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META-ANALYSIS

Probiotic monotherapy and *Helicobacter pylori* eradication: A systematic review with pooled-data analysis

Giuseppe Losurdo, Rossella Cubisino, Michele Barone, Mariabeatrice Principi, Gioacchino Leandro, Enzo Ierardi, Alfredo Di Leo

Giuseppe Losurdo, Rossella Cubisino, Michele Barone, Mariabeatrice Principi, Enzo Ierardi, Alfredo Di Leo, Section of Gastroenterology, Department of Emergency and Organ Transplantation, University "Aldo Moro", Piazza Giulio Cesare, Bari 70124, Italy

Gioacchino Leandro, National Institute of Gastroenterology, "S De Bellis" Research Hospital, Via Turi, Castellana Grotte (BA) 70013, Italy

ORCID number: Giuseppe Losurdo (0000-0001-7038-3287); Rossella Cubisino (0000-0001-5983-3726); Michele Barone (0000-0001-8284-5127); Mariabeatrice Principi (0000-0003-0545-5656); Gioacchino Leandro (0000-0001-6624-4532); Enzo Ierardi (0000-0001-7275-5080); Alfredo Di Leo (0000-0003-2026-1200).

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Correspondence to: Enzo Ierardi, MD, Professor, Section of Gastroenterology, Department of Emergency and Organ Transplantation, University "Aldo Moro", Piazza Giulio Cesare 11, Bari 70124, Italy. e.ierardi@virgilio.it
Telephone: +39-80-5593452
Fax: +39-80-5593088

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Abstract

AIM

To define probiotic monotherapy effect on *Helicobacter pylori* (*H. pylori*) status by performing a systematic review.

METHODS

Methods of analysis and inclusion criteria were based on PRISMA recommendations. Relevant publications were identified by searching PubMed, MEDLINE, Science Direct, and EMBASE. The end-point was to estimate eradication rate and urea breath test delta value before and after probiotic monotherapy across all studies and, overall, with a pooled data analysis. Adverse events of probiotic therapy were evaluated. The data were expressed as proportions/percentages, and 95%CIs were calculated. For continuous variables, we evaluated the weighted mean difference. Odd ratios (ORs) were calculated according to the Peto method for the comparison of eradication rates between probiotics and placebo.

RESULTS

Eleven studies were selected. Probiotics eradicated *H. pylori* in 50 out of 403 cases. The mean weighted eradication rate was 14% (95%CI: 2%-25%, *P* =

0.02). Lactobacilli eradicated the bacterium in 30 out of 235 patients, with a mean weighted rate of 16% (95%CI: 1%-31%). *Saccharomyces boulardii* achieved eradication in 6 out of 63 patients, with a pooled eradication rate of 12% (95%CI: 0%-29%). Multistrain combinations were effective in 14 out of 105 patients, with a pooled eradication rate of 14% (95%CI: 0%-43%). In the comparison of probiotics *vs* placebo, we found an OR of 7.91 in favor of probiotics (95%CI: 2.97-21.05, $P < 0.001$). Probiotics induced a mean reduction in delta values higher than placebo (8.61% with a 95%CI: 5.88-11.34, *vs* 0.19% for placebo, $P < 0.001$). Finally, no significant difference in adverse events was found between probiotics and placebo (OR = 1, 95%CI: 0.06-18.08).

CONCLUSION

Probiotics alone show a minimal effect on *H. pylori* clearance, thus suggesting a likely direct role.

Key words: *Helicobacter pylori*; Probiotics; Eradication; Meta-analysis; Breath test

Core tip: Despite several lines of evidence in the literature having demonstrated a pivotal role of probiotics as adjunctive treatment for *Helicobacter pylori* (*H. pylori*) eradication, national and international guidelines do not have a uniform consensus about their clinical application. Many meta-analyses have confirmed that co-administration of probiotics may have a beneficial effect on the prevention of side effects and eradication rates. Herein, we found that probiotic monotherapy may eradicate *H. pylori* in 14% of cases. Lactobacilli, *Saccharomyces boulardii* and multistrain combinations eradicated the bacterium with a rate of 16%, 12% and 14%, respectively. Probiotics were significantly more effective than placebo (OR = 7.91).

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INTRODUCTION

Helicobacter pylori (*H. pylori*) is a ubiquitous Gram-negative, flagellated organism, residing in the human stomach, where it may cause both malignant and nonmalignant diseases[1-3]. The treatment of *H. pylori* relies mainly on a combination of antibiotics. However, despite several therapeutic schemes having been proposed, the way towards ideal therapeutic management remains an unsolved issue[4].

Until a few years ago, triple therapy (based on a proton pump inhibitor, amoxicillin and clarithromycin) was considered as the standard first-line regimen. However, failure rates have increased recently, due to the spreading of antibiotic resistances, which are due to point mutations of the *H. pylori* genome[5]. For this reason, alternative first-line regimens have been proposed (sequential, concomitant, quadruple with and without bismuth, and hybrid). In this context, the geographic pattern of antibiotic resistances must also be studied as a relevant matter[6-9]. To now, the "ideal therapy" does not exist and this is the real limit for worldwide effective therapeutic guidelines[6].

A relevant problem related to *H. pylori* therapy failure is linked to patient compliance, which is often affected by antibiotic-associated adverse events, including diarrhea, nausea, vomiting and abdominal pain. Therefore, the development of a new strategy which could improve the eradication rate as well as reduce the frequency of adverse effects is advisable.

Probiotics are defined as "live microorganisms that, when administered in adequate amounts, confer a health benefit on the host"[10]. The intestinal microbiota is the community of microorganisms which colonizes the gut. It is an essential component of the luminal intestinal environment. Antibiotic-induced alteration of the microbiota may lead to diarrhea and other side effects[11]. Consequently, probiotic supplementation could restore microbial balance, thus preventing antibiotic-associated adverse events[12,13]. In particular, this benefit may be useful in *H. pylori* management for the need to administer a combination of antibiotics at high dose.

Furthermore, it is supposed that probiotics could interfere with potential pathogens which may colonize the stomach[14]. Indeed, probiotics may compete with *H. pylori* for host surface receptors and, thereby, inhibit its adhesion to epithelial cells[15]. Furthermore, it has been demonstrated that, *L. acidophilus* may hamper *H. pylori* urease activity *in vitro*[16]. Finally, lactobacilli produce lactic acid, which is able to counteract *H. pylori*-induced hypochlorhydria and has bactericidal effect itself[17]. For these reasons, it is possible to hypothesize that probiotics may exert a direct inhibitory effect on *H. pylori* growth.

Several meta-analyses have demonstrated that probiotics, when given in combination with the standard therapy, induce an improvement in both eradication rates and reduction of adverse events. In this regard, Zhang *et al*[18] demonstrated that probiotic administration along with triple therapy achieved a success rate of 82.31% (against the 72.08% of the control group), with a risk ratio of 1.11 in favor of probiotics. Another study[19] showed that probiotics have a positive effect on preventing diarrhea [odds ratio (OR) = 0.21] and increase the eradication rate, with an OR of 1.68.

Until now, meta-analyses have investigated pro-

biotic effects on *H. pylori* only in association with antibiotics. To the best of our knowledge, there are no meta-analyses concerning probiotic monotherapy effects on *H. pylori* infection. Therefore, our aim was to perform a systematic review with pooled data analysis regarding this uninvestigated topic.

MATERIALS AND METHODS

Eligibility criteria and study selection

Methods of analysis and inclusion criteria were based on "Preferred Reporting Items for Systematic reviews and Meta-Analyses" (PRISMA) recommendations[20], and the Meta-analysis Of Observational Studies in Epidemiology (MOOSE) checklist has been enclosed as supplementary material. We excluded review articles, experimental *in vitro* studies and single case reports.

Data collection process

A literature search was performed in May 2017. Relevant publications were identified by a search of PubMed, MEDLINE, Science Direct and Scopus. The search terms were *Helicobacter pylori*, probiotics, lactobacilli, bifidobacteria, saccharomyces, treatment, eradication, breath test. We used the following string, with Boolean operators AND/OR: ([Helicobacter pylori OR H. pylori] AND [probiotic* OR lactobacil* OR bifidobacteria OR saccharomyces OR bacillus OR treatment OR eradication OR breath test]). We excluded studies that used probiotics in combination with antibiotics, while co-administration of other molecules, such as proton pump inhibitors, was not considered as an exclusion criterion. We excluded, as well, studies in which patients with major gastrointestinal surgery interventions were enrolled.

Titles and abstracts of papers were screened by two reviewers (Losurdo G and Ierardi E). Studies were independently prescreened in blinded fashion for relevance by the two reviewers using full reports. Discussion put an end to any disagreements. Successively, data were extracted from the relevant studies by one reviewer and checked by a second reviewer, and thus inserted into dedicated tables. A third reviewer (Leandro G) came to a decision on any disagreements.

Reviewers independently extracted the following data from each paper: (1) year of publication; (2) country where the study was performed; (3) single- or multicenter study; (4) study design; (5) number of patients included; (6) mean age and sex of enrolled patients; (7) test used to diagnose *H. pylori* infection; (8) type of probiotic and modality of administration; (9) success rate; (10) delta values of urea breath test (UBT); and (11) adverse events. We did not include studies reporting only the results of UBT delta value without detailing eradication rate.

Summary measures and planned methods of analysis

The end-point was to estimate the mean eradication rate and variations of delta value at UBT across all studies and, overall, with a pooled data analysis. The data were expressed as proportions/percentages, and 95%CIs were calculated using the generic inverse variance method, as described in the Cochrane Handbook, Chapter 9.4.3.2[21], and as we already performed in a previous meta-analysis[22]. The inverse variance methods allow a "weighting" of the eradication rate according to the sample size. For continuous variables (delta value of UBT), we entered mean, standard deviations and sample size in order to calculate the weighted mean difference. OR and 95%CI were calculated, where available, based on the Peto method, for the comparison of two groups (probiotics *vs* placebo).

Data were entered into the RevMan 5.3 software (The Nordic Cochrane Centre, Copenhagen, Denmark) (Cochrane library) in order to draw forest plots. A P value < 0.05 was considered statistically significant. Heterogeneity was assessed by using the χ^2 and I^2 statistics. In particular, heterogeneity was considered to be present if the χ^2 test delivered a $P < 0.05$ and, therefore, the I^2 statistic was used to quantify the proportion of heterogeneity between the studies. In the presence of heterogeneity, a revision of included studies was carried out to assess the main reasons explaining the phenomenon and, therefore, a subgroup analysis was performed. Only if this attempt failed, a random effects model was employed, in order to minimize the impact of heterogeneity. We preferred a fixed effects model if less than 4 studies per outcome were included in the analysis[23].

The degrees of freedom (df) were reported for each analysis. We evaluated the quality of enrolled studies by the Jadad scale[24] for randomized clinical trials (RCTs) or by the Quality Assessment Tool for Case Series Studies (QATCSS) of the National Institutes of Health[25] for nonrandomized, open label pilot studies. Finally, when comparison between two groups (probiotics *vs* placebo) was performed, we drew funnel plots and applied Egger's regression method to estimate the asymmetry of the funnel plots, considering non-statistically significant results as absence of publication bias[26].

RESULTS

Study selection

The literature search found 1537 articles overall. After study selection, reported in detail in Figure 1, 11 studies were eligible for the analysis[27-37]. Only 7 of them were RCTs[27,29,30,32,33,36,37]. A total of 517 *H. pylori*-infected patients were recruited. Of these, 114 received a placebo treatment and served as a control group, and the remaining 403 had probiotic

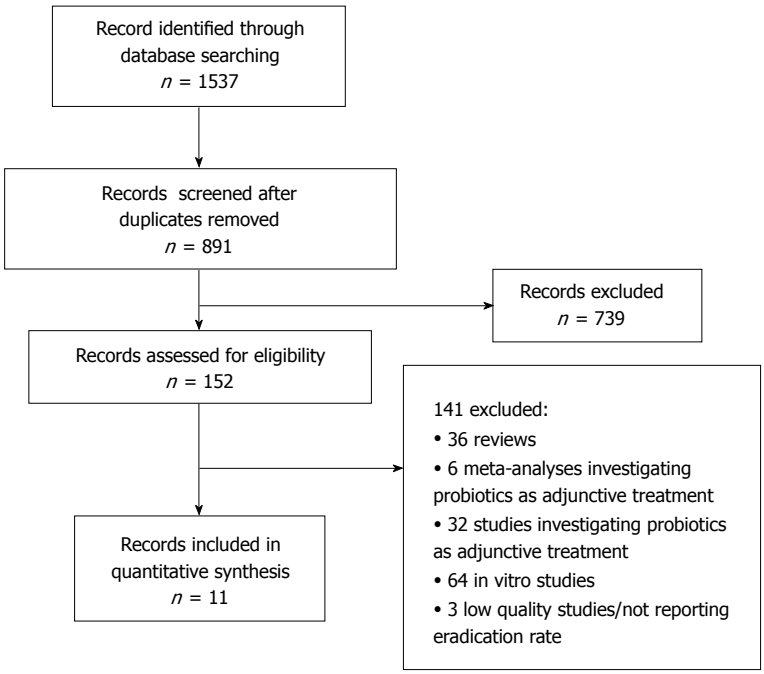

Figure 1 Flowchart showing the process of study selection for the systematic review.

supplementation. In all studies except 2, the diagnosis was achieved by UBT[27,37], but in most cases the initial diagnosis was established by the combination of more than one test, including UBT, upper endoscopy with histology or rapid urease test, serology or stool antigen test (SAT). The verification of eradication of treatment was performed by UBT in all but 2[27,37], which used SAT both for diagnosis and eradication control. Details of the cut-offs used for diagnosis and timing of UBT are reported in Table 1.

Only 3 studies were conducted on the pediatric population[30,35,37]. In most cases (7), a lactobacilli-based formulation was employed, while only 2 studies administered *Saccharomyces boulardii*[28,35] and 3 investigated probiotic multistrain formulations[31,33,36]. Of note, only in 1 study[28] was probiotics given in combination with proton pump inhibitor. The duration of probiotic supplementation varied across the studies, from 10 d to 1 year. Quality assessment is reported in Table 2.

Overall effectiveness of probiotics in eradicating H. pylori

In the 11 selected studies, probiotics eradicated *H. pylori* in 50 out of 403 cases. The mean weighted eradication rate was 14%, with a 95%CI of 2%-25% (df = 4, P = 0.02). In 6 studies, probiotic treatment was unsuccessful[29,31,32,34,35,37], while the highest percentage of eradication (32.5%) was achieved in an Italian study[33]. The forest plot of such analysis is displayed in Figure 2A.

Further, we performed a sub-analysis comparing the success rate in RCT *vs* non-randomized studies (Figure 2B). The pooled rate was 14% for RCT (95%CI: 1%-27%, df = 3, P = 0.04) and 14% for non-randomized trials (95%CI: 0%-44%, P = 0.34). No difference was found between these two groups (P = 0.99).

Eradication rate according to the probiotic strain

Most of studies investigated a probiotic formulation based on a single lactobacilli strain (further details of species are listed in Table 1)[27-30,32,34,35]. Lactobacilli eradicated the bacterium in 30 out of 235 patients, with a mean weighted rate of 16% (95%CI: 1%-31%, df = 2). Multistrain combinations[31,33,36] were effective in 14 out of 105 patients, with a pooled eradication rate of 14% (95%CI: 0%-43%, df = 1). In the two studies evaluating *Saccharomyces boulardii*[30,37], the treatment was successful in 6 out of 63 subjects (pooled rate of 12%, 95%CI: 0%-29%). We did not find any statistically significant difference among these three formulations (P = 0.94). The forest plot of this analysis is reported in Figure 3.

Probiotics vs placebo in the eradication of H. pylori

Six RCTs[27,29,32,33,36,37] compared probiotics to a placebo (see Figure 4). In total, probiotics eradicated the bacterium in 38 out of 238 patients (15.9%), while placebo alone did not achieve any success (0 out of 114, 0%). The analysis, reported in Figure 4, provided an OR of 7.91 in favor of probiotics, with a 95%CI of 2.97-21.05. In this case, we used a fixed effects model since heterogeneity was absent (χ^2 = 0.75, df = 2, P = 0.69). A funnel plot, reported in Figure 5, showed that a possible bias could be detected, as confirmed by Egger's test (P = 0.02). However, the low number of included studies and the presence of 0% eradication rates (which are void for the test) imply that the test has a low statistical power, and therefore the possibility

Table 1 Main characteristics of the studies included in the quantitative analysis

Ref.	Nation	Age and sex	Probiotic strain and dose	Diagnosis	Control of eradication	Eradication rate % (n/N)
Boonyaritichaikij *et al*[27], 2009	Japan	62 ± 14 yr Male sex: 54.5%	Cheese with *L. gasseri* OLL2716 5 × 10^8 CFU/g for 12 mo	SAT	SAT after 12 mo	Probiotic: 29.3% (24/82) Placebo: 0% (0/6)
Dore *et al*[28], 2014	Italy	Mean age: 51 yr (range, 21-68) Male sex: 13.6%	*L. reuteri* 10^8 CFU/tablet bid + Pantoprazole 20 mg bid for 60 d	UBT	UBT after 30-40 d	14.3% (3/21)
Francavilla *et al*[29], 2008	Italy	53.3 ± 13.3 yr (probiotics) 52.4 ± 13.1 yr (placebo) Male sex: 57.5%	*L. reuteri* ATCC55730 10^8 CFU/tablet bid for 28 d	UBT (cut-off 3.5%), SAT, RUT, histology	UBT after 4 wk	Probiotic: 0% (0/20) Placebo: 0% (0/20)
Gotteland *et al*[30], 2005	Chile	8.5 ± 1.7 Male sex: 49.6%	*L. acidophilus* LB 10^9/tablet bid or *S. boulardii* 250 mg + inulin 5 g bid for 8 wk	UBT (cut-off 5‰)	UBT after 1 d	9.3% (9/97) *L. acidophilus* 6.5% (3/46) *S. boulardii* 11.8% (6/51)
Myllyluoma *et al*[31], 2007	Finland	Mean age: 51 yr (range, 40-69)	Multi-strain (*L. rhamnosus* GG, *L. rhamnosus* LC705, *P. freudenreichii* JS, *B. lactis* Bb12) 2.5 × 10^9 CFU/day for 8 wk	UBT (cut-off 2.2%), RUT, histology	UBT after 8 wk	0% (0/6)
Pantoflickova *et al*[32], 2003	Switzerland	25 ± 5 yr Male sex: 50%	*L. johnsonii* bid for 3 wk, then once daily for 13 wk	UBT (cutoff 5%), histology, culture, RUT, serology	UBT, culture at the end of treatment	Probiotic: 0% (0/25) Placebo: 0% (0/25)
Rosania *et al*[33], 2012	Italy	52.4 ± 21.7 yr (probiotics) 48.7 ± 25.3 yr (placebo) Male sex: 42.5%	Multi-strain (*S. termophilus*, *L. acidophilus*, *B. longum*, *L. plantarum*, *B. brevis*, *L. paracasei*, *B. infantis*, *L. delbrueckii*) 1800 × 10^9/d for 10 d	UBT (cut-off 4%)	UBT after 4 wk	Probiotic: 32.5% (13/40) Placebo: 0% (0/40)
Sakamoto *et al*[34], 2001	Japan	50.1 ± 7.4 yr Male sex: 93.1%	Yoghurt + *L. gasseri* OLL2716 1-1.4 × 10^7 CFU/g bid for 8 wk	UBT (cut-off 5%)	UBT after 9 wk	0% (0/29)
Shimizu *et al*[35], 2002	Japan	Mean age: 12.1 yr (range, 7.4-15.8) Male sex: 41.7%	Yoghurt + *L. gasseri* OLL2716 1-1.4 × 10^7 CFU/g bid for 8 wk	SAT, UBT	SAT, UBT after 4 and 10 wk	0% (0/12)
Wang *et al*[36], 2004	China	Not available	Multi-strain yoghurt (*L. acidophilus* La5, *B. lactis* Bb12, *L. bulgaricus*, *S. termophilus*) > 10^7 bacteria/mL for 6 wk	UBT (cut-off 3.5%), histology	UBT after 8 wk	Probiotic: 1.7% (1/59) Placebo: 0% (0/11)
Namkin *et al*[37], 2016	Iran	Age range of 9-12 yr Male sex: 20.8%	*S. boulardii* 250 mg/d for 1 mo	SAT	SAT after 8 wk	Probiotic: 0% (0/12) Placebo: 0% (0/12)

CFU: Colony forming units; RUT: Rapid urease test; SAT: Stool antigen test; UBT: Urea breath test.

Table 2 Quality assessment according to the type of studies

Ref.	Type of study	Jadad score[1]	QATCSS score[2]
Boonyaritichaikij *et al*[27], 2009	Randomized, single blind placebo-controlled, pilot	3	NA
Dore *et al*[28], 2014	Prospective, single center, open label pilot study	NA	8
Francavilla *et al*[29], 2008	Randomized, double blind placebo-controlled	4	NA
Gotteland *et al*[30], 2005	Randomized, open study	3	NA
Myllyluoma *et al*[31], 2007	Prospective, single center, open label pilot study	NA	7
Pantoflickova *et al*[32], 2003	Randomized, double blind placebo-controlled	4	NA
Rosania *et al*[33], 2012	Randomized, double blind placebo-controlled	4	NA
Sakamoto *et al*[34], 2001	Single center, open label pilot study	NA	6
Shimizu *et al*[35], 2002	Single center, open label pilot study	NA	6
Wang *et al*[36], 2004	Randomized, double blind placebo-controlled	2	NA
Namkin *et al*[37], 2016	Randomized, double blind placebo-controlled	5	NA

[1]Jadad scale reaches a maximum score of 5; [2]QATCSS reaches a maximum score of 9. NA: Not applicable.

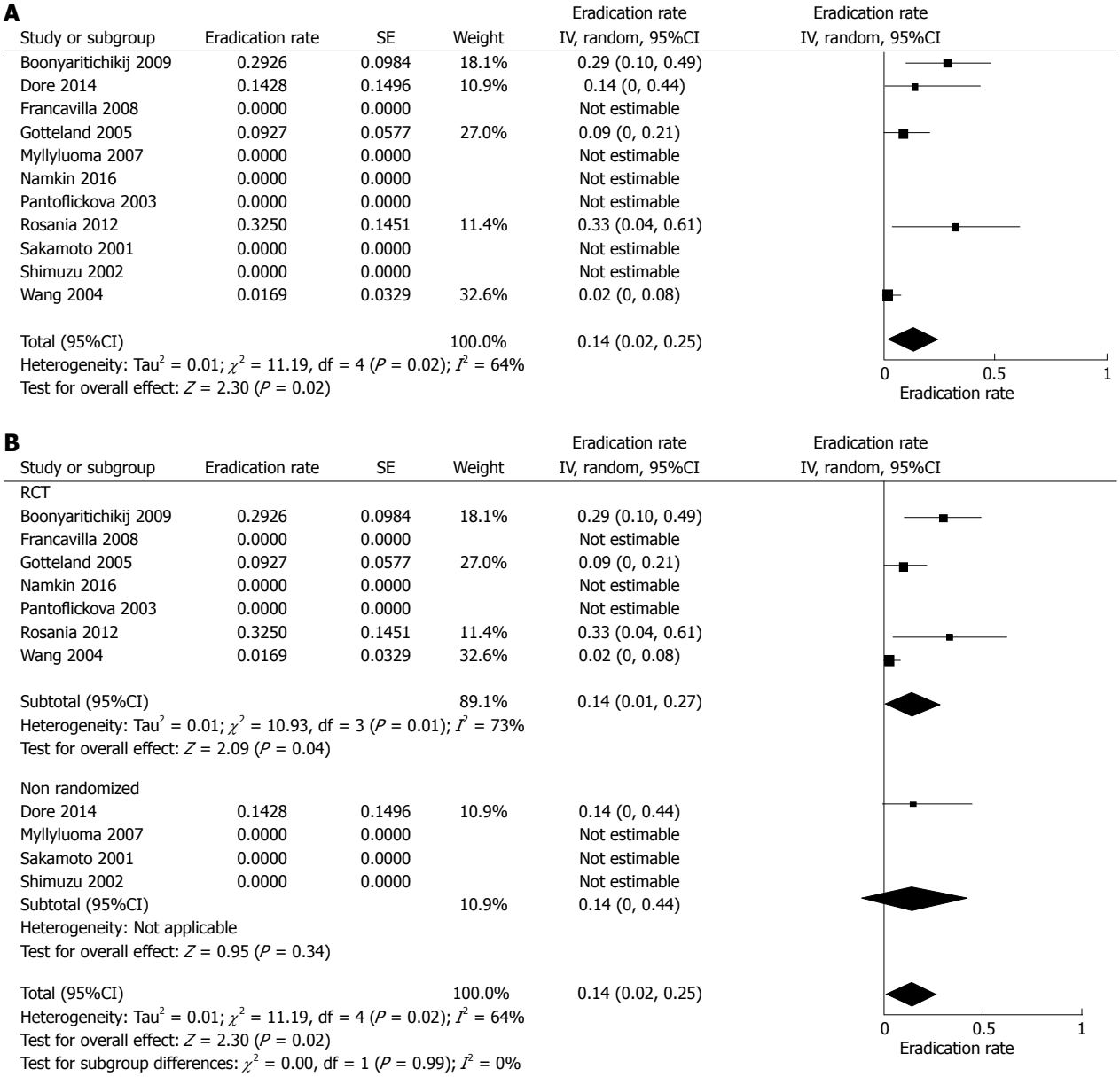

Figure 2 Mean eradication rate of probiotics for *H. pylori* infection (A). B: A sub-analysis according to the type of studies (randomized controlled trials (RCTs) *vs* open label studies) is reported.

of bias is questionable anyway.

Variations in delta values for UBT

We aimed to evaluate whether probiotics' administration alone could reduce the expired ^{14}C-marked CO_2 during the UBT. Six studies provided sufficient data (delta values expressed as ‰) to perform such analysis[29,30,33-36]. In two studies, delta values for placebo were reported[29,33].

Overall, probiotics induced a statistically significant mean reduction in delta values of 8.61‰ (95%CI: 5.88-11.34, df = 6) which was statistically significant. On the other hand, placebo implied a reduction of 0.19‰, which was not statistically significant (95%CI: -5.16-5.53, P = 0.94, df = 1). The test for subgroup differences demonstrated that probiotics significantly reduced delta compared to placebo (P = 0.006). In this analysis, despite a high heterogeneity (χ^2 = 47.08, df = 8, P < 0.001, I^2 = 83%) we used a fixed effects model since the number of included studies was low and the heterogeneity could be explained by the different type of probiotics and the different study design of enclosed trials. The forest plot of this analysis is reported in Figure 6.

Adverse events

Only 3 studies described adverse events during probiotic administration[28,30,37], and only 1 case of side effect was reported in 39 treated patients, with a pooled prevalence of 8% (95%CI: 0%-39%, P = 0.59). In only 1 study[37], side effect rate was reported both for placebo and probiotic groups. In this case, the

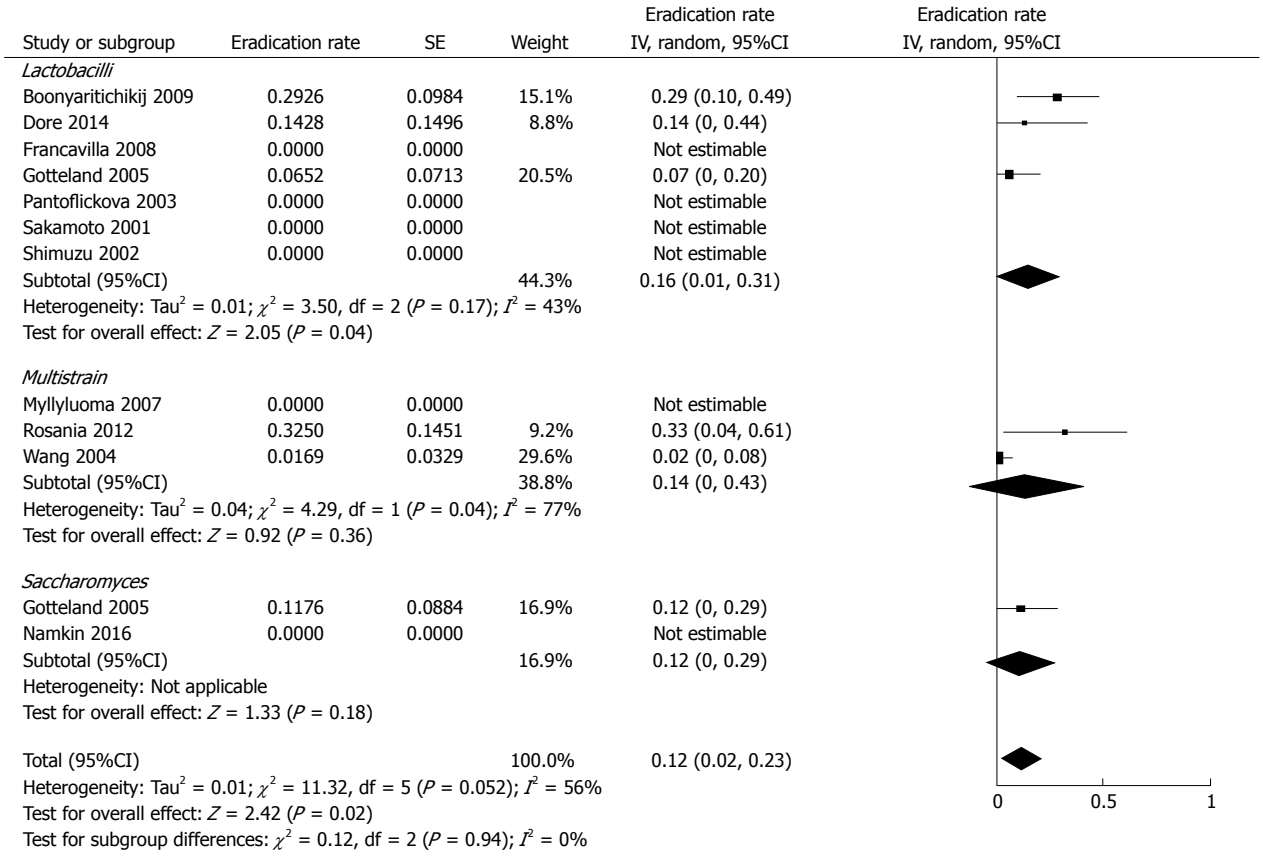

Figure 3 Sub-analysis of probiotics' effectiveness in *H. pylori* eradication according to the strain.

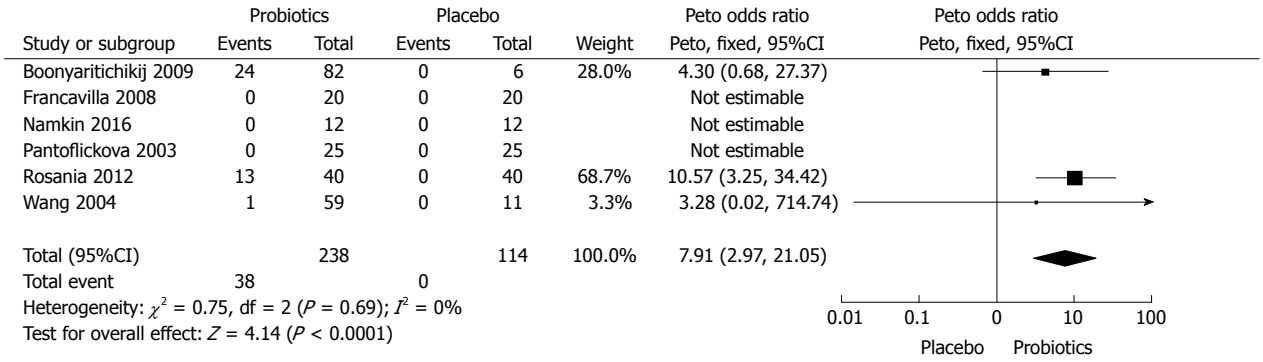

Figure 4 Meta-analysis comparing the eradication rate of probiotics against placebo.

meta-analysis did not show any difference between the two groups (OR = 1, 95%CI: 0.06-18.08, *P* = 1).

DISCUSSION

Despite several lines of evidence in the literature having demonstrated a consistent role of probiotics as adjunctive treatment for *H. pylori* eradication[38], national and international guidelines do not address a uniform consensus about their clinical application. The last Maastricht guidelines state that certain probiotics may have a beneficial impact on the eradication[39]. Similarly, Italian guidelines advise their use since they may reduce antibiotics-related side effects[40]. On the other hand, Toronto guidelines discourage routine probiotic administration in order to reduce side effects and improve the efficacy, since clinical trials and meta-analyses are characterized by low quality[41].

The most important issue that sets a limit to draw conclusions about the effects of probiotics in the treatment of *H. pylori* is that they have been considered only as an adjunctive treatment to antibiotics. In this context, probiotics demonstrated effectiveness mainly in reducing adverse events (especially diarrhea). However, these studies did not provide adequate evidence regarding a direct role in the eradication. Few studies have focused probiotic alone activity on bacteriotherapy in this field and, to date, this is the first systematic review on this topic.

In our analysis, the exclusive inclusion of studies

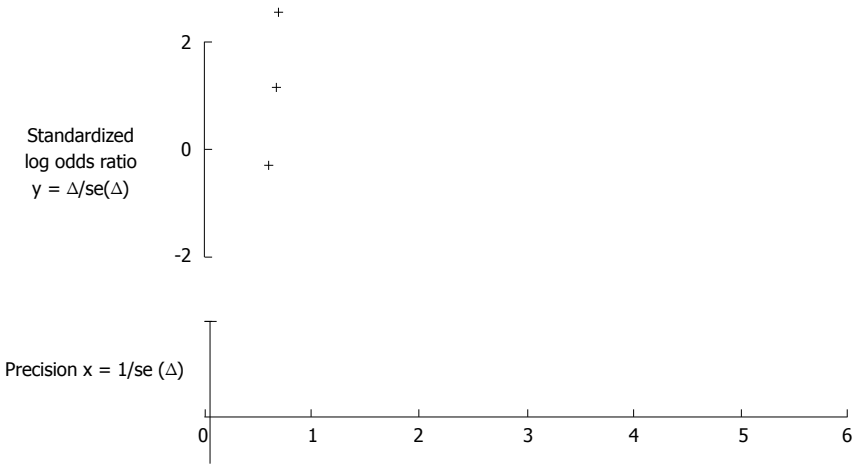

Figure 5 Funnel plot of the meta-analysis comparing the eradication rate of probiotics against placebo.

Study or subgroup	Before Mean	Before SD	Before Total	After Mean	After SD	After Total	Weight	Mean difference IV, fixed, 95%CI
Probiotics								
Francavilla 2008	33.8	15.0	20	27.3	12.1	20	8.3%	6.50 (-1.95, 14.95)
Gotteland lactobacilli	33.4	19.3	46	32.7	15.2	46	11.7%	0.70 (-6.40, 7.80)
Gotteland Saccharomyces	31.2	17.4	51	27.5	18.2	51	12.4%	3.70 (-3.21, 10.61)
Rosania 2012	39.5	19.3	40	12.5	8.7	40	13.7%	27.00 (20.44, 33.56)
Sakamoto 2001	26.6	13.7	29	20.9	11.8	29	13.7%	5.70 (-0.88, 12.28)
Shimuzu 2002	28.1	14.2	12	26.2	13.1	12	5.0%	1.90 (-9.03, 12.83)
Wang 2004	36.2	19.4	59	28.2	15.8	59	14.5%	8.00 (1.62, 14.38)
Subtotal (95%CI)			257			257	79.3%	8.61 (5.88, 11.34)
Heterogeneity: χ^2 = 39.37, df = 6 (P < 0.00001); I^2 = 85%								
Test for overall effect: Z = 6.18 (P < 0.00001)								
Placebo								
Francavilla 2008	35.8	15.5	20	37.3	16.2	20	6.1%	-1.50 (-11.33, 8.33)
Rosania 2012	33.5	15.5	40	32.6	13.5	40	14.6%	0.90 (-5.47, 7.27)
Subtotal (95%CI)			60			60	20.7%	0.19 (-5.16, 5.53)
Heterogeneity: χ^2 = 0.16, df = 1 (P = 0.69); I^2 = 0%								
Test for overall effect: Z = 0.07 (P = 0.94)								
Total (95%CI)			317			317	100.0%	6.86 (4.43, 9.30)
Heterogeneity: χ^2 = 47.08, df = 8 (P < 0.00001); I^2 = 83%								
Test for overall effect: Z = 5.53 (P < 0.00001)								
Test for subgroup differences: χ^2 = 7.55, df = 1 (P = 0.006); I^2 = 86.8%								

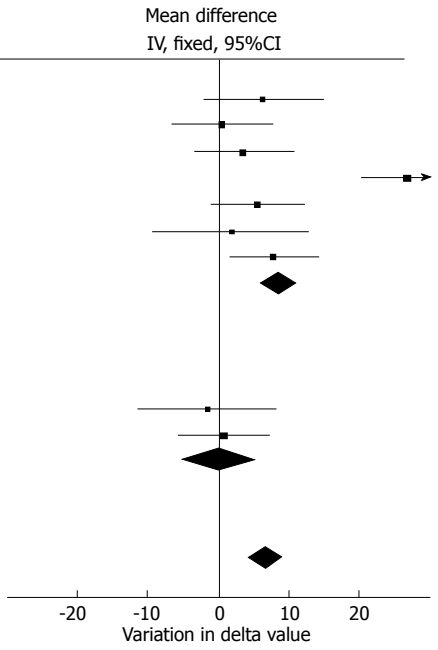

Figure 6 Variations of delta value for urea breath test before and after the treatment, both for probiotics and placebo.

using probiotics alone allowed us to draw more solid conclusions about the role of probiotics, since we removed the interference of factors and bias related to antibiotics such as inhomogeneous resistance pattern, variations in doses and administration modalities, patient compliance and adverse events. On the other hand, our analysis implied other limitations, such as the low number of enrolled patients, the differences of administered probiotic strains and the lack of randomization and/or a placebo arm as control group. For this reason, we attempted to limit these sources of heterogeneity by adding subgroup analyses and by choosing a random effect model heterogeneity that was high, a strategy that can minimize this phenomenon[23]. Finally and unfortunately, none of the included studies reported any data about smoking habits nor on alcohol assumption. Therefore, we were unable to perform a sub-analysis. This is another drawback, since it is known that such factors could influence the eradication. However, most of studies were conducted in pediatric populations, so that we may assume that such cases patients did not consume alcohol nor cigarettes.

The first relevant finding of this review is that probiotics alone may eradicate *H. pylori*, in 14%. From a clinical point of view, this is an unsatisfactory rate; however, taking into account that this percentage is considerably higher than placebo (0%, with a Peto OR = 7.91; Figure 4), we could assume that probiotic direct antibacterial action against *H. pylori* is consistent. Our analysis failed to ascertain whether some formulations may be more effective than others, but this limitation is due to the low number of included studies. Indeed, better outcome (32.5% of successful eradication) was achieved in the study which employed a multistrain

combination with the highest bacterial charge[33]. On the other hand, in 4 out of 7 studies using a single lactobacillus strain, no eradication was recorded. These observations may suggest that an association of more bacterial species could be more effective[42]. One study explored the effect of *Saccharomyces boulardii*, a yeast species, demonstrating a success rate of 11.8% and, thus, indicating a reliable performance in *H. pylori* gastritis[43,44].

The second important result concerns the variations in delta values for UBT. Indeed, as shown in Figure 6, in all studies, a reduction of delta values was observed in the probiotic arm, while delta values remained stable in subjects assuming placebo. This result is in agreement with evidence from the literature[45,46] and may suggest that probiotics could reduce the bacterial load in any case, despite a complete eradication not being obtained[47,48]. Indeed, labeled CO_2 in the expirate is considered as an indirect indicator of the density of gastric *H. pylori* colonization[49,50]. A probiotic-induced intragastric bacterial load reduction has been confirmed by histological semiquantitative analysis in some included studies[31] and even by a study, which used an original assessment of bacterial stool antigen[29,51].

In conclusion, preliminary data show that a primary therapeutic effect of probiotics may be hypothesized for *H. pylori*, but the low number of studies, their inhomogeneity in the design and the low number of enrolled patients are a critical limit to drawing evidence-based conclusions. However, the modulation of gastric microbiota could represent an intriguing aspect, since it does not imply the drawback of antibiotics (induction of dysbiosis, side effects) and is safe and probably more acceptable for patients[11,52].

ARTICLE HIGHLIGHTS

Research background

Probiotics have been largely used as adjunctive treatment for *Helicobacter pylori* (*H. pylori*) eradication, showing good results.

Research motivation

Until now, meta-analyses have investigated probiotic effects on *H. pylori* only in association with antibiotics. Therefore, our aim was to perform a systematic review with pooled data analysis regarding this uninvestigated topic.

Research objectives

The objective was to perform a meta-analysis aiming to calculate a pooled eradication rate for probiotic monotherapy, overall and according to the strain.

Research methods

Article search and selection was conducted according to the PRISMA criteria. We performed a pooled-data analysis using to the inverse variance method to calculate the mean weighted eradication rate. Peto odd ratio (OR) was calculated for the comparison "probiotics *vs* placebo". For continuous variables (delta value of urea breath test), we entered mean, standard deviations and sample size in order to calculate the weighted mean difference.

Research results

We found that probiotic monotherapy may eradicate *H. pylori* in 14% of cases. Lactobacilli, *Saccharomyces boulardii* and multistrain combinations eradicated the bacterium with a rate of 16%, 12% and 14%, respectively. Probiotics were significantly more effective than placebo (OR = 7.91). Moreover, probiotics were able to reduce delta values in the expirate of urea breath test.

Research conclusions

The eradication rate of probiotics' monotherapy is disappointing; however, our meta-analysis showed that, in some cases, they are able to defeat the bacterium. They compete with *H. pylori* for host surface receptors and, thereby, inhibit its adhesion to epithelial cells. Furthermore, it has been demonstrated that probiotics could hamper *H. pylori* urease activity. On these bases, since probiotics administration does not carry the risk of antibiotic resistance, it could represent an optimal strategy in selected cases.

Research perspectives

Further studies on large sample size are necessary to draw more solid conclusions about a direct inhibitory effect of probiotics on *H. pylori*.

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