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Quadruple Rescue Therapy after First and Second Line Failure for *Helicobacter pylori* Treatment: Comparison between Two Tetracycline-Based Regimens

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ABSTRACT

Background & Aims: Antibiotic resistance is the main reason for failure of *Helicobacter pylori* (*H. pylori*) treatment. Currently, guidelines recommend a treatment guided by antimicrobial susceptibility testing after two failures. However, microbial culture is not feasible everywhere, and the limited number of effective antibiotics against the bacterium narrows the options; thus a rescue therapy combining antibiotics with a low resistance may be fitting.

Methods: Patients who have failed a first-line treatment (either prolonged triple or sequential regimens) and, successively, a levofloxacin-based triple therapy were considered for the study. Subjects underwent urea breath test (UBT), stool antigen test (ST) and endoscopy/histology to confirm the diagnosis. Cytopenia and impaired liver and kidney function were exclusion criteria. Fifty-four subjects were randomized 1:1 to two regimens: RMB Rabeprazole/Rifabutin/Minocycline/Bismuth sub-citrate or MTB Rabeprazole/Tinidazole/Minocycline/Bismuth sub-citrate both for 10 days. The results were checked 6 weeks after the end of therapy with ST/UBT plus endoscopy when indicated.

Results: RMB eradicated the bacterium in 21 patients. Two subjects dropped out. The eradication rate was 77.7% (CI 62.0-93.4%) at intention-to-treat and 84.0% (CI 69.6-98.4%) at per-protocol analysis. MTB was successful in 14 patients (51.9%, CI 33.1-70.7%). No patient withdrew from the treatment for adverse events. Drug-related side effects were reported only in 3 subjects, but in all cases the treatment was carried on.

Conclusions: The association minocycline/rifabutin seems to have a synergic effect and a good therapeutic outcome in patients who have failed at least two previous regimens, although a trial on a large population is needed.

Key words: *Helicobacter pylori* – antibiotic resistance – rescue therapy – rifabutin – minocycline.

INTRODUCTION

The increasing antibiotic resistance is the main reason for *Helicobacter pylori* (*H. pylori*) treatment failure [1]. After an unsuccessful first-line regimen (usually triple or sequential/concomitant/hybrid therapies), guidelines recommend a triple therapy with levofloxacin or a bismuth/tetracycline/metronidazole quadruple regimen [2]. When a second-line treatment fails, the possible options are very limited [3]. In this case, Maastricht-IV Consensus advises a treatment guided by antimicrobial susceptibility testing, i.e. culture [2]. However, despite the fact that this technique may permit the achievement of good eradication rates [4], culture is not feasible everywhere and difficult to perform even in expert hands. Moreover, the limited number of effective antibiotics against the bacterium narrows the field of options. On the other hand, Italian guidelines [5] suggest a rifabutin based triple therapy as a third line. The possibility of using an empiric third-line regimen when culture is not available or most antibiotics have failed is essential, and rifabutin has been investigated in many studies, with a success rate ranging from 44 to 95% [6]. In a recent meta-analysis, the mean percentage of success of a rifabutin-based triple therapy (rifabutin, amoxicillin and a proton pump inhibitor - PPI) was 66% when used in third line [6].

Tetracycline is a class of antibiotics that is largely used in second and third line therapy [7], usually in association with bismuth and metronidazole, the so called "quadruple therapy". However, in Italy the commercial package of tetracycline contains tablets with a dose of 250 mg. Therefore, for an effective treatment, the patient is requested to assume 8 pills/day of tetracycline in addition to the other drugs with a total number of 16 tablets/day for at least 10 days. It may be argued that relevant consequences on the compliance are very common. Minocycline belongs to the tetracycline class. It is used at a dose of 100 mg (one tablet) twice daily, and this could push the patient to a complete adherence to the therapeutic regimen.

Therefore, herein we describe a preliminary experience with a novel quadruple regimen containing rifabutin, minocycline and bismuth (RMB) as a rescue therapy in patients who had failed at least two previous regimens, compared to a classical quadruple therapy which comprised minocycline, tinidazole and bismuth (MTB).

PATIENTS AND METHODS

Patients who failed a first-line regimen (either prolonged triple or sequential therapy) and successively a levofloxacin-based triple therapy were considered for the study. Before starting the treatment, all eligible patients underwent a full blood count and evaluation of renal (creatinine, blood urea nitrogen) and hepatic function (transaminases, bilirubin, alkaline phosphatase and gamma-glutamyl transpeptidase). All subjects with cytopenia (defined as white blood cells < 4,000/μL, hemoglobin < 10g/dL and platelets < 150,000/μL), impaired renal function and liver cytolysis/cholestasis were excluded. All subjects had *H. pylori* infection diagnosed by the concordance of the urea breath test (UBT) and stool test antigen (ST). The diagnosis was confirmed by upper endoscopy with histology before starting the therapy. The patients were randomized 1:1 to the rifabutin-based quadruple therapy (RMB) or to the modified conventional quadruple therapy (MTB). Both treatments lasted 10 days.

The RMB eradication regimen was given as follows: Rabeprazole 20mg b.i.d, Rifabutin 150 mg b.i.d, Minocycline 100 mg b.i.d and Bismuth sub-citrate 120 mg four times/day. The MTB regimen consisted of Rabeprazole 20 mg b.i.d, Tinidazole 500 mg b.i.d, Minocycline 100 mg b.i.d and Bismuth sub-citrate 120mg four times/day. The patients had PPI 30 minutes before meals and antibiotics 30 minutes after meals. Bismuth administration was independent by meals. Informed consent was obtained from each patient.

The result of the eradication treatment was checked by UBT and ST 6 weeks after its cessation, and therapy was considered successful only when both tests confirmed *H. pylori* negativity. When necessary (ulcer or low grade MALT lymphoma) a second endoscopy/histology was performed. At the 5th day and the end of therapy, the side effects were investigated by a personal interview.

The $\chi 2$ test or the Fisher's exact probability test were used, as appropriate, to compare the percentages and nominal variables. For continuous variables, differences between the patients in the two treatment arms were compared using the Student *t*-test for unpaired samples. The eradication rates were expressed both as intention-to-treat (ITT) and per-protocol (PP) analysis, and 95% confidence interval (95% CI) was provided. The statistical analysis was performed using the statistical software GraphPad Prism version 5.00 for Windows, GraphPad Software, San Diego California USA.

RESULTS

Fifty-four patients (32 females, 22 males) fulfilled inclusion criteria and received the rescue regimen in the period July 2013-July 2014. The mean age was 53.3 years (range 23-80, standard deviation 13.3, median 54). All patients were referred to endoscopy for long-lasting dyspeptic and/or ulcer-like symptoms. Endoscopy revealed peptic ulcer in 9 cases. Histology and immunohistochemistry revealed in three patients a low grade MALT lymphoma. Diffuse antral incomplete intestinal metaplasia was observed in 6 patients.

In the first line, 22 patients (40.7%) had failed sequential regimen, while 32 (59.3%) a 10-day triple therapy. All patients had successively failed a second line levofloxacin-amoxicillin 10-day triple therapy.

In the RMB group, 21 patients eradicated the bacterium. Two subjects dropped out, so no data regarding their therapy results are available. The eradication rate was 77.7% (CI 62.0-93.4%) at ITT and 84.0% (CI 69.6-98.4%) at PP analysis.

In the MTB group, 14 patients eradicated *H. pylori*. We did not record any drop out. The eradication rate was 51.9% (CI 33.1-70.7%) at both PP and ITT analysis.

H. pylori related disorders and therapeutic outcome of enrolled patients are described in Table I.

No patient withdrew from the treatment for adverse events. Main drug-related side effects were reported only in 3 subjects (5.6%): diarrhea in two cases (one in RMB and one in MTB) and severe asthenia in the other one (RMB). However, in both cases the treatment was not halted and in the first case a probiotic supplementation was given.

DISCUSSION

H. pylori is a well-known cause of gastritis, peptic ulcer and malignant diseases of the stomach [8, 9]. Since its eradication is able to avoid the evolution of gastric lesions into severe gastric malignancies, such as adenocarcinoma or lymphoma, effective eradication regimens are required. However, antibiotic resistances are a frequent cause of therapy failure; therefore, rescue therapies are of strong relevance. Rifabutin is an anti-tuberculosis agent used in patients affected by acquired immune-deficiency syndrome. Its use in *H. pylori* eradication regimens is indicated today only for the rescue therapy given the low rates of antibiotic resistance [10], the high economic costs and the potential severe side effects, such as neutropenia or anemia [11]. A recent meta-analysis showed a mean percentage of success of a third-line rifabutin-based triple therapy (rifabutin, amoxicillin and a PPI) of 66% [6].

The second antibiotic used in our regimens, minocycline, belongs to the tetracycline drug class, which has been associated with low rates of antibiotic resistance in Italy [12]. A single trial with minocycline and amoxicillin is available and

Table I. Demographic, clinical, endoscopic and histological characteristics of enrolled patients and the related therapeutic outcome.

	RMB		MTB		p*	p**
Features	Number (%)	Eradicated patients n(%)	Number (%)	Eradicated patients n (%)		
Total	27	21 (77.7)	27	14 (51.9%)	-	0.02
Age (mean ± SD)	53.8 ± 12.6	51.6 ± 12.1	52.6 ± 14.5	50.4 ± 13.3	0.45	0.43
Sex (female)	15 (55.5)	12 (80.0)	14 (51.9)	10 (71.4)	1	0.68
Dyspepsia	17 (62.9)	13 (76.5)	19 (70.4)	10 (52.6)	0.77	0.18
Peptic ulcer	5 (18.5)	5 (100)	4 (14.8)	2 (50)	1	0.17
Duodenal	4(14.8)	4 (100)	4 (14.8)	2 (50)		
Gastric	1 (3.7)	1 (100)	0 (0)	-		
Antral diffuse intestinal metaplasia	3 (11.1)	1 (33.3)	3 (11.1)	1 (33.3)	1	1
Low grade MALT lymphoma	2 (7.4)	2 (100)	1 (3.7)	1 (100)	1	1
Side effects	2 (7.4)	-	1 (3.7)	-	1	-

* *t*-test or Fisher exact text/χ2 test where needed. The comparision was performed between the total number of patients enrolled in each group.
** *t*-test or Fisher exact text/χ2 test where needed. The comparision was performed between the number of patients who eradicated *H. pylori* between the two groups.
RMB = rifabutin, minocycline, bismuth; MTB = minocycline, tinidazole, bismuth.

shows a disappointing effectiveness (38.5%) [13]. However, the case of a multi-resistant *H. pylori* strain successfully treated with minocycline/amoxicillin/bismuth has been described [14]. Tetracycline has been employed in third line regimens, enclosed in classical bismuth-containing quadruple therapy, with results varying from 65% in a recent Spanish trial [15] to 36% in another study conducted in the year 2000 in the same country [16].

The data of the present preliminary report demonstrated that the association minocycline/rifabutin has a satisfactory eradication rate (PP: 84%; ITT: 77.7%) when compared to previous studies using tetracycline or rifabutin alone. This effect could be explained by two main reasons: i) two different antibiotics with low resistance rates were combined, and ii) these antibiotics had not been used in precedent eradication regimens in our patients. On the other hand, a conventional bismuth based quadruple therapy, although with minocycline instead of tetracycline, did not reach a satisfactory eradication rate (51.9%). The use of tinidazole, which was already employed in first line by some patients, could explain this result. Nevertheless, the different outcomes of the two regimens could not have been affected by the previous tinidazole assumption, as clearly shown in Table I.

Based on this hypothesis, the association rifabutin-minocycline could have a synergic action and fulfills its maximal potential therapeutic effect when compared with other third line schemes. The possibility of the combination of these two drugs is secondary to the approval of local pharmaceutical agencies. Both drugs were approved by the Food and Drug Administration (FDA) [17] and by the European Medicines Agency (EMA), so they are available both in Europe and the United States [18]. Studies related to their use in Asian and African countries are also available [19-22].

Of relevance, *H. pylori* eradication was obtained in two patients with low grade MALT lymphoma.

In our study, no patient developed severe drug reactions. Possible explanations may be : i) the short duration of the treatment (rifabutin side effects are common in a 14-day lasting regimen [23] or when the dose is 600mg/day for mycobacterial infection [24]), and ii) the exclusion criteria that did not allow the enrolling of patients at high risk of severe reactions to both antibiotics.

Despite the encouraging results, this is a preliminary study and the present data come from a small-size group of subjects. This last aspect is due to the rarity of multiple failures, since the patients were recruited during a period of one year. In conclusion, further investigations are required to confirm our results and to definitively ascertain the effectiveness and safety of the combination rifabutin-minocycline in *H. pylori* multi-resistant infection.

Conflicts of interests: none declared.

Authors' contribution: E.I. and A.D.L. designed the study. G.L., A.A., A.G. and V.D.F. collected the data. M.P. performed the endoscopies. F.G. and G.L. extracted and performed statistical analysis. E.I. and G.L. wrote the manuscript. All authors revised the final version before approval.

REFERENCES

1. Ierardi E, Giorgio F, Losurdo G, Di Leo A, Principi M. How antibiotic resistances could change Helicobacter pylori treatment: A matter of geography? World J Gastroenterol 2013;19:8168-8180.
2. Malfertheiner P, Megraud F, O'Morain CA, et al. Management of Helicobacter pylori infection--the Maastricht IV/ Florence Consensus Report. Gut 2012;61:646-664.
3. Song M, Ang TL. Second and third line treatment options for Helicobacter pylori eradication. World J Gastroenterol 2014;20:1517-1528.
4. Fiorini G, Vakil N, Zullo A, et al. Culture-based selection therapy for patients who did not respond to previous treatment for Helicobacter pylori infection. Clin Gastroenterol Hepatol 2013;11:507-510.

5. Caselli M, Zullo A, Maconi G, et al. „Cervia II Working Group Report 2006": guidelines on diagnosis and treatment of Helicobacter pylori infection in Italy. Dig Liver Dis 2007;39:782-789.
6. Gisbert JP, Calvet X. Review article: rifabutin in the treatment of refractory Helicobacter pylori infection. Aliment Pharmacol Ther 2012;35:209-221.
7. Cianci R, Montalto M, Pandolfi F, Gasbarrini GB, Cammarota G. Third-line rescue therapy for Helicobacter pylori infection. World J Gastroenterol 2006;12:2313-2319.
8. Ierardi E, Goni E, Losurdo G, Di Mario F. Helicobacter pylori and nonmalignant diseases. Helicobacter 2014;19 Suppl 1:27-31.
9. Venerito M, Nardone G, Selgrad M, Rokkas T, Malfertheiner P. Gastric cancer--epidemiologic and clinical aspects. Helicobacter 2014;19 Suppl 1:32-37.
10. O'Connor A, Taneike I, Nami A, et al. Helicobacter pylori resistance rates for levofloxacin, tetracycline and rifabutin among Irish isolates at a reference centre. Ir J Med Sci 2013;182:693-695.
11. Brogden RN, Fitton A. Rifabutin. A review of its antimicrobial activity, pharmacokinetic properties and therapeutic efficacy. Drugs 1994;47:983-1009.
12. De Francesco V, Giorgio F, Hassan C, et al. Worldwide *H. pylori* antibiotic resistance: a systematic review. J Gastrointestin Liver Dis 2010;19:409-414.
13. Murakami K, Sato R, Okimoto T, et al. Effectiveness of minocycline-based triple therapy for eradication of Helicobacter pylori infection. J Gastroenterol Hepatol 2006;21:262-267.
14. Nakajima S, Inoue H, Inoue T, Maruoka Y. Multiple-antibiotic-resistant Helicobacter pylori infection eradicated with a tailor-made quadruple therapy. J Gastroenterol Hepatol 2012;27 Suppl 3:108-111.
15. Gisbert JP, Perez-Aisa A, Rodrigo L, et al Third-line rescue therapy with bismuth-containing quadruple regimen after failure of two treatments (with clarithromycin and levofloxacin) for *H. pylori* infection. Dig Dis Sci 2014;59:383-389.
16. Gomollón F, Sicilia B, Ducóns JA, Sierra E, Revillo MJ, Ferrero M. Third line treatment for Helicobacter pylori: a prospective, culture-guided study in peptic ulcer patients. Aliment Pharmacol Ther 2000;14:1335-1338.
17. U S Food and Drug Administration. www.fda.gov/
18. European Medicines Agency. www.ema.europa.eu/
19. Seo JH, Jun JS, Yeom JS, et al. Changing pattern of antibiotic resistance of Helicobacter pylori in children during 20 years in Jinju, South Korea. Pediatr Int 2013;55:332-336.
20. Tan Y, Hu Z, Zhao Y, et al. The beginning of the rpoB gene in addition to the rifampin resistance determination region might be needed for identifying rifampin/rifabutin cross-resistance in multidrug-resistant Mycobacterium tuberculosis isolates from Southern China. J Clin Microbiol 2012;50:81-85.
21. Miyazaki M, Nagata N, Matsuo K, Takata T, Tanihara S, Kamimura H. Trends and antimicrobial susceptibilities of clinical methicillin-resistant Staphylococcus aureus isolates in Fukuoka University Chikushi Hospital 2008-2012. Yakugaku Zasshi 2014;134:269-276.
22. Kanj SS, Whitelaw A, Dowzicky MJ. In vitro activity of tigecycline and comparators against Gram-positive and Gram-negative isolates collected from the Middle East and Africa between 2004 and 2011. Int J Antimicrob Agents 2014;43:170-178.
23. Miehlke S, Hansky K, Schneider-Brachert W, et al. Randomized trial of rifabutin-based triple therapy and high-dose dual therapy for rescue treatment of Helicobacter pylori resistant to both metronidazole and clarithromycin. Aliment Pharmacol Ther 2006;24:395-403.
24. Griffith DE, Brown BA, Girard WM, Wallace RJ Jr. Adverse events associated with high-dose rifabutin in macrolide-containing regimens for the treatment of Mycobacterium avium complex lung disease. Clin Infect Dis 1995;21:594-598.