Does the urea breath test predict eradication of *Helicobacter pylori* infection ?

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Abstract

Background/Aim : ***Helicobacter pylori*** **infection is common worldwide and has been linked to development of gastric and duodenal ulcers, gastric adenocarcinoma, and gastric lymphoma. However, antimicrobial resistance has decreased *H. pylori* eradication rates worldwide. This study aimed to evaluate the effect of bacterial load on eradication rate.**

Method : **This prospective study included 237 consecutive patients who presented to our institution with dyspeptic symptoms and underwent both upper endoscopy and urea breath tests (UBT). The patients were divided into three equal sized groups according to their UBT values. All subjects received a standard triple eradication regimen, followed by a bismuth- based quadruple eradication regimen if triple eradication was not successful. The three groups were compared with respect to age, endoscopic findings, sex, and eradication rates.**

Results : **Our results were consistent with those of previous studies : higher UBT values were associated with failure of standard 14-day triple treatment ($p < 0.05$). However, in patients who received a quadruple eradication regimen, differences between groups were not significant ($p = 0.434$). There was no relationship between UBT values and gastric pathologies ($p = 0.751$). Age and sex also did not differ significantly between groups ($p = 0.061$).**

Conclusions : **Our study and others have found that high bacterial loads are negatively associated with achievement of eradication with triple treatment. However, differences between groups were not significant in patients who received a quadruple eradication regimen. Comparisons of treatment results according to bacterial density may be informative. The importance of *H. pylori* density should be further evaluated with new treatment protocols.** (Acta gastroenterol. belg., **2016**, 79, **3-7**).

Key words : *Helicobacter pylori*, eradication, urea breath test.

Introduction

Helicobacter pylori infection is the most common infection worldwide. This infection is contracted in childhood and, if untreated, persists as a lifelong, chronic infection. It is well established that *H. pylori* is involved in the pathogenesis of chronic gastritis, peptic ulcers, B cell gastric lymphoma, and gastric adenocarcinoma (1-3).

Once research provided evidence that *H. pylori* infection is the main cause of many gastric diseases, antibiotic treatment became standard. Currently, a triple combination treatment consisting of two antibiotics (clarithromycin plus amoxicillin or metronidazole) and a proton pump inhibitor (PPI) is the first-line therapy for *H. pylori* eradication in regions with clarithromycin resistance rates below 15-20%. The Maastricht III Consensus Report indicated that effective *H. pylori* treatment should achieve an intention-to-treat (ITT) eradication rate of over 80% and that the triple eradication regimen provides eradication rates close to 80% in certain regions with low resistance (4). However, recently reported eradication rates achieved by PPI-based triple therapy are lower than 80% ; rescue regimens have therefore emerged. The main factors associated with poor therapy responses are antibiotic resistance and poor patient compliance. Bacterial load has also been reported to affect eradication rates (2-4). Both C^{14} urea breath tests (UBTs) and histologic examination of biopsy specimens can be used to measure *H. pylori* bacterial load. In this study, we evaluated the effect of bacterial load on eradication rates.

Materials and Methods

Patient Population

This study involved 380 consecutive eligible patients with upper gastrointestinal symptoms admitted to the gastroenterology clinic of Baskent University Konya Hospital in Konya, Turkey, between March and August 2009. Exclusion criteria included prior *H. pylori* eradication therapy ; use of PPIs, H-2 receptor blockers, bismuth preparations, prokinetics, non-steroidal anti-inflammatory drugs, or antibiotics in the preceding two months ; alcoholism ; history of gastric surgery or cholecystectomy ; decompensated congestive heart failure ; liver cirrhosis ; chronic renal failure requiring dialysis ; prior diagnosis of malignancy ; pregnancy ; and active infections requiring antibiotic therapy.

Diagnosis of H. pylori *Infections*

All patients included in this study underwent upper gastrointestinal endoscopy (Pentax 2940 ; Tokyo, Japan). *H. pylori* infection status was evaluated by a rapid urease test (RUT) Following endoscopy, patients with positive RUT results underwent C^{14} UBT. After overnight fasting, they swallowed a urea capsule (Helicap ; Noster System AB Stockholm, Sweden) labeled with 1 mCi C^{14} with 150 mL of water. Breath samples were collected in a special cartridge 20 minutes after ingestion of the C^{14} urea

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capsule until the indicator membrane on the cartridge changed from yellow to orange. The cartridge was then placed into a scintillation counter that reported counts per minute (CPM). Samples with less than 25 CPM were considered negative, those with between 25 and 50 CPM doubtful, and those over 50 CPM positive for *H. pylori* infection. Patients who were positive for *H. pylori* by both histopathology and RUT were included in the study. Of the 380 patients, 237 patients who were positive for *H. pylori* by RUT and UBT were included in the study.

Treatment Regimen

A 14-day triple therapy consisting of clarithromycin (500 mg b.i.d), amoxicillin (1,000 mg b.i.d.), and lansoprazole (30 mg b.i.d.) was administered to the 237 patients who were positive for *H. pylori*. Eradication was defined as the absence of *H. pylori* according to C^{14} UBT testing six weeks after completing antimicrobial therapy. The only agents allowed for symptomatic relief were oral antacids ; patients were not allowed PPIs or other antisecretory drugs. Patients were divided into three groups using a computer-generated randomised sequence. This procedure resulted in patients with UBT values of 50-100 CPM being assigned to group I, those with values of 100-135 CPM to group II, and those with values of 135 CPM and above to group III.

Compliance and Safety

The frequency of adverse effects was assessed from diaries completed by the patients during treatment. Additionally, at each clinic visit patients were asked to report any adverse events since the previous visit. All adverse events were recorded, irrespective of their possible relationship to the study medication. Therapy compliance of each patient was assessed by counting returned pills.

Ethics and Consent

This study was approved by the Ethics Committee of Baskent University and all patients provided written informed consent in accordance with the Helsinki Declaration.

Statistical Analyses

Data were analyzed using SPSS (Statistical Package for the Social Sciences, version 13.0, SPSS, Chicago, IL, USA). Results are expressed as means ± standard deviation. Statistically significant differences between groups were assessed using Student's *t*- or χ^2 tests. P values below 0.05 were considered statistically significant in all analyses. Per- protocol (PP), ITT eradication rates, and 95% confidence intervals (CIs) were calculated for both treatments. For the PP analysis, only patients who had taken all of their medications for the prescribed therapy duration were evaluated. For the ITT analysis, all patients were included, including those who were non-compliant or did not complete the full course of therapy.

Results

The study cohort consisted of three groups, each of 79. Group I comprised patients with UBT values ≤ 100 CPM (42 women and 37 men ; mean age 42.1 ± 12 years), Group II patients with UBT values between 100 CPM and 135 CPM (47 women and 32 men ; mean age 44 ± 12 years), and Group III patients with UBT values > 135 CPM 49 women and 30 men ; mean age 39 ± 11.4 years). Patient age, sex, and endoscopic findings did not differ significantly between groups ($p > 0.05$). Treatment was not completed by three patients in Group I (because of severe nausea/vomiting and abdominal pain), four patients in Group II (two because of severe nausea and vomiting and two because of loss to follow-up), and two in Group III (one because of severe nausea and vomiting and because of loss to follow-up). Drug compliance was acceptable, compliance rates for Groups I, II, and III being 96%, 97%, and 98%, respectively.

H. pylori eradication rates according to ITT and PP analyses were 128/237 (54%, 95% CI : 46.4-66.5) and 128/228 (56.1%, 95% CI : 47.3-67) for all patients. *H. pylori* eradication rates according to ITT and PP analyses were 62/79 (79.1% 95% CI : 65.1-86.7) and 62/76 (81.5%, 95% CI : 70.4-86.2) for group I ; 36/79 (45.6% 95% CI : 34-58) and 36/75 (48% 95% CI : 36-61) for group II ; and 30/79 (37.9% 95% CI : 26-51) and 30/77 (38.9% 95% CI : 27-52) for group III, respectively. The differences between groups in the success of triple eradication treatment were statistically significant. UBT values were negatively associated with treatment success rate ($p < 0.05$). Eradication rates and 95% CIs of treatment differences are summarized in Figure 1.

Patients in Groups I, II, and III (14, 36, and 42, respectively) who failed triple therapy were retreated with quadruple therapy. One patient in Group I, two in Group II, and two in Group III were unable to continue treatment due to adverse effects such as diarrhea and taste disorders. *H. pylori* eradication rates according to ITT and PP analyses were 71.7% (95% CI : 63.4-84.2) and 75.8% (95% CI : 66.4-87) for all patients. The quadruple therapy was successful in nine patients in Group I (ITT : 64.2, PP : 69.2), 25 in Group II (ITT : 69.4, PP : 73.5), and 32 in Group III (ITT : 76 ; PP : 80). There were no statistically significant differences between the groups in terms of quadruple therapy success rate ($p = 0.434$).

Discussion

In the present study, we observed that successful treatment with the standard 14-day regimen was strongly negatively associated with UBT values ($p = 0.00$). However, there were no significant differences between groups ($p = 0.434$) in rate of eradication by quadruple treatment in patients whose first-line treatment had been

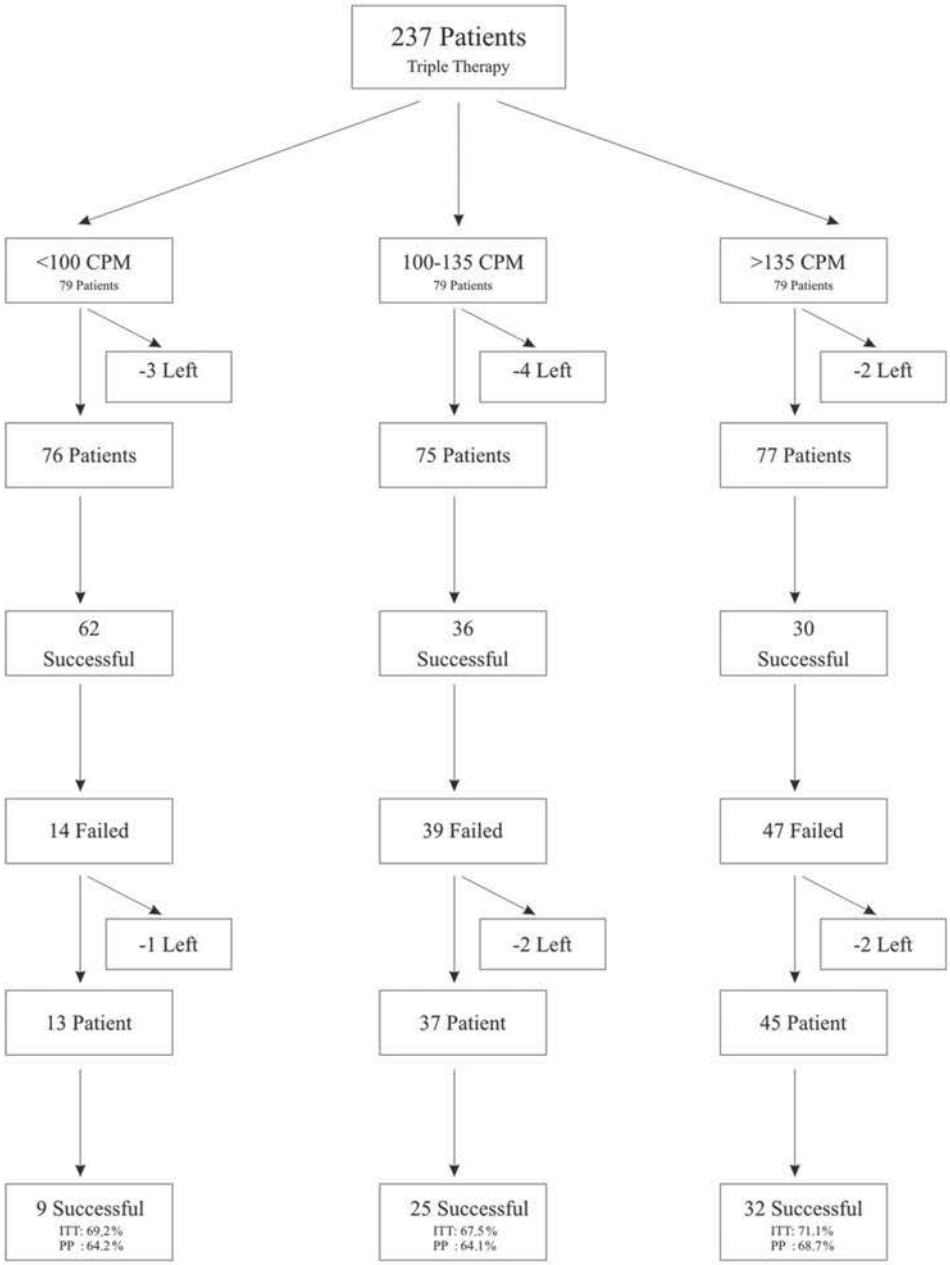

Fig. 1. — Schema depicting patient selection

unsuccessful. There was no relationship between UBT values and gastric pathologies (p = 0.751).

H. pylori is a gram-negative, microaerophilic bacterium that was first detected and cultured from the stomach in 1983. Nearly half of the world's population is infected by this microorganism which, since its identification, has been implicated in the pathogenesis of various gastric and extra-gastric diseases (2).

A number of studies have shown that *H. pylori* is involved in the pathogenesis of gastritis, atrophic gastritis, intestinal metaplasia, peptic ulcers, mucosa-associated lymphoid tissue lymphomas (MALTomas), and gastric cancer. Patients with peptic ulcer disease, atrophic gastritis, MALTomas ; those who have undergone gastric cancer surgery ; and first-degree relatives of patients with gastric cancer should receive treatment for *H. pylori* infection (2,4,6).

According to the Maastricht IV/Florence Consensus Report of March 2011, 7- or 14-day treatments consisting of PPIs, clarithromycin, amoxicillin or metronidazole are the recommended primary treatments for *H. pylori* infections. However, in countries with high rates of clarithromycin resistance, quadruple therapy consisting of tetracycline, metronidazole, PPI, and bismuth may be a preferable first option (6). Standard triple therapy has been associated with reduced *H. pylori* treatment success rates in our country (Turkey) and worldwide ; the eradication rate has decreased from 90% to 50% because of

Table 1. — **Relevant patient variables and endoscopic findings by UBT values**

UBT values	Age (years)	Male to female ratio	Pangastritis	Erosive gastritis	Duodenal ulcer	Gastric ulcer
< 100 CPM	42 ± 12	37/42	46	8	9	*4*
100–135 CPM	44 ± 12	32/47	40	11	11	*5*
> 135 CPM	39 ± 11	30/49	48	5	9	*5*

bacterial resistance (7). Therefore, sequential and new alternative therapies consisting of antibiotics like levofloxacin, rifabutin, and furazolidone have been developed (4-8).

The causes of eradication failure have been the subject of many studies. Insufficient patient compliance with prescribed therapy, including inappropriate doses and treatment duration, is a fundamental cause of eradication failure. The combination of a large number of drugs and side effects that reduce the quality of life (e.g. ; metallic taste in mouth, nausea, vomiting, and diarrhea) and the long duration of therapy may reduce patient treatment compliance (9).

The second main reason for eradication failure is antibiotic resistance : clarithromycin resistance increases failure rates. Development of resistance has varied from country to country and even regionally. Clarithromycin should not be used in countries with resistance rates over 20% unless based on antibiotic sensitivity test results (7, 8,10).

However, a number of studies have investigated other reasons for reduced treatment success rates, including sex, smoking, bacterial load, presence of dyspeptic symptoms, and gastric pathology (11). Since 1992 many similar studies in other countries have assessed the impact of bacterial load on eradication rates, with different reported results. In several of these studies, bacterial density has been evaluated histopathologically and by UBT (12-16). For histopathological evaluation, upper gastrointestinal endoscopies were performed and biopsies taken from the corpus and antrum. The degree of gastritis in these samples was determined using Sydney Classification Multiple studies in which C^{13} and C^{14} UBTs were performed on the same patient groups have shown UBT numerical values to be correlated with gastritis histopathology (17-21).

Comparisons of these two tests have been based on results of histopathological examination, which requires an endoscopy – an invasive examination – and multiple biopsies from the corpus and antrum. An insufficient number of biopsies may fail to reflect the bacterial load ; moreover, pathological evaluations can vary with different examiners. UBT can also be problematic. False negativities can occur in patients with rapid gastric emptying, caused by previous surgery. Standardization difficulties make it important to collect breath samples at an appropriate and reported interval after administration of the urea substrate. UBT is an easy, noninvasive, and highly sensitive test under appropriate conditions (22). In this study, we selected C^{14} UBT to determine the density of *H. pylori* in the stomach because of its cost-effectiveness ; this test has been shown to adequately assess gastric *H. pylori* density in previous studies (17-21). Because ongoing studies have shown that *H. pylori* densities vary between patients ; the effects of density on eradication rates have been studied.

In 1998, Perri *et al*. reported administering a triple therapy of clarithromycin, amoxicillin, and omeprazole for 1 week ; their eradication rate was negatively related to UBT values (82%, 56%, and 17%, respectively) (9). In a study conducted in 1996, amoxicillin (500 mg t.i.d.), metronidazole (500 mg t.i.d), and bismuth subcitrate (120 mg q.i.d.) were administered to 136 patients with bleeding duodenal ulcers who were positive for *H. pylori* infections and the bacterial load during the first endoscopy graded histopathologically. The eradication rate was 76.4% and the histopathological bacterial load was high in the group with eradication failure (23). In 2001, Maconi *et al*., similar to other studies, found that high intragastric load was negatively associated with achievement of eradication. This was statistically significant especially for a 1-week treatment regimen ; a 2-week treatment was relatively less successful in the group with high bacterial loads ; however, this difference was not statistically significant (10).

In a study of 119 patients by Kawai *et al*. in 2008, the subjects underwent upper gastrointestinal endoscopy, UBT, *H. pylori* culture, and antibiograms. These authors also reported lower rates of treatment success in patients with higher UBT values ; they attributed the low treatment success rate in patients with high UBT values to clarithromycin resistance (24). However, in a study by Perri *et al*. involving 162 patients, clarithromycin resistance was similar in groups with high and low UBT values (25). In a 2008 study, Zullo *et al*. reported no significant differences in UBT values between clarithromycin-sensitive and -resistant strains (11). In another study conducted in 2006 by Gisbert *et al*., no relationship was observed between the success of primary and salvage treatments and UBT values. Clarithromycin resistance was reported to be the main factor for treatment failure (26).

It is worth noting that antimicrobial sensitivity is maintained at neutral pH *in vitro*. Furthermore, sensitivity assessment is performed for single antibiotics ; the synergistic effects of multiple antibiotics cannot currently be measured.

In our study, we found that the success rate of the standard 14-day regimen decreased with increasing UBT values ($p = 0.001$). However, when quadruple treatment

was administered to patients in whom eradication had not been achieved, there were no significant differences in success rates between the groups (p = 0.434). Following the standard treatment regimen, UBT values decreased in individuals with treatment failure, but not enough to qualify as treatment success (p = 0.001).

Our aim in this study was to predict treatment effectiveness based on outpatient UBT values. Because the study protocol specified a standard treatment for all patients, antibiograms were not performed.

The relationship between UBT values and gastric pathology has also been examined. Alam *et al*. reported in 1992 that high bacterial densities were associated with development of acute and chronic inflammation and duodenal ulcers (27). However, in 2005, Tseng *et al*. studied 564 patients and found no relationship between high UBT values and incidence of gastric cancer, peptic ulcer, and gastritis (28). In the current study, we also found no relationship between UBT values and gastric pathology.

Numerous previous studies have found that patient compliance with treatment and antibiotic resistance are important factors that determine treatment success. However, other factors such as bacterial load may also play a role in achieving responses to treatment : our study and others have found that high bacterial loads are associated with lower treatment success rates. Future research could further investigate the impact of bacterial density on treatment outcomes by assessing the effectiveness of primary and secondary treatment regimens according to UBT values.

Conclusions

Additional research is necessary to confirm our finding that UBT, a noninvasive test, can predict treatment success and identify alternative drug combinations for eradicating *H. pylori* in patients with high bacterial density. This would facilitate sparing patients from unpleasant and ineffective treatments.

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