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REVIEW ARTICLE

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Adjunctive probiotics after periodontal debridement versus placebo: a systematic review and meta-analysis

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ABSTRACT

Objective: To comprehensively investigate the efficacy of adjunctive probiotics compared to placebo, using conventional and novel treatment outcomes.

Materials and methods: Three databases (MEDLINE, EMBASE, and CENTRAL) were searched. Outcomes included percent change in the total number of deep sites before and after therapy, change in mean probing pocket depth (mm), percentage patients requiring additional therapy, risk for disease progression, and microbiological and immunological results. Meta-analysis was conducted to evaluate treatment effects wherever appropriate.

Results: Ten studies were selected from 818 records. Meta-analysis showed that adjunctive probiotics had no additional benefit for percentage change of the total number of deeper sites ($\geq$5 mm, $\geq$6 mm, $\geq$7 mm) before and after therapy. No significant difference was observed for mean probing pocket depth reduction at 3 and 6 months. Statistically significant beneficial odds ratios for need for additional therapy (OR = 0.19, 95% CI [0.07–0.56]) and risk of disease progression (OR = 0.32, 95% CI [0.14–0.73]) were observed with probiotic administration. Immunological rather than microbiological outcomes correlated more consistently with clinical findings. No adverse events were reported.

Conclusions: Adjunctive probiotics are safe in systemically healthy individuals and could offer additional patient-level benefits compared to placebo, hence its use can sometimes be justified.

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Introduction

Periodontitis is a prevalent chronic inflammatory disease associated with several negative downstream sequelae [1]. Advances in next-generation sequencing support a polymicrobial model of community dysbiosis, and the transition to disease resembles ecological succession culminating in a higher proportion of oral pathobionts [2]. Current understanding is that non-resolving inflammation drives the conversion to a dysbiotic community, and intrinsic host-related factors and defense mechanisms may play an overriding role in disease pathogenesis [3]. Although non-surgical therapy remains the gold standard for the treatment of periodontitis, some sites may not improve significantly even after treatment [4,5]. To achieve better outcomes, adjuncts such as local or systemic antibiotics, antiseptics, lasers, or photodynamic therapy have been used [6–9]. Antibiotics have been investigated extensively, however there are concerns about resistance and patient compliance [10,11]. Thus, probiotics represent an alternative intervention to improve non-surgical outcomes, with no known adverse effects [12].

As inflammation is a self-sustained forward loop that perpetuates disease, the use of adjunctive host-modulation therapies to inhibit or promote resolution of inflammation may improve treatment outcomes [13]. Probiotics are one such modality, and their principles of use and applications in periodontology have been discussed in a recent review [14]. To exert their effects, oral probiotic bacteria need to possess the ability to adhere to and colonize periodontal tissues and become part of the biofilm [15]. Evidence for their mode of action can be divided into four categories: Competitive exclusion, signaling interference, production of antimicrobial substances, and immune modulation [16]. Competitive displacement refers to the displacement of an existing microorganism with a more competitive one, this may include competition for adhesion sites and nutrients. *In vitro* studies have supported the ability of probiotic species to bind to saliva-coated hydroxyapatite and buccal epithelial surfaces, thus preventing the adherence of other bacteria [17,18]. Probiotic strains have also demonstrated the ability to co-aggregate with other bacterial strains and inhibit their growth and biofilm formation [19,20].

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Probiotics may also act *via* signaling interference. Quorum sensing is the process by which bacteria produce and detect signal molecules to coordinate their behavior [21]. An example is the accessory gene regulator (agr) quorum sensing system used by *Staphylococcus aureus* which is important for up-regulation of virulence factors during infection and the development of acute disease [22]. An *in vitro* study demonstrated that signaling molecules produced by *Lactobacillus reuteri* have the potential to interfere with this quorum-sensing system within *S. aureus*, resulting in decreased production of virulence factors [23]. More recently, a human study demonstrated that probiotic *Bacillus* was able to block this signaling system, thus reducing the infectivity of *S. aureus* infections [24]. While these actions have not yet been described in the dental literature, quorum-sensing regulation appears to be a valid mechanism for further research.

Probiotics such as *Lactobacillus* also have antibacterial activity *via* bacteriocins or other products. Lactic acid production from *Lactobacillus* lowers pH and disrupts the outer membrane of Gram-negative bacteria [25,26]. Fortunately, short-term consumption of these probiotics does not appear to influence the acid production of plaque [27]. *Lactobacillus* and *Streptococcus* species also produce hydrogen peroxide which is bactericidal and fungicidal *via* its oxidizing effect [28,29]. Bacteriocins are antimicrobial peptides produced by bacteria and these toxins play a critical role in mediating competitive dynamics between bacterial strains [30]. Reuterin, a bacteriocin produced by *L. reuteri*, can inhibit bacterial growth, yeasts, fungi, and viruses [31,32]. In the oral cavity, *L. reuteri* has demonstrated degradation activity and growth inhibition on pathogens such as *Porphyromonas gingivalis* and *Actinomyces actinomycetemcomitans* [33–35]. On the other hand, bacteriocin production by *Bifidobacterium* species appears to be a rarer trait [36].

Probiotic bacteria and their products can increase the phagocytic capacity of macrophages, enhance natural killer cell activity, and regulate the expression of phagocytosis receptors [37–39]. *Streptococcus dentisani* strongly increased the secretion of the anti-inflammatory cytokine IL-10 and reduced the expression of interferon gamma after incubation with *P. gingivalis* and *Fusobacterium nucleatum in vitro* [40]. Another immunomodulatory mechanism of probiotics is *via* the expression of matrix metalloproteinases. Reduced gingival crevicular fluid levels of matrix metalloproteinases have been reported after administration of probiotics [41,42]. Furthermore, different *Lactobacillus* strains can enhance macrophage activation and reduce the levels of tumor necrosis factor-alpha and interleukin-6 in gingival tissue [43,44].

There is increasing interest towards the use of probiotics to improve non-surgical outcomes after periodontal debridement, and randomized clinical trials published over the past decade have demonstrated the potential of probiotics. However, conflicting results have obfuscated its clinical efficacy and an appraisal of enough stringently selected studies is lacking. Limitations of previous reviews include a limited number of included studies [45–47], heterogenous study inclusion criteria (e.g. probiotics as monotherapy) and the selection of studies with a lack of blinding [12,46–48]. Thus, the present study seeks to investigate the use of probiotics as an adjunct to non-surgical therapy for periodontitis patients, when compared to placebo intervention. Conventional outcomes are used, and novel treatment outcomes for the first time to evaluate the efficacy of probiotics on a site and patient-level.

Materials and methods

Study protocol

This review was conducted according to PRISMA (Preferred Reporting Items for Systematic Review and Meta-analysis) guidelines [49]. The study protocol has been registered on PROSPERO (CRD42020142699).

Focused question

The focused PICO question of this systematic review was 'in the non-surgical management of patients with chronic periodontitis, do adjunctive probiotics compared to placebo improve clinical outcomes?'

Inclusion criteria—type of studies

Randomized controlled trials in humans using probiotics as an adjunct to treat periodontitis, with a baseline and at least one follow-up visit were included.

Type of participants (study population)

Systemically healthy adults ($\geq$18 years old) diagnosed with periodontitis.

Treatment groups

The intervention group consisted of patients receiving non-surgical therapy with adjunctive probiotic therapy. No restrictions were placed on type of probiotic or method of administration. The comparator group consisted of patients receiving non-surgical therapy with a placebo.

Outcomes

The primary outcome chosen for this study was percentage change of the total number of deeper sites ($\geq$5 mm, $\geq$6 mm, $\geq$7 mm) before and after therapy. Secondary outcomes included change in mean pocket probing depth (mm), percentage patients in need for additional therapy [50,51], risk for disease progression [52], microbiological, and immunological results.

Exclusion criteria

Non-randomized clinical trials and non-blinded randomized clinical trials without use of a placebo were excluded. Studies reporting on less than 10 patients per treatment group, patients with systemic compromise, absence of

periodontal therapy, study duration less than three months, and articles not in English were excluded.

Search methods for identification of studies

An electronic search of the databases PubMed (Medline), EMBASE, and Cochrane Register of Controlled Trials (CENTRAL) was conducted in Feb 2020, with the last update in June 2020. Additionally, bibliographies of all studies included were screened. The detailed search strategy can be found in Supplementary material S1.

Study selection

Search results were imported into EndNote x8 (Thomson Reuters, New York City, NY) and duplicates were removed. Titles and abstracts were independently screened for relevance by two reviewers (EN and JT) for potential inclusion. The full article was reviewed if a decision could not be made by screening the title and abstract alone. Full text of selected articles was carried out for the final selection of studies. Any disagreement was resolved by discussion until a consensus was obtained.

Data extraction

Data collection was done using data collection forms designed to extract study characteristics and outcomes, as well as to carry out methodological quality assessment. Data were independently extracted by EN and JT, who were not blinded to the authors and institution of the studies undergoing review. In cases of missing or unclear information, an attempt was made to contact the corresponding author of included reports.

Quality assessment

Risk of bias was independently assessed by EN and JT using a checklist of items from the Cochrane Handbook [53]. The following domains were assessed: randomization, effect of assignment to intervention, missing outcome data, measurement of outcome, and selection of reported result. Risk of bias for the individual domains was categorized as low, some concerns, or high according to the algorithm suggested by Higgins et al. and an overall risk of bias was assigned for each study. Overall, a study was at low risk of bias if all domains were low risk, some concerns if at least one domain had some concerns but no domain was at high risk, and high risk if at least one domain was at high risk of bias or had some concerns for multiple domains. Test product and industry funding was also evaluated, and any disagreements between reviewers were resolved through discussion.

Summary measures

The primary outcome was the percentage change of the total number of deeper sites before and after therapy, as assessed on three levels (PPD $\geq$5 mm, $\geq$6 mm, $\geq$7 mm). Secondary outcomes included change in mean pocket probing depth (mm) at three time points (3, 6, and 12 months), percentage patients in need of additional therapy [50,51], risk of disease progression [52], microbiological and immunological parameters.

Data synthesis and statistical analysis

Extracted data were presented into tables to assess the quantity of data, and to determine its suitability for further quantitative analysis. Meta-analysis was performed to estimate weighted mean differences with 95% confidence interval for the primary and three of the four secondary outcomes. The random-effects model was performed to allow for differences in treatment effect from study to study. Where mean change and mean SD were not provided, they were estimated using baseline and end mean values with a correlation coefficient of 0.9 [53]. Odds ratios were calculated for the outcomes for patients in need of additional therapy and risk for disease progression. The statistical heterogeneity between studies was assessed using Cochran's Q test and I^2 statistic. The I^2 values of 25%, 50%, and 75% were considered as low, moderate, and high level of heterogeneity, respectively. A narrative approach was chosen for the other two secondary outcomes (microbiological and immunological results). Review Manager (RevMan, Computer program, Version 5.3. Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2014) was used for data analysis, and a p value $<.05$ was considered as statistically significant. Inter-reviewer agreement for study selection and quality assessment was assessed *via* Cohen's κ scores. Sensitivity analyses were conducted to assess the effect of outliers or studies at high risk of bias.

Results

Study selection

A total of 818 studies were identified. After removal of duplicates, 20 were screened for full text. A further 10 studies were excluded and the reasons for exclusion are provided in Supplementary material S2. Finally, 10 studies were included for qualitative analysis and nine for meta-analysis (Figure 1). Inter-rater agreement for selection of studies at the title and abstract level was high with a κ of 0.89 (95% CI 0.79–1.00).

Study characteristics

All 10 included studies were double-blinded, randomized, placebo-controlled clinical trials. Most were conducted in a University setting. The general characteristics of included studies are summarized in Supplementary material S3. Four studies were performed in Turkey, two in Chile, two in Brazil, one in India, and one in Hong Kong. All studies featured systemically healthy adult subjects diagnosed with chronic periodontitis. Two studies included a small number of smokers; however, these were evenly distributed between test and

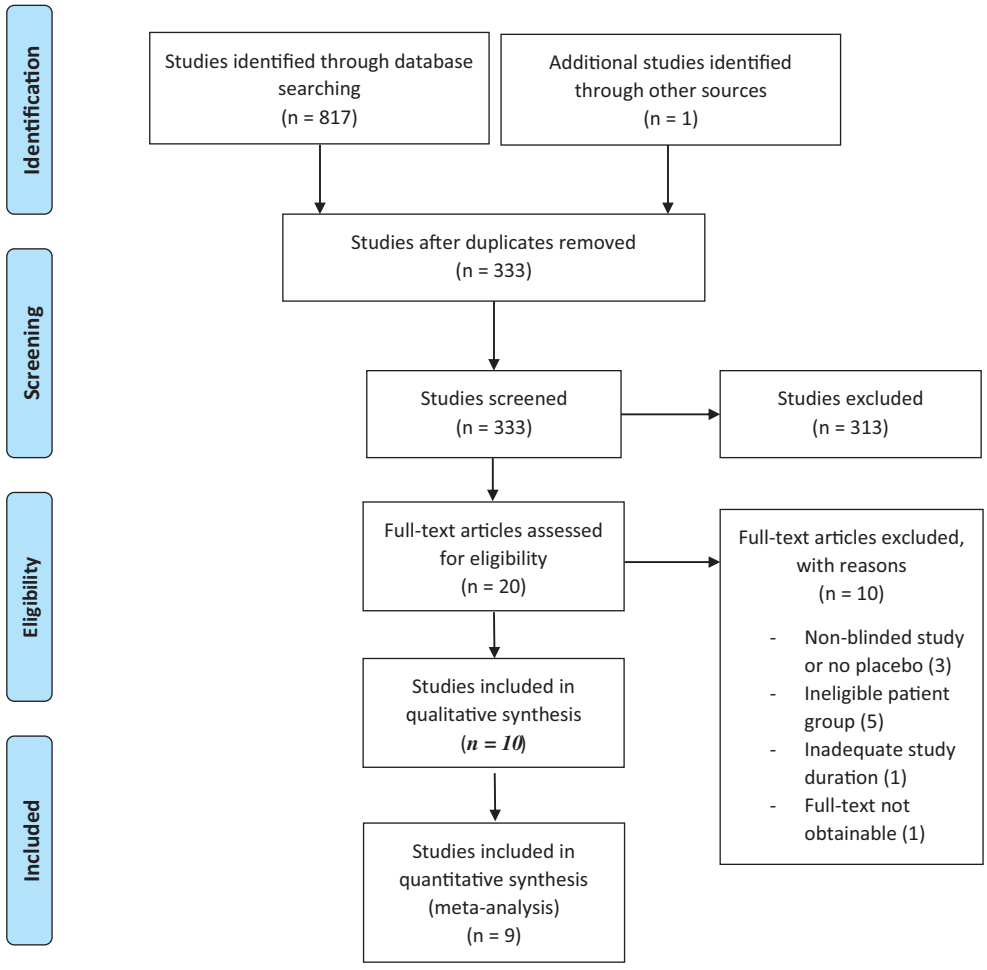

Figure 1. PRISMA flowchart showing the study selection process.

control groups [54,55]. The mean age of participants ranged from 42 to 57 years of age, although one study did not report this detail [56]. The evaluation period of studies ranged from 3–12 months. Included patients received standard non-surgical periodontal therapy and oral hygiene instructions, and either probiotic or placebo. One study supplemented their non-surgical debridement with xylitol [57], one included subgingival delivery of probiotics [58], and two employed a full mouth disinfection protocol [51,59,60]. The probiotic administration protocol of included studies is summarized in Supplementary material S4. All the studies started probiotic therapy immediately after root surface debridement was completed except for two studies, which started at the onset of periodontal therapy [42,61].

Response rate, compliance, and adverse events

The response rate ranged from 69.5–100% and the dropouts were reported from two studies [58,62]. All but two studies [57,58] reported checking for compliance with the probiotic or test drug. Eight studies reported a high compliance rate of 80–100% to the probiotic regime. Adverse events were not associated with probiotic consumption during the study duration, although one study did not report on this parameter [57].

Percentage change of the total number of deeper sites before and after therapy

Meta-analysis of five studies revealed a combined nonsignificant benefit of −4.33% (95% CI [−12.33 to 3.67], $p = .29$) for sites $\geq$5 mm when probiotics were used (Figure 2(a)). This benefit remained nonsignificant when analysis was performed on deeper pockets $\geq$6 mm (WMD = −2.19%, 95% CI [−7.48 to 3.09], $p = .42$) and $\geq$7 mm (WMD = −0.15%, 95% CI [−1.60 to 1.30], $p = .84$) (Figure 2(b,c)). Considerable heterogeneity was observed. Among the five studies, three studies provided 3-month data [51,56,62] and two studies only published 12-month data [54,61]. Separate analysis on 3-month and 12-month data revealed no significant benefits for all three levels of deep probing depth (Supplementary material S5 and S6).

Overall change in mean pocket probing depth (PPD)

Analysis of mean probing depth reduction was performed on three levels—3, 6, and 12 months. One study [58] could not be included for further analysis due to insufficient information, and an attempt to contact the authors was unsuccessful. Meta-analysis of nine studies revealed a significant benefit in mean PPD reduction at 3 months (WMD=

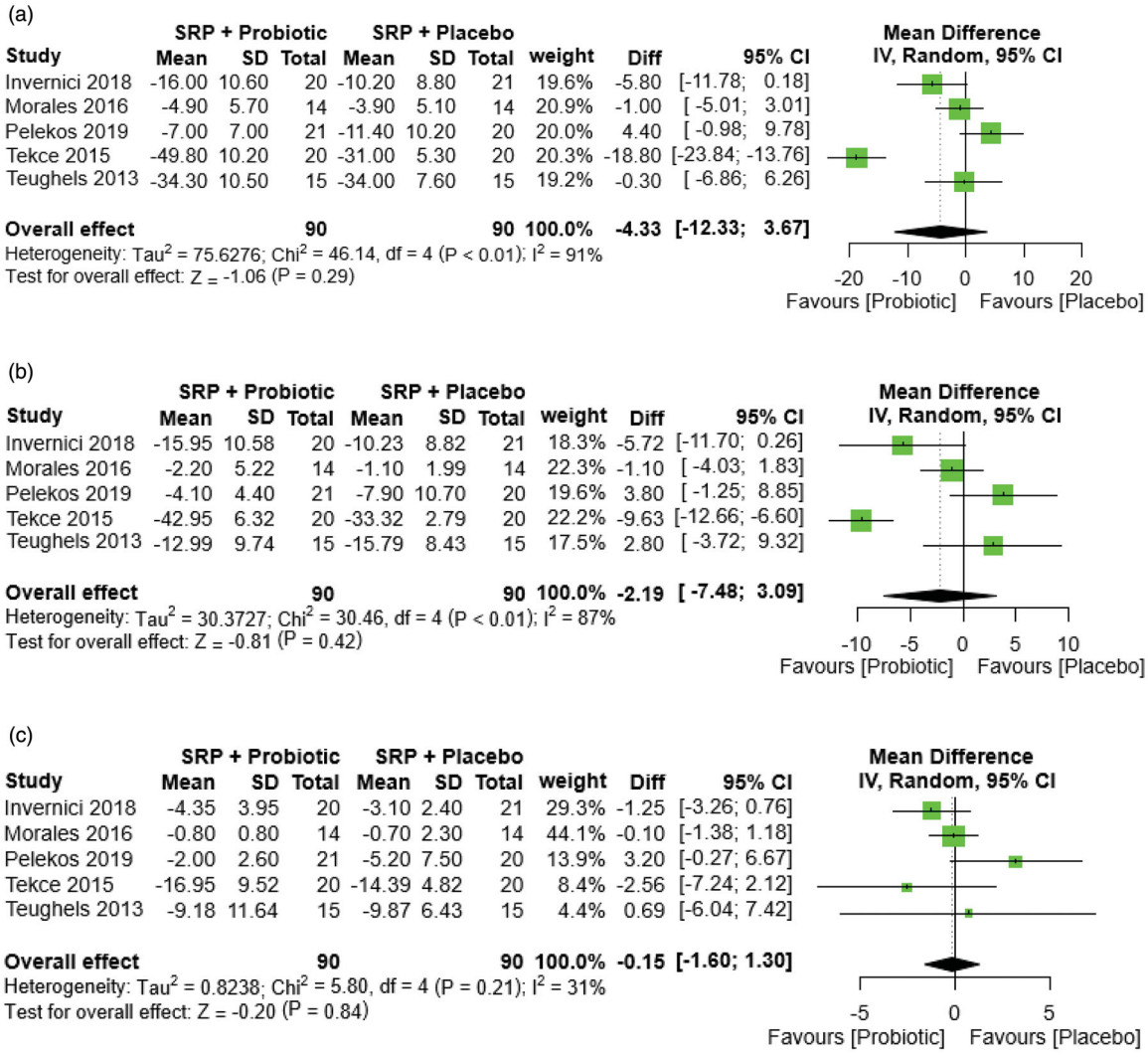

Figure 2. Forest plot: percent change in the remaining number of deeper sites (combined 3- and 12-month data). (a) ≥5 mm, (b) ≥6 mm, (c) ≥7 mm.

–0.26 mm, 95% CI [–0.48 to –0.04], $p = .02$) (Supplementary material S7). However, sensitivity analysis was performed excluding one study [57] due to high risk of bias. Meta-analysis of the remaining eight studies revealed that compared with placebo, treatment with probiotics resulted in a nonsignificant ($p = .08$) benefit in mean PPD reduction of –0.18 mm (95% CI [–0.39 to 0.02]) (Figure 3(a)). Inter-study heterogeneity remained significant ($\chi^2 = 49.02$, $I^2 = 86\%$). Six studies provided outcomes at six months. No significant differences were found between treatment with probiotics and placebo ($p = .22$) (Figure 3(b)). Three studies were included in the meta-analysis for 12-month outcomes. A weighted mean difference of –0.83 mm (95% CI [–1.49 to –0.18], $p = .01$) was found, but high heterogeneity was observed ($\chi^2 = 30.44$, $I^2 = 93\%$) (Figure 3(c)).

Patients in need of additional therapy (0–2 sites compared to ≥3 sites)

The use of adjunctive probiotics had an odds ratio of 0.19 (95% CI [0.07 to 0.56], $p = .002$), or an 81% lower chance, of having ≥3 sites requiring additional therapy (Figure 4(a)). This result is based on sensitivity analysis, after exclusion of one study [61], where all 20 patients using probiotics fell in the 0-2 sites category and all 20 patients using placebo fell into the ≥3 sites requiring additional therapy category. Meta-analysis including this study demonstrated an odds ratio of 0.05 (95% CI [0.00–0.64], $p = .02$) or a 95% lower chance of having ≥3 sites requiring additional therapy (Supplementary material S8). There was no heterogeneity after exclusion of this study ($\chi^2 = 0.72$, $I^2 = 0\%$), and significant heterogeneity otherwise ($\chi^2 = 9.03$, $I^2 = 78\%$).

Risk of disease progression at the end of therapy

An odds ratio of 0.32 (95% CI [0.14–0.73], $p = .006$) in favor of probiotics was observed, indicating that adjunctive use of probiotics is associated with a 68% lower chance of being high risk *vs.* low/medium (Figure 4(b)). No significant heterogeneity was observed ($\chi^2 = 2.56$, $I^2 = 0\%$).

Microbiological and immunological results

Six studies evaluated microbiological findings [51,55,56,58,60,61], with all except two studies using cell cultures in

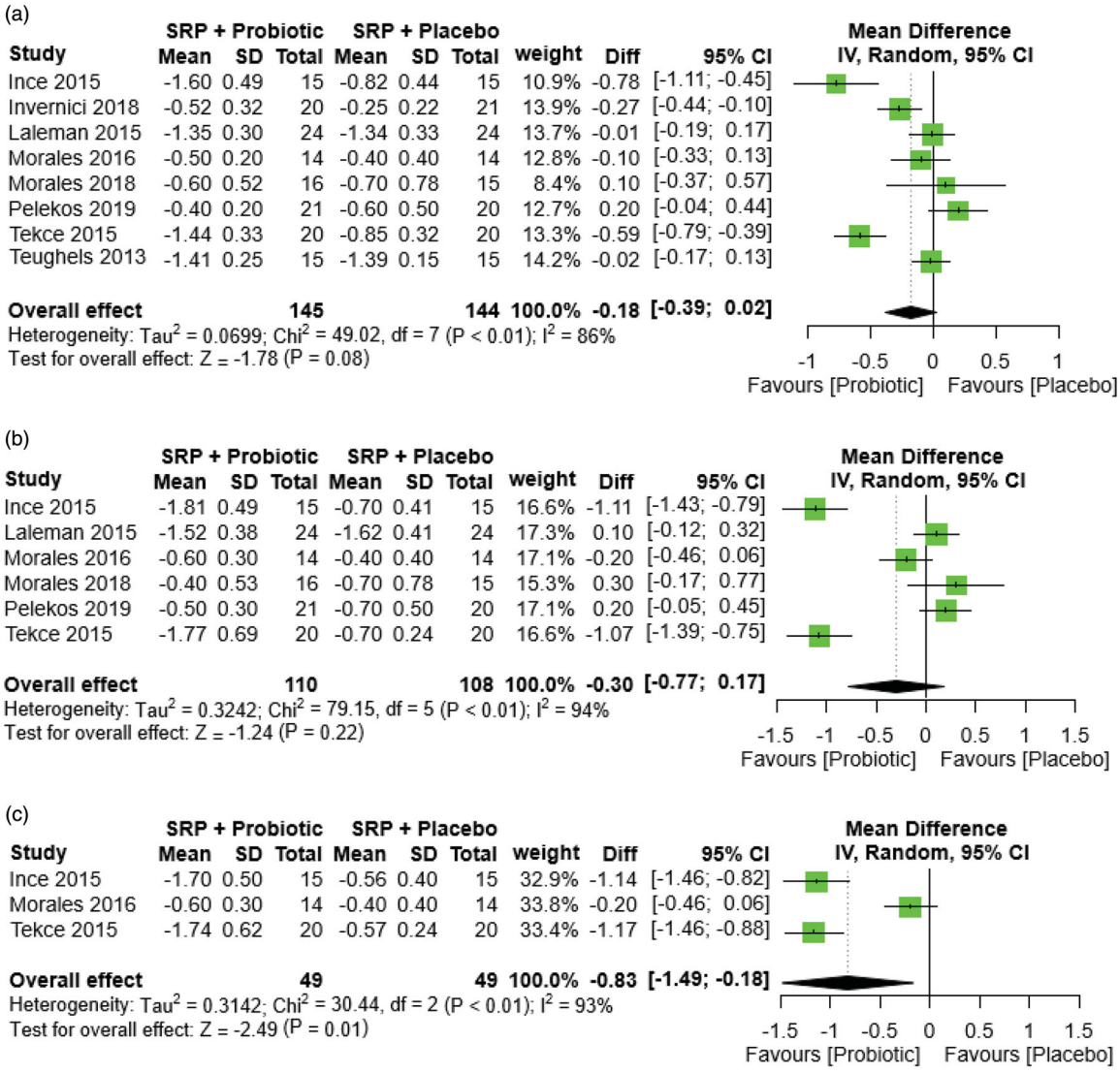

Figure 3. Forest plot: change in mean probing pocket depth (a) 3 months, (b) 6 months, (c) 12 months.

Supplementary material S9. All studies collected subgingival plaque samples, and two studies also examined supragingival plaque, tongue biofilm, and saliva samples [51,60]. A significant difference was only reported in some studies, and periodontal pathogens were not uniformly reduced across these studies.

Two studies investigated immunological responses with ELISA [42,56]. One study [56] showed a higher concentration of IL-1β and IL-8 levels at 30 days after periodontal therapy in the control group compared to the test group, while another study [42] showed that treatment with probiotics led to a greater decrease in gingival crevicular fluid volume and MMP-8 concentrations, and a concomitant greater increase in TIMP-1 in the test group compared to the control group. However, these immunological parameters returned to baseline values in both groups after 360 days.

Quality assessment

Risk of bias of included studies is presented in Supplementary material S10. Most studies had an overall low risk of bias, two had some concerns, and one was at high risk. A κ coefficient of 1.00 (95% CI 1.00–1.00) for overall risk of bias was found, indicating perfect agreement in quality assessment. Seven studies received industry support [42,51,56,57,60–62], although it was reported that the company was not involved in data management.

Discussion

Summary of main findings

The present systematic review aimed to evaluate the efficacy of probiotics when used as an adjunct to periodontal debridement. Selective criteria were used for the inclusion of clinical trials in this study, and this resulted in the inclusion of nine relevant randomized controlled trials for meta-analysis. Most studies evaluated the use of *Lactobacillus* species, and lozenges were the most common delivery vehicle.

A meta-analysis of five studies did not show a significant beneficial reduction in percentage change of the total number of deeper sites before and after therapy when adjunctive probiotics were used, and this remained true even when

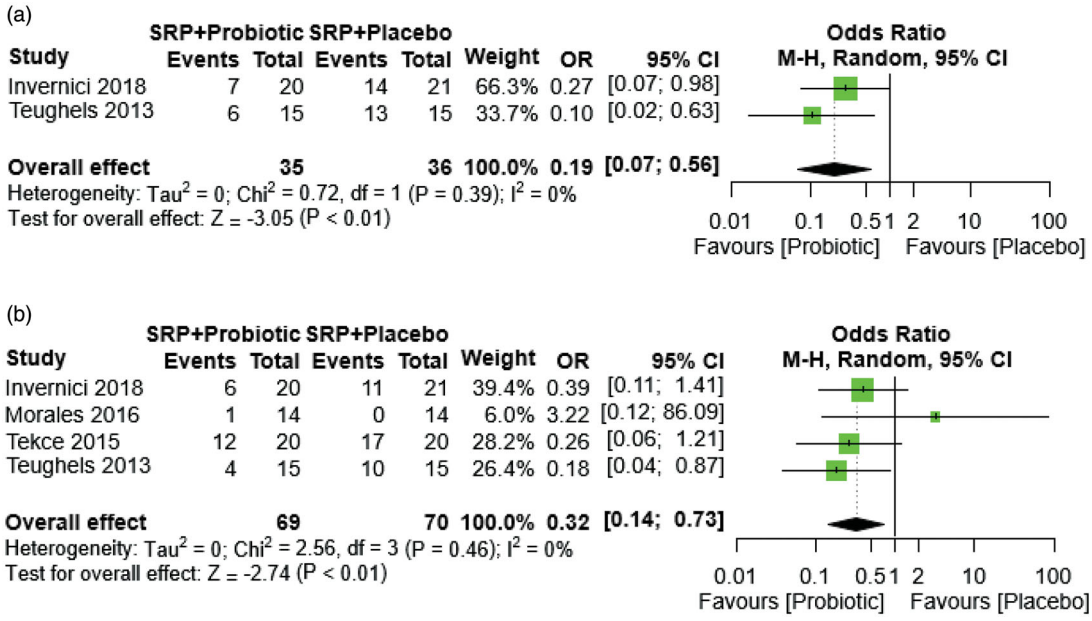

Figure 4. Forest plot: secondary outcomes. (a) Odds ratio for patients in need of additional therapy, 0–2 compared to ≥3 sites. (b) Odds ratio for risk of disease progression at the end of therapy, low/medium compared to high risk.

deeper thresholds were analyzed. Change in mean probing depth was analyzed at three time points. No significant difference between probiotic and placebo groups was observed at three months (eight studies) and six months (six studies), although a significant difference was noted at 12 months (three studies). It should be noted, however, that two of the three studies with a 12-month follow-up consistently reported a large mean difference between probiotic and placebo compared to the other studies.

The concept of 'need for additional therapy' was based on the 11-year maintenance results by Matuliene et al. [63], who reported that a pocket probing depth of 6 mm was a risk factor for disease progression and tooth loss, and thus represented an incomplete periodontal outcome. Three studies reported on this outcome, previously defined as a site with a pocket probing depth ≥5 mm with bleeding or ≥6 mm [50,51]. After sensitivity analysis, the odds of a patient having 0–2 sites in need of addition therapy compared to ≥3 sites were significantly lower with the use of adjunctive probiotics.

Risk of disease progression at the end of therapy was assessed by some included studies using the Periodontal Risk Assessment by Lang and Tonetti [52]. Patients in the high-risk group have been shown to suffer from a higher rate of tooth loss [64]. Four studies reported on this outcome, and adjunctive probiotics were found to confer a protective odds ratio from a patient being at high-risk at the end of therapy.

Microbiological outcomes were reported in six studies. However, heterogeneity in bacteria identification method and bacteria tested meant that a meta-analysis for this parameter was not possible. While the use of adjunctive probiotics could modulate the oral bacteriome, data reported by existing studies are not consistent. Of interest are the results of one study which reported the presence of *Lactobacillus Reuteri* probiotic in addition to periodontal pathogens. The probiotic was present at 90 days but no longer detectable at 180 days [61], perhaps indicating that its effects may not last more than a few months. Two studies reporting on immunological parameters suggest that adjunctive probiotics appear to favor a state of inflammation resolution and less periodontal breakdown.

Compliance with probiotic therapy in a clinical trial setting appears to be high, and this ranged from 80–100% [60,62]. None of the included studies reported any associated adverse events, thus probiotics may be considered a safe intervention in systemically healthy patients with chronic periodontal disease.

Quality of included studies

This study also assessed the quality of evidence, patient compliance, and adverse effects. Seven studies were of high quality with an overall low risk of bias. Two studies had some concerns and this was due to inadequate analyses in the presence of dropouts in one study [58] and insufficient information provided on whether outcome assessors were aware of the intervention received by study participants [61]. One study was at high risk of bias because of baseline differences between intervention groups, insufficient information provided on the awareness of assigned intervention of participants or carers, and the likelihood that outcome assessment could have been influenced by knowledge of intervention received [57]. This study was therefore not included in the meta-analysis. Though most studies received industry support, some of these trials reported equivocal findings [60,62].

Comparison with previous reviews or studies

This study found that adjunctive probiotics do not confer additional benefits for deeper sites, which is not in

agreement with other studies that suggested deeper pockets and higher baseline disease severity may benefit more [45,47,65]. The difference in results could be due to differences in study selection, number of included studies, and parameters analyzed. In this study, percentage change of the total number of deeper sites before and after therapy was used rather than the use of mean probing depth values for the respective categories. The site-level findings in this study are more similar to another systematic review and meta-analysis on host-modulators in periodontal therapy, which concluded that administration of probiotics conferred limited clinical benefits which were not clinically significant [66]. However, this study also only assessed mean probing depth changes overall. In the present study, assessment of novel treatment outcomes suggest benefits on a patient-level. This may be more relevant to clinical practice, as it is the patient who may develop recurrent periodontitis, who has to decide whether to proceed with periodontal surgery, and who ultimately assesses the benefits of further treatment with regard to their individual quality of life [67]. We observed a lack of robustness of microbiological data, and this agrees with other reviews which found no significant effect on bacteria [65,68,69]. Our observations on immunological findings are consistent with others in the literature, where probiotics have been shown to reduce the pro-inflammatory cytokine response in tandem with improved clinical parameters [70,71].

Strengths and limitations

This systematic review and meta-analysis has several strengths and limitations. The most significant is the use of percentage change of the total number of deeper sites before and after therapy as a primary outcome for the first time to assess the efficacy of probiotics, rather than generalizing their effectiveness from changes in mean probing depth. Stringent selection of studies was also carried out, and our review was restricted to trials with a minimum duration of three months. Included studies were stratified according to the time points reported, thus minimizing loss of data or effectiveness of the intervention in between baseline to the last follow-up. Meta-analysis was carried out on secondary outcomes that could have more clinical relevance, such as 'need for additional therapy' and 'risk of disease progression'. Sensitivity analyses were also performed to evaluate the robustness of data.

There are also several limitations, both in our review and in the reported literature. Different probiotics were pooled together and compared to placebo and it is therefore not possible to make definitive conclusions on the individual efficacy of the respective probiotics used and these results cannot be generalized. Significant heterogeneity was present, and this could be explained by various factors related to study design, including the non-surgical therapy protocol, disease severity, probiotic administration protocol (dose, duration of administration, delivery vehicle), duration and frequency of follow-up, and analyses performed. It was not within the scope of this study to propose the most appropriate administration protocol for probiotics. How much probiotic reaches the bottom of the pocket, and whether this colonizes the biofilm at a significant level to provide modulatory effects remains unclear. Finally, the method of power calculation was also very heterogenous and many studies based this on the pilot study of Vivekananda et al. [72]. However, a *post hoc* power calculation by Teughels et al. at the conclusion of his study suggested that at least 63 subjects are required per group [51]. It is therefore likely that many of these studies are underpowered.

Conclusion

With global concerns for the judicious use of antibiotics mounting, the prospect of probiotics replacing antibiotics as an intervention is appealing. This study aimed to clarify the relevance of results published over the last decade, and their translatability to daily clinical practice. Based on the studies included in this review, and within the limitations of this study, adjunctive probiotic therapy on a site-level did not demonstrate statistically significant greater reductions to the percentage of remaining deeper sites or mean probing depth after therapy. However, a subset of studies indicates beneficial odds ratios on a patient-level, for need for additional therapy and risk of disease progression. These results should be considered when evaluating the clinical relevance of probiotic therapy.

Clinical implications

As demonstrated by most of the included studies, adjunctive probiotics should only be used after the completion of adequate debridement. Adjunctive probiotic therapy is safe in systemically healthy individuals and could offer additional benefits in reducing risk of disease progression and need for additional therapy. Therefore, its use can sometimes be justified. However, the observed heterogeneity of treatment effect precludes definitive recommendations for clinical practice.

Future research

More appropriately powered randomized controlled trials will be beneficial to further clarify the efficacy of adjunctive probiotics and their mode of action. Ideally, these should be accompanied with at least a 6-month follow-up, with outcome measures at baseline, 6 months, and an intermediate time point. Clinically relevant parameters as outlined in this study, and patient-level outcome measures should be assessed as well. Correlation of 16S rRNA or cytokine analysis with clinical findings will be useful to better understand the mechanisms of probiotic therapy.

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Disclosure statement

This study was self-funded by the authors. No potential competing interest was reported by the authors.

Author contributions

EN contributed to conception, design, data acquisition and interpretation, and drafted and critically revised the manuscript. JT contributed to conception, design, data acquisition and interpretation, and critically revised the manuscript. SS contributed to data acquisition, analysis, and interpretation. LL, CK, and MO contributed to data interpretation and critically revised the manuscript. All authors gave their final approval and agreed to be accountable for all aspects of the work.

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