



# The Effect of Probiotic Plus Prebiotic Supplementation on the Tolerance and Efficacy of *Helicobacter Pylori* Eradication Quadruple Therapy: a Randomized Prospective Double Blind Controlled Trial

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## ABSTRACT

### BACKGROUND

Standard anti-*Helicobacter pylori* (*H. pylori*) treatment fails in the eradication of the organism in almost 10-35% of the patients and has different side effects. Recent studies have proposed that probiotic supplementations with or without prebiotic may improve the eradication rate and diminish the side effects, although it is still a controversial issue. We aimed to investigate the effect of probiotic with prebiotic supplementation on the eradication rate and side effects of anti *H. pylori* quadruple therapy.

### METHODS

76 patients with a positive biopsy specimen for *H. pylori* were enrolled. They were randomized to receive quadruple therapy of bismuth, clarithromycin, amoxicillin, and omeprazole for 14 days and also the synbiotic or the placebo. We asked them to answer study questionnaires at the beginning and during the treatment. Finally, urea breath test was done 8 weeks after the treatment.

### RESULTS

The eradication rate was significantly better in the synbiotic group by intention-to-treat analysis ( $p < 0.05$ ). Treatment side effects such as diarrhea, nausea, vomiting, epigastric pain, flatulence, constipation, and taste abnormality were similar in both groups but anorexia was significantly better in the synbiotic group ( $p < 0.05$ ).

### CONCLUSION

The eradication rate was significantly better in the synbiotic group by intention-to-treat analysis ( $p < 0.05$ ). Treatment side effects such as diarrhea, nausea, vomiting, epigastric pain, flatulence, but could improve the eradication by augmenting the treatment tolerance and compliance.

### KEYWORDS:

*H. pylori* eradication; Probiotic; Prebiotic; Synbiotic; Quadruple therapy; RCT

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## INTRODUCTION

*Helicobacter pylori* (*H. Pylori*), as a common and significant health problem plays a major contributory role in the pathogenesis of chronic gastritis, peptic ulcer disease, gastric mucosa associated lymphoid tissue (MALT) lymphoma, and adenocarcinoma.<sup>1-4</sup> Currently, there are various therapeutic regimens; although the most successful one cannot eradicate the bacteria in at least 10% of the affected patients.<sup>5</sup> Quadruple therapy is appropriate as initial therapy in areas where the prevalence of resistance to clarithromycin or metronidazole is >20%, or in patients with recent or repeated exposure to clarithromycin or metronidazole.<sup>6</sup> A meta-analysis of 93 studies showed that in populations with either clarithromycin or metronidazole resistance, quadruple therapy resulted in a higher rate of eradication than triple therapy.<sup>7</sup> Beside the eradication rate of treatment regimen, undesirable side effects, and patients' poor compliance play considerable role in the outcome of treatment. A considerable number of patients experience undesirable side effects. The most common complaints during antimicrobial treatments are diarrhea, epigastric pain, nausea, and bloating.<sup>8</sup>

Probiotic bacteria, defined as living microorganisms that have beneficial health effects to the host, have been successfully used in the treatment or prevention of several gastrointestinal (GI) disorders, such as antibiotic-associated diarrhea, inflammatory bowel disease, and irritable bowel syndrome.<sup>9-10</sup> They may be useful adjuncts to improve tolerability and compliance of anti *H. pylori* treatment.<sup>11-12</sup> Because some probiotics have antimicrobial effects, they have been proposed as a treatment option for *H. pylori* infection but they should not be considered as a substitute for standard antibiotic treatments.<sup>13</sup> Several studies have reported that certain probiotic bacteria, such as *Lactobacillus* spp., exhibit inhibitory activity against *H. pylori* in vitro and in vivo.<sup>14-15</sup>

This study was designed to investigate the effect of probiotic-prebiotic supplementation on the eradication rate of anti *H. pylori* quadruple therapy, treatment side effects, and the improvement of dyspepsia after treatment. We aimed to investigate the effect of this supplementation, which contains *Lactobacillus casei*, *Lactobacillus rhamnosus*, *Streptococcus thermophilus*, *Bifidobacterium breve*, *Lactobacillus acidophilus*, *Bifidobacterium longum*, and *Lactobacillus bulgarius* on the eradication

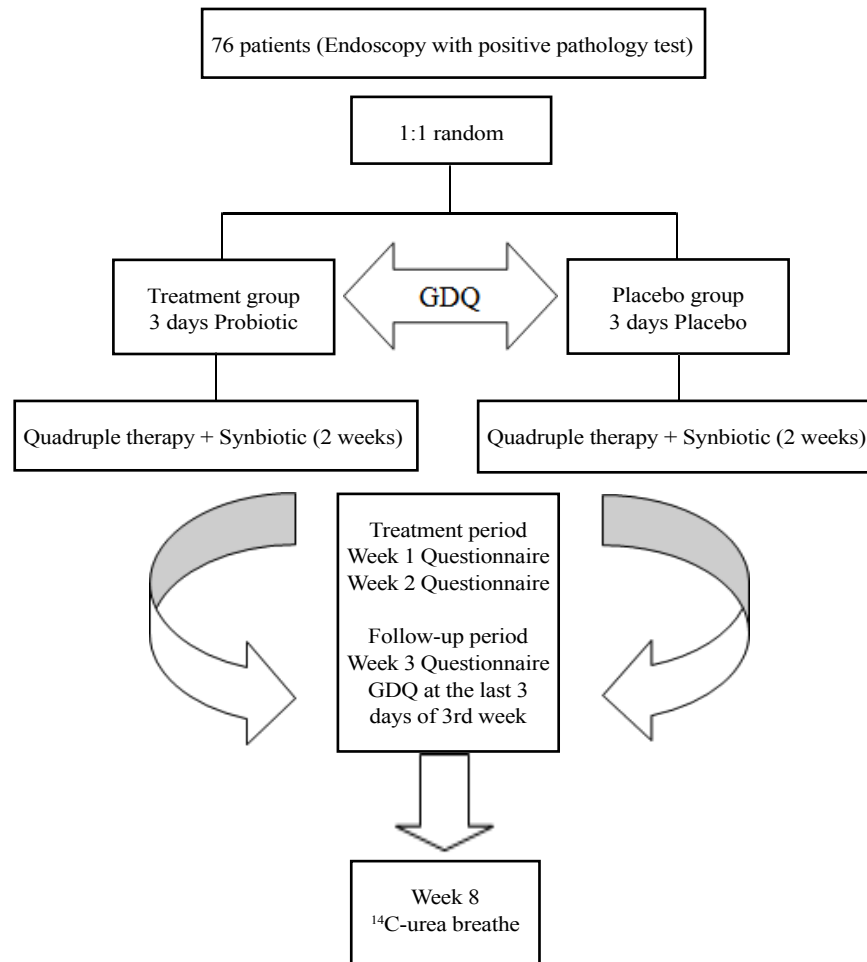
rate of anti *H. pylori* quadruple therapy, its side effects, and patients' compliance.

## MATERIALS AND METHODS

### Study design

This study was a prospective randomized double blind placebo controlled study. 76 patients with a positive biopsy specimen for *H. pylori* infection were enrolled from May 2011 to March 2012 in Rasht and Anzali, Iran (male/female: 35/41, mean age: 43.5±13.31). The patients who received 14 days of quadruple therapy consisted of bismuth subcitrate four times a day, and amoxicillin 1 g, clarithromycin 500 mg, and omeprazole 20 mg, all twice a day were randomly assigned to receive synbiotic (Protexin Balance®) or placebo 2 capsules before the lunch during the treatment and also three days earlier. We asked them to begin the synbiotic (treatment group, n=38) or placebo (control group, n=38) three days before initiation of the main treatment to make a better chance for colonization of probiotic bacteria. The patients were divided into two groups by random allocation. The placebo was manufactured by Sobhan Daru Company and there was no difference in shape, color, weight, and taste between the two products. Boxes containing placebo or probiotic were identical in shape, size, and color and did not have any trade mark and contained the same number of drugs. All the patients were followed up for 4-8 weeks after the end of treatment and to evaluate the patients' compliance, pill counting was done. 8 weeks after the end of treatment, the eradication was confirmed by urea breath test (figure 1).

The inclusion criterion was the presence of *H. pylori* in the histopathological examination of biopsy specimen obtained during upper GI endoscopy. The gastric body greater curvature and prepyloric antrum were the preferred sites of biopsy sampling and a large channel biopsy forceps was used. Both hematoxylin and eosin (H&E) and Giemsa staining were used and all the samples were evaluated by an expert histopathologist. A rapid urease test was also performed for a better confirmation. The exclusion criteria were GI bleeding, malignancy, inflammatory bowel disease, prior gastrectomy, recent history of *H. pylori* eradication, immunodeficiency, and the use of antibiotic or probiotics in the weeks before and during the study. Dyspeptic symptoms were



**Fig.1:** Study design and patients follow-up

recorded daily before the initiation of treatment in three days and also in the last three days of the third week after the initiation of treatment using modified version of the Glasgow Dyspepsia Questionnaire (GDQ).<sup>16</sup> The dyspepsia score were ranged from 0-12. The assessment of side effects was done at the end of the 1st, 2nd, and 3rd week after initiation of quadruple therapy by the modified version of the questionnaire by de Boer and colleagues.<sup>17</sup> The subjects were asked to report any side effects during and after therapy such as taste disturbance, nausea, vomiting, epigastric pain, bloating, diarrhea, and constipation. They were also asked to grade each side effect according to severity as: mild (effect observed, but could be disregarded), moderate (effect sometimes interfered with daily activities), or severe (effect continuously interfered with daily activities). In order to obtain the highest compliance in registering any treatment related

side effects, verbal explanations and printed instructions were both given to patients on how and when to fill in the questionnaires. This study was approved by the Ethics Committee of Guilan University of Medical Sciences and registered on the Iranian Registry of Clinical Trials website (IRCT registration no. IRCT201011135174N1). A written informed consent was obtained from each patient before the initiation of the study.

### ***H. pylori* Eradication**

*H. pylori* eradication was evaluated by a <sup>14</sup>C-urea breath test performed 8 weeks after the end of treatment. During this period the patients did not take any antibiotics or acid reducing drugs. After taking urea labeled with <sup>14</sup>C by the patients, *H. pylori* infection could be diagnosed by using Helitracer HUBT-20 (Headway Bio-Sci&Tech Co, LTD, Shenzhen Zhonghe, China, sen-

sitivity 95%, specificity 95-100%) to detect the volume of [<sup>14</sup>C] in exhaled breath due to action of urease generated by *H.pylori* that dissolve urea into <sup>14</sup>CO<sub>2</sub>. The entire test is finished in 25 minutes.

### Synbiotic Supplementation

Protexin Balance provides a complex blend of 7 strains of friendly bacteria and prebiotic, to help maintain healthy digestive and immune systems in all the family packed in easy to swallow capsules. Protexin probiotics are *Lactobacillus casei*, *Lactobacillus rhamnosus*, *Streptococcus thermophilus*, *Bifidobacterium breve*, *Lactobacillus acidophilus*, *Bifidobacterium longum*, *Lactobacillus bulgaricus*, (TVC: 200 million CFU TVC: 2×10<sup>8</sup> CFU) and also FOS (fructooligosaccharide-prebiotic), magnesium stearate (source: mineral and vegetable), and vegetable capsule (hydroxypropyl methyl cellulose). These processes were accelerated by the oral administration of lactulose, a non-digestible disaccharide used as substrate by the *Lactobacillus*. Protexin is manufactured in state-of-the-art facility in Somerset, UK. (Probiotics International Ltd Somerset, TA13 5JH, UK Registered Company No: 1122942) {available at <http://www.protexin.com>}

### Statistical analysis

Eradication rates were determined for both groups on a per-protocol (PP) and intention-to-treat (ITT) basis. For ITT analysis, all the enrolled patients were included, but for PP analysis, those who had taken less than 80% of any of the prescribed drugs, had dropped out due to severe adverse events, had not undergone final <sup>14</sup>C-urea breath testing, or those who had violated the study protocol were excluded. The side effects, dyspepsia improvement, and the eradication rates were compared by paired Student t test, Mann-Whitney U- test, Chi-square or Fischer's exact test. Results were considered significant when p values were less than 0.05 and the analysis was done by SPSS software version 18 for windows.

## RESULTS

### Patients/population

76 patients (35 men, 41 women) were randomly allocated into placebo group (17 men, 21 women) and

probiotic group (18 men, 20 women). In the placebo group, three patients could not complete the pre-treatment regimen i.e. the short course of placebo before the main therapy, and thereby were excluded from the study (one due to severe infectious diarrhea at the beginning of treatment, the other due to fascioliasis and hospital admission, and the last one due to the possibility of pregnancy). 11 patients in the placebo group were also excluded due to follow-up loss and non-compliance. Of the 14 excluded patients, 12 had non-ulcer dyspepsia (NUD) and 2 had duodenal ulcer (DU). In placebo group, of the 24 patients enrolled in PP analysis, 19 (79.16%) had NUD and 5 (20.84%) had DU with no case of gastric ulcer (GU). Out of 38 patients enrolled in ITT analysis, 31 (81.57%) had NUD and 7 (18.43%) had DU with no case of GU. There was no statistical difference in underlying diseases between the two groups (table 1). In the synbiotic group, all the 38 patients completed treatment regimen with favorable compliance and there was no missed case.

### H. pylori eradication rate

The overall eradication rate in patients who completed the treatment regimen was 95.2% (CI 95%: 89.9-100). 79% of these patients had NUD, which the eradication rate was 93.9% in this group of patient.

Based on ITT analysis, the eradication rates were 92.1% (CI 95%: 83.5-100%) in the synbiotic group and 63.2% (CI 95%: 47.9- 78.5%) in the placebo group. The difference was statistically significant. However, there was no statistically difference between the two groups based on PP analysis (table 2).

### Adverse effects and dyspepsia improvement

We did not find any statistically significant difference in frequency and severity of flatulence, taste abnormality, epigastric pain, nausea and vomiting, constipation, and diarrhea between the two groups but anorexia was significant in treatment period (p<0.05). The most common symptoms were taste abnormality and flatulence that were detected in most of the patients in treatment period (table 3).

The mean GDQ score before (3.36±3.25) and after (1.76±2.61) treatment in placebo group was statisti-

**Table 1: Comparison of patients' characteristic in study groups**

| Variables           | Probiotic (n=38) | Placebo (n=38) | P value |
|---------------------|------------------|----------------|---------|
| Sex                 |                  |                | NS      |
| Male                | 18 (47.36%)      | 17 (44.73%)    |         |
| Female              | 20 (52.63%)      | 21 (55.27%)    |         |
| Age (years)         |                  |                | NS      |
| Mean ± SD           | 43.75±13.32±     | 43.35±12.29    |         |
| Weight (Kg)         |                  |                | < 0.05  |
| Mean±SD             | 66.87±10.29      | 74.35±16.07    |         |
| Smoking             |                  |                | NS      |
| No                  | 30 (78.94%)      | 30 (78.94%)    |         |
| Ex                  | 5 (13.15%)       | 2 (5.26%)      |         |
| Active              | 3 (7.91)         | 6 (15.78)      |         |
| Endoscopic findings |                  |                | NS      |
| DU                  | 8(21.05%)        | 7(18.42%)      |         |
| GU                  | 0                | 0              |         |
| NUD                 | 30(78.95%)       | 31(81.58%)     |         |

NS= Not Significant

**Table 2: Eradication rate between two study groups based on method of analysis**

| Variable     | Probiotic group   | Placebo group       | P value |
|--------------|-------------------|---------------------|---------|
| Eradication  |                   |                     |         |
| ITT analysis | 92.1% (83.5-100%) | 63.15% (47.9-78.5%) | P<0.05  |
| PP analysis  | 92.1% (83.5-100%) | 100% (24 of 24)     | NS      |

NS= Not Significant

cally different ( $P<0.05$ ). The mean GDQ score before ( $2.76\pm 2.07$ ) and after ( $1.53\pm 1.54$ ) treatment in symbiotic group was statistically different ( $P<0.05$ ) but there was no difference between GDQ scores of patients in the two groups before and after treatment, respectively (table 4).

## DISCUSSION

*H. pylori* associated diseases that require effective eradication, has increased and we encounter some problems such as treatment resistance and failure. Patients frequently experience GI upset, diarrhea, nausea, vomiting, anorexia, and bloating associated with eradication. Drugs side effects can lead to discontinuation of therapy. The most important predictors of anti *H. pylori* treatment failure are poor compliance and antibiotic resistance.

Probiotics are defined by the FAO/WHO as live microorganisms, which are administered in adequate amounts, confer a health benefit on the host and are widely used as a live microbial food supplement that can improve the intestinal microbial balance.<sup>18</sup> Prebiotics are

currently defined as a non-digestible food ingredient that beneficially affects the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria in the colon.<sup>19</sup> Galactooligosaccharides are one category of prebiotics, which contain growth-promoting factors for *Bifidobacterium*. Synbiotics are generally considered as a combination of probiotics and prebiotics.

Shimizu and colleagues showed that *Bifidobacterium breve* strain Yakult and *Lactobacillus casei* strain Shirota as probiotics and galactooligosaccharides as prebiotics, may cause significantly greater levels of benefit in sepsis. *Bifidobacterium* and *Lactobacillus* and short chain fatty acids (SCFAs) cause lower incidence of infectious complications such as enteritis, pneumonia, and bacteremia than those who received no synbiotics.<sup>20</sup> The presumptive mechanism of synbiotics is that their administration increases the levels of beneficial bacteria such as *Bifidobacterium* and *Lactobacillus*. The increased level of total anaerobes induces increased production of SCFAs in GI tract. These environmental changes can

**Table 3:** Prevalence of the most frequent tolerable side effects occurred in patients of each groups and comparison of them

| Variables         |                      | Placebo group | Probiotic group | p-value |
|-------------------|----------------------|---------------|-----------------|---------|
| Diarrhea          | 10 <sup>th</sup> day | 18.2 %        | 8.3 %           | NS      |
|                   | 17 <sup>th</sup> day | 10 %          | 8.3 %           | NS      |
|                   | 24 <sup>th</sup> day | 0             | 5.6 %           | NS      |
| Epigastric pain   | 10 <sup>th</sup> day | 59.1 %        | 47.2 %          | NS      |
|                   | 17 <sup>th</sup> day | 40 %          | 36.1 %          | NS      |
|                   | 24 <sup>th</sup> day | 50 %          | 39.4 %          | NS      |
| Flatulence        | 10 <sup>th</sup> day | 50 %          | 50 %            | NS      |
|                   | 17 <sup>th</sup> day | 45 %          | 45.7 %          | NS      |
|                   | 24 <sup>th</sup> day | 30%           | 55.9 %          | NS      |
| Taste abnormality | 10 <sup>th</sup> day | 81.8 %        | 77.8 %          | NS      |
|                   | 17 <sup>th</sup> day | 75 %          | 61.1 %          | NS      |
|                   | 24 <sup>th</sup> day | 45 %          | 38.2 %          | NS      |
| Constipation      | 10 <sup>th</sup> day | 40.9 %        | 33.3 %          | NS      |
|                   | 17 <sup>th</sup> day | 20 %          | 36.1 %          | NS      |
|                   | 24 <sup>th</sup> day | 15 %          | 33.3%           | NS      |
| Nausea            | 10 <sup>th</sup> day | 26.1 %        | 28.6 %          | NS      |
|                   | 17 <sup>th</sup> day | 33.3 %        | 36.1 %          | NS      |
|                   | 24 <sup>th</sup> day | 20 %          | 17.6 %          | NS      |
| Vomiting          | 10 <sup>th</sup> day | 4.3 %         | 5.6 %           | NS      |
|                   | 17 <sup>th</sup> day | 4.8 %         | 0               | NS      |
|                   | 24 <sup>th</sup> day | 0             | 6.1 %           | NS      |
| Anorexia          | 10 <sup>th</sup> day | 40.9 %        | 13.9 %          | P< 0.05 |
|                   | 17 <sup>th</sup> day | 30 %          | 8.3 %           | P< 0.05 |
|                   | 24 <sup>th</sup> day | 15 %          | 15.2 %          | NS      |

NS= Not Significant

help preserve the gut flora. The beneficial changes in gut flora and environment by synbiotics administration may enforce systemic immune function and decrease the incidence of septic complications such as pneumonia, enteritis, and bacteremia in patients with severe systemic inflammatory response syndrome (SIRS).<sup>21</sup> A common mistake is the belief that prebiotic can increase the population and/or function of probiotic, whereas prebiotics stimulate the flora that is already present. According to similar evidence, we chose this regimen in our study design. Study drug contained *Lactobacillus casei*, *Lactobacillus rhamnosus*, *Streptococcus thermophilus*, *Bifidobacterium breve*, *Lactobacillus acidophilus*, *Bifidobacterium longum*, *Lactobacillus bulgaricus*, and also Fructooligosaccharide as prebiotic, Magnesium, and a vegetable capsule (hydroxypropyl methyl cellulose).

In our study, ITT analysis showed that adding synbiotic to commercial anti *H. pylori* regimen led to a better drug tolerance. Therefore this group of patients uses their medications well. The eradication rate of 92.1% in this group is a reasonable rate. Cure rates below 80% by ITT analysis, are the accepted threshold separating acceptable from unacceptable treatment results. Grading clinical studies into effectiveness categories using pre-specified criteria would allow clinicians to objectively identify and compare regimens. Graham and colleagues offer a therapy report card similar to that used to grade the performance of school children. The ITT cure rate categories are: “F” or unacceptable (80%), “D” or poor (81-84%), “C” or fair (85-89%), “B” or good (90-95%), and “A” or excellent (95-100%). Regimens scoring as

**Table 4:** Comparison of GDQ score in the two groups before and after treatment

|           | (SD ± mean) GDQ score |                 | Difference  | 95% CI      | P value |
|-----------|-----------------------|-----------------|-------------|-------------|---------|
|           | Before treatment      | After treatment |             |             |         |
| Placebo   | 3.36 ± 3.25           | 1.76 ± 2.61     | 1.6 ± 3.09  | 0.15 – 3.04 | < .05   |
| Probiotic | 2.76 ± 2.07           | 1.53 ± 1.54     | 1.23 ± 2.37 | 0.39 – 2.07 | < .05   |

NS= Not Significant

“B” or “good” can be used if “excellent” results are not obtainable.<sup>22</sup> As a general rule, clinicians should prescribe therapeutic regimens that have a  $\geq 90\%$  or, preferably,  $\geq 95\%$  eradication rate locally as mentioned by Rimbara and co-workers.<sup>23</sup> They emphasized that if no available regimen can achieve a  $\geq 90\%$  eradication rate, clinicians should use the most effective regimen(s) available locally.<sup>23</sup> This goal was achieved by our proposed regimen.

Gastroduodenal microbiota could protect the gut mucosa. Consequently the intake of exogenous lactic acid bacteria, particularly those with probiotic properties, may reinforce these protective functions in the stomach by maintaining local microbiological homeostasis, interfering with *H. pylori* and/or decreasing inflammatory processes.<sup>24</sup>

Two main types of substances have been implicated in the inhibition of *H. pylori* by lactic acid bacteria: SCFAs and bacteriocins. Probiotics produce SCFAs and have an important role in decreasing pH. A dose-dependent inhibition of *H. pylori* growth has been observed with acetic and lactic acid, the later demonstrating the most intense effect.<sup>25</sup> Such antimicrobial activity may be due to a direct effect on *Helicobacter* and also the inhibition of its urease activity. Bacteriocins are small, heat-resistant peptides, which are synthesized by several bacterial species including lactic acid bacteria with potential anti-*H. pylori* activity. This peptide with other heat-stable proteinaceous compounds are capable of inhibiting the growth of both antibiotic-resistant and -sensitive strains of *H. pylori*.<sup>26-27</sup>

Accumulating evidence suggests an important role of interleukin-8 (IL-8) in *H. pylori* infection associated peptic ulcer and it is an important mediator in the immunopathogenesis of chronic gastritis caused by *H. pylori*.<sup>28</sup>

It has been demonstrated that *cagA* and *vacA-s1* positive strains of *H. pylori* induce production of IL-8 in the gastric mucosa, both in vivo and in vitro. In addition,

an association between the mucosal levels of IL-8 and severity of gastritis and presence of PUD has also been reported.<sup>29</sup> IL-8 appears paramount in the acute inflammatory response to *H. pylori* infection, as this gene is involved in all significant response pathways in the initial cellular response to infection. Several authors have demonstrated increase in IL-8 in response to *H. pylori* in both in vivo<sup>30</sup> and in vitro studies.<sup>31-32</sup> IL-8 is a key chemokine in accumulating neutrophils. Gastric mucosal IL-8 levels have shown a positive correlation with the degree of stomach corpus inflammation.<sup>33</sup> Some probiotics exert an anti-inflammatory effect by decreasing dose dependently the release of IL-8, thus they prevent *H. pylori* colonization and the development of gastritis. *Lactobacillus acidophilus* (*johnsonii*) La1 decrease gastric inflammation in colonized animals.<sup>34</sup>

Increased levels of some growth factors and prostaglandins have been implicated in the protective effect of some strains of *B. breve* (that was present in our study drug) and *B. bifidum* against gastric ulcers induced by acetic acid or ethanol in rats.<sup>35</sup>

Other possible mechanisms of protection induced by probiotics including the stimulation of the expression of gastric mucins, decreases in bacterial overgrowth, stimulation of local immune responses and release of antioxidant substances.<sup>36</sup>

In our study, the most prominent benefit of synbiotics on drug side effects was improvement of anorexia. Among the various strains of probiotic, *Lactobacillus* causes some benefits. Increased appetite is one of them. There were four different *Lactobacillus* species in our study used product. In a recent meta-analysis the incidence of diarrhea was significantly reduced in the probiotic group (OR=0.21, 95%CI: 0.06-0.74), whereas the incidence of taste disorders, metallic taste, vomiting, nausea, and epigastric pain did not differ significantly between the probiotic group and the control group.<sup>37</sup> In another recent comprehensive analysis, at a first glance,

probiotics seem effective in decreasing side effects. At a closer look, however, evidence only supports these claims for specific probiotic strains, ineffective antibiotic therapies, and low-quality trials.<sup>38</sup>

Previous meta-analyses and review articles have suggested that probiotic supplements could improve *H. pylori* eradication.<sup>37-38</sup> Our study showed that probiotics with prebiotics could raise this rate significantly.

Probiotics are under valuable studies, as the idea holds promise for human health and well-being, and corresponding commercial opportunities. Protection of consumers requires health claims to be confirmed by important scientific documentations. Overall scientific demonstration of probiotic effects requires defining a healthy body microbiota and interactions between these microbiota and host factors, and the difficulty to define probiotic effectiveness in health and disease. Recent developments of high-throughput sequencing technology and the consequent progresses of metagenomics represent a new approach for the future of probiotics research.<sup>39</sup>

Our study had some limitations. First of all, a high dropout rate in conventional regimen was occurred. Clarithromycin was the main cause of treatment discontinuation. We could not obligate patients with intolerable side effects to continue the regimen despite their desire.

Cost is a major consideration in many countries such as ours. The cost of adding probiotic supplementations to standard regimens was a problem. Accompanying antibiotics with probiotics may decrease the number of effective bacteria in consumed capsules. This may be another difficulty of our work that could influence our results. On the other hand, the results may not be applicable to other countries and populations. The low rate of withdrawal in probiotic group may not be reproducible outside the trial setting, as the prescribing physician does not explain the temporary nature of drug side effects and the significance of completing the prescribed regimen. Finally, although adding probiotics causes an improvement over current therapies, it does not decrease the duration of therapy. Combined probiotic supplementation appears to be a promising therapeutic agent for treatment of some diseases in adults. However, there is concern that although probiotics are considered non-pathogenic,

it may be infective when the patient is acutely ill or immunosuppressed. Some factors should be considered, including the safety, strain selection, product stability, and formulation. It should always be kept in mind that probiotic must stay viable to bring the beneficial effect and its viability is affected by temperature, humidity, and oxygen concentration. The product should also be delivered in right dose. Lower dose of probiotic in the market may not provide the beneficial effects as reported in clinical studies. Therefore, further controlled clinical studies, as well as good storage, handling and distribution is essential to explore the therapeutic benefit for possible practical clinical application. We recommend designing other studies in order to define the best duration and dose of such probiotic added combination regimens.

In the future, considerable advances will be made in understanding the evolution of the organism and pathogenesis of diseases. Although combination therapies have high rates of eradication, the preferred treatment regimen would be one which use less medications especially with a lower dose and with a short course with more tolerability or without any adverse effects.

Finally, we recommend adding synbiotic supplementations to various anti *H. pylori* regimens, for a better eradication in patients without any contraindications to their use.

#### CONFLICT OF INTEREST

The authors declare no conflict of interest related to this work.

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