982[1,2]. *H. pylori* is a gram negative, spiral-shaped organism that is associated with many gastrointestinal diseases in humans. The discovery of *H. pylori* infection plays role in several diseases of the stomach and duodenum which has been a breakthrough in gastroenterology. It has a world wide prevalence, with approximately 50% of the world's population infected. The natural history of *H.pylori* infection is highly variable and influenced by environmental, host genetic and microbial factors. The pattern and distribution of gastritis correlate with risks of clinical outcomes including peptic ulcers and its complications; atrophic gastritis, gastric carcinoma and gastric lymphoma[3]. In 1994, the International Agency for Research on Cancer (IARC) declared H.pylori to be a group I human carcinogen for gastric adenocarcinoma[4]. According to the Maastricht 2-2000 consenses, strong recommendations to eradicate *H. pylori* include, active or inactive peptic ulcer diseases, MALToma, atrophic gastritis, post-gastric cancer resection and patients who are first-degree relatives of gastric cancer patients[5].

First-line therapy for *H. pylori* eradication should be with triple therapy using a proton pump inhibitor or ranitidine bismuth citrate, combined with clarithromycin and amoxicillin or metronidazole. Treatment failure may be attributed generally to poor compliance with drug regimen or to acquired antibiotic resistance to organism. Clarithromycin resistance in *H. pylori* is a growing problem in many parts of the word.

The prevalence of *H. pylori* resistant has been reported as 24% in Mexico[6], 19% in France[7], 13.5% in China[8], 12.9% in Japan[9], 12% in United State[10], 10.8% in Hong Kong[11], 5.9% in Korea[12] and the latest data 19% in Thailand[13]. Few data are available from clinical trials to evaluate the impact of clarithromycin resistance on eradication. This study was designed to determine the effect of clarithromycin resistance on the efficacy of pantoprazole-amoxicillin-clarithromycin for *H. pylori* eradication in Thai patients with non ulcer dyspepsia.

PATIENTS AND METHODS

A total of 470 patients who had undergone upper gastrointestinal endoscopy for dyspeptic symptoms diagnosed as non-ulcer dyspepsia at King Chulalongkorn Memorial Hospital were included in the study. Patients were excluded from the study if they had history of gastric surgery, previous antibiotics, bismuth preparations or proton pump inhibitors within a month before the study, had serious concomitant medical conditions such as chronic renal failure, cirrhosis, cancer, AIDS, stroke, coagulopathy, history of allergy to proton pump inhibitor,penicillin, macrolide, were pregnant or on breast feeding.

Three specimens of biopsy from gastric antrum were obtained from each patient, one specimen for rapid urease test and two specimen for culture and antibiotic susceptibility test. Minimal inhibitory concentrations (MICs) of clarithromycin was determined by the E-test method (AS Brodisk, Solna Sweden) Strains were considered resistant to clarithromycin if MIC >1.0 (g/ml)[13]. Infection by *H. pylori* was defined as positive by rapid urease test and culture. A total of 69 patients out of 470 patients had positive *H. pylori* by rapid urease test and culture. These 69 patients received 1 week triple therapy with pantoprazole (Controloc®) 40 mg and amoxicillin 1 gm twice daily. Clarithromycin (Klacid MR®) 500 mg 2 tablets daily.

Patients were then asked to return after completing 1 week of triple therapy for assessment of compliance. Eradication was determined by ^{14}C-urea breath test (PY test®) 4 to 6 weeks after completion of triple therapy. Successful eradication was defined by a negative urea breath test. The successful *H. pylori* eradication rate was calculated as the percentage of patients whose is *H. pylori* were undetectable by urea breath test after triple therapy.

The data was analyzed by SPSS for window. Descriptive data was presented as mean and standard deviation. Comparison between the efficacy of treatment in each patient group was evaluated by the two tailed chi-square and Fisher's exact tests. A $p < 0.05$ was considered statistical significance.

RESULTS

Between September 2002 and December 2003 *H. pylori* was identified by positive rapid urease test in 285 patients (55.7 %) Then, cultures for *H. pylori* were achieved in 113 patients (43.1%) and E-test for clarithromycin were successfully identified in 69 isolations. (Figure 1) There were 29 males (42.1%) and 40 females (57.9%). With a mean ages of 40.22 years

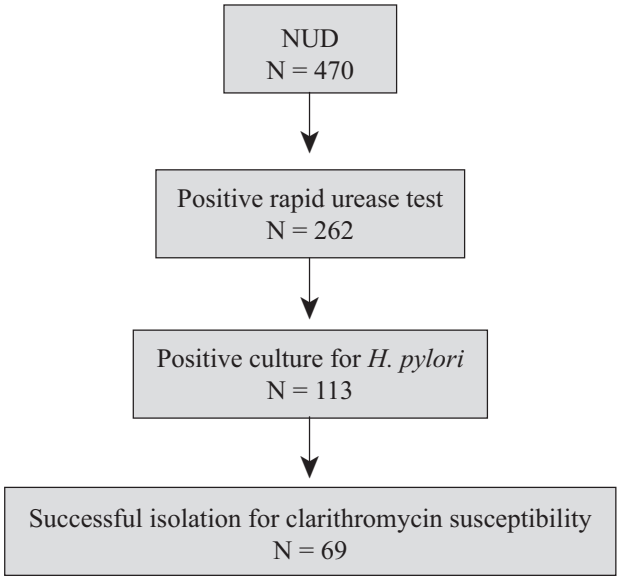

Figure 1 Flow chart of patients recruited.

Table 1 Baseline characteristic of the patients

Characteristics	Sensitive Strains N = 53	Resistant Strains N = 16
Sex		
Male:Female	24:29	5:9
(%)	(45:55)	(31:69)
Age (yr.)	39.4 ± 13.7	45.7 ± 17.3
Smoking		
No. of patient (%)	7 (13.2)	5 (68.8)
Alcohol		
No. of patient (%)	12 (22.6)	4 (25.0)
Severity of dyspepsia		
- moderate	23 (43.4)	4 (25)
- severe	30 (56.6)	12 (75)

(range 19-75 years). Clarithromycin-sensitive *H. pylori* was isolated from 53 patients (76.8%), and clarithromycin-resistant *H. pylori* from16 patients (23.2%). Baseline-characteristics of the patients were shown (Table 1).

The most common endoscopic finding in both groups is antral gastritis (40.6%). Out of these 69 patients, 16 patients (23.2%) were found to have clarithromycin-resistant *H. pylori* and 53 patients (76.8%) were infected with clarithromycin-sensitive *H. pylori*. Both groups received clarithromycin containing triple therapy. *H. pylori* eradication was assessed by urea breath test (^{14}C UBT) which resulted in successful eradication in 9 out of 16 patients (56.2%) in clarithromycin resistant strains, and 48 out of 53 patients (90.6%) in clarithromycin resistant strains as shown (Table 2). The difference in eradication rate between both groups was statistically significant ($p = 0.002$). There were no significant differences between clarithromycin sensitive and resistance groups in regards to age, sex, number of patients who smoked or consumed alcohol, or severity of dyspepsia. Few adverse events were observed in these patients taking this regimen as shown (Table 3) and there were no serious side effects. None of the patients from the study were lost to follow up.

DISCUSSION

Antibiotic-resistant *H. pylori* is increasing in many parts of the world. Acquired resistance of *H. pylori* to clarithromycin and metronidazole have been reported worldwide[6-13]. This could have a significant clinical impact on eradication using standard triple therapy. Primary resistance of *H. pylori* to metronidazole is increasing and is typically high in popu-

Table 2 Eradication rate between clarithromycin sensitive and resistant *H. pylori* using combination of Pantoprazole-Amoxicillin-Clarithromycin.

H. pylori / Patients	Successful Eradication (patients)	Failed Eradication (patients)	Total (patients)
Sensitive strains (n = 53)	48 (90.6%)	5 (9.4%)	53
Resistant strains (n = 16)	9 (56.2%)	7 (43.8%)	16

p = 0.002

Table 3 Adverse events from pantoprazole-amoxicillin-clarithromycin regimen

Adverse Events	No. of Patients	Percents
Nausea / Vomiting	40	58.0
Metallic tongue	8	11.6
Diarrhea	6	8.7
Dizziness	5	7.2
Headache	4	5.8
Constipation	3	4.3

lations where metronidazole is used leberally, which is associated with a reduction in eradication rate. In the United States, the frequency of resistance to metronidazole ranges from 20-50% but is higher in woman and immigrants from semitropical countries[15]. Meta-analysis by Maria *et al.*[16], found that metronidazole resistance reduced the eradication rate by an average of 37.3% (95%CI = 29.6-45.7%). Wongkusoltham *et al.*[17], reported, prevalence of metronidazole resistant strains was 51.92%, but the eradication rate for *H. pylori* by metronidazole containing regimen between metronidazole resistant (MR) and sensitive (MS) group was not statistically significant (MR = 91.30, MS = 95.65%, p >0.05).

Graham *et al.*[18] found that metronidazole resistance can be overcome by increasing the dosage (e.g. from 250 mg three or four times daily to 500 mg three times daily). The prevalence of primary clarithromycin resistance *H. pylori* reported in this study (23.2%) is higher than other data reported from other asian countries[8,9,11,12].

In the United states[15], clarithromycin resistance is reported to be in the range of 7-1 %. Metaanalysis[16] showed that clarithromycin resistance reduced effectiveness by an average of 55% (95%CI = 33-78%).

In this study, the eradication rate in clarithromycin resistant group is lower than that in clarithromycin sensitive group (56.2% vs 90.6%; p = 0.002), indicated that primary clarithromycin resistant *H. pylori* can result in failure of eradication using clarithromycin containing triple therapy. Previous studies revealed that clarithromycin-resistant *H. pylori* decreased the outcome of clarithromycin containing regimen. Tankovic *et al.*[7], found that eradication rate for clarithromycin sensitive and resistant *H. pylori* were 79% and 12% respectively, the difference was statistically significant (p <0.05). Other studies had similar outcomes, but only a small number of clarithromycin resistant strains were included. Lamouliatte *et al.*[19] found eradication rate for clarithromycin resistant *H. pylori* was 0% (0 of 5 patients), Wurzer *et al.*[20] showed eradication rate for clarithromycin resistant *H. pylori* of 50% (3 of 6 patients), but Lind *et al.*[21] found that the two clarithromycin resistant strains were successfully eradicated. There were no significant differences between patients in groups of clarithromycin resistant and sensitive strains regards to age,sex,number of patients who smoked or consumed alcohol and severity of dyspepsia.

The mechanism involved in clarithromycin resistant *H. pylori* is now well recognized at the molecular level[22]. Resistance to clarithromycin in *H. pylori* is due to lack of clarithromycin binding to the 23 S ribosomal RNA gene in one of two positions (2143 and 2144)[23]. More recently, it has been suggested that mutations in positions 2116 and 2142 have been described but seem to occur quite seldomly[24]. The consequence is a high level of resistance, which is easy to detect phenotypically. To avoid the emergence of resistance to both key antibiotics, the combination of metronidazole and clarithromycin should be avoided where possible. In case of persistent treatment failure, it is useful to consider repeating gastroscopy to obtain tissue for culture and then prescribe antibiotics according to the bacterial susceptibility pattern. Molecular assays for detecting clarithromycin resistance in *H. pylori* are based on detection of mutations in the 23S RNA genes. The PCR line probe assay (LiPA) system is simple and cost effective alternative capable of detecting seven distinct resistance mutations and is highly suitable for testing large numbers of samples.

In conclusion,we reported a high rate of resistance to clarithromycin in *H. pylori* isolates in this part of the world. Primary resistance to clarithromycin can have a significant impact on eradication using clarithromycin based regimen. The simple and cost effective molecular assays for detecting clarithromycin resistance in *H. pylori* is potentially useful in this part of the world.

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