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Probiotics and the Treatment of Peri-implant Diseases: a systematic review

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Probiotics and the treatment of peri-implant diseases: a systematic review.

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“Work gives you meaning and purpose and life is empty without it.” Stephen Hawking

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To you, my mother, I dedicate this work. I hope I can achieve all my goals and I wish you help me overcome any difficulties that may arise.

I love you.

RESUMO

Introdução/Objetivo

Os probióticos são microrganismos benéficos que desempenham certas funções no hospedeiro, e estudos mostram que os benefícios à saúde que eles podem oferecer vão além do intestino. O objetivo desta revisão sistemática foi identificar associações suportadas por evidências atuais entre os probióticos e o tratamento de doenças peri-implantares.

Materiais e Métodos

As bases de dados eletrônicas PubMed, Web of Science e Scopus foram pesquisadas até 25 de novembro de 2019. Esta revisão explorou estudos que relataram a avaliação do efeito clínico dos probióticos numa população de pacientes com implantes com mucosite peri-implantar e peri-implantite. A pergunta (PICO) abordada foi: "O uso de probióticos no tratamento de doenças peri-implantares melhora o seu *outcome* quando comparados com os tratamentos convencionais?"

Resultados

A análise de 203 títulos/resumos selecionou 8 artigos para a leitura em texto completo. 2 artigos foram posteriormente excluídos por não preencherem os critérios de inclusão. Um total de 6 estudos foram incluídos nesta revisão sistemática e todos eles foram ensaios clínicos randomizados que avaliaram a aplicação de probióticos em implantes orais. Metade dos estudos incluídos nesta revisão mostrou um efeito benéfico adicional significativo dos probióticos nas doenças peri-implantares, embora inconsistente. 3 estudos apresentaram uma melhoria na saúde peri-implantar, com uma redução significativa nos parâmetros clínicos (1 estudo no índice de placa e na profundidade de sondagem, $p=0.001$; outro no índice de placa, $p<0.001$; e outro estudo no sangramento à sondagem e à profundidade de sondagem, $p=0,024$). Os restantes

três estudos incluídos não relataram diferenças estatisticamente significativas consistentes entre os grupos de teste e controlo. Houve uma heterogeneidade metodológica significativa nos estudos incluídos sobre as definições de caso e controlo e uma existência de fatores de confusão que podem afetar os resultados do estudo.

Conclusão

Devido aos resultados heterogêneos encontrados no presente estudo, pode-se concluir que as evidências científicas atuais são insuficientes para apoiar a eficiência dos probióticos na saúde peri-implantar.

PALAVRAS-CHAVE

Probióticos; Peri-implantite; Implante dentário; Mucosite peri-implantar; Doenças peri-implantares; Terapia peri-implantar; Saúde Periodontal

ABSTRACT

Background/ Objective

Probiotics are beneficial microorganisms that perform certain functions in the host, and studies show that the health benefits they can offer extend beyond the gut. The aim of this systematic review was to identify associations supported by current evidence between probiotics and the treatment of peri-implant diseases.

Methodos

The electronic databases PubMed, Web of Science and Scopus were searched, up to 25th of November, 2019. This review explored studies that reported evaluation of the clinical effect of the probiotics in a population of patients with implants with peri-implant and peri-implantitis mucositis. The addressed (PICO) question was: "Does using probiotics to treat peri-implant diseases improve your outcome compared with conventional treatments?"

Results

The screening of 203 titles/abstracts selected 8 papers for full-text reading. 2 papers were subsequently excluded because did not fullfil the inclusion criteria. A total of 6 studies were included in this systematic review and all of them were randomized controlled trials that evaluated the application of probiotics in oral implants. Half of the studies included in this review showed a significant additional beneficial effect of probiotics on peri-implant diseases, although inconsistent. 3 studies showed an improvement in peri-implant health, with a significative reduction in clinical parameters (one study in plaque index and probing pocket depth, $p=0.001$; another one in plaque index, $p<0.001$; and another study in bleeding on probing and probing pocket depth, $p=0.024$). The remaining 3 included studies were unable to report consistent statistically significant differences between test and control groups. There was a significant methodological heterogeneity in the included studies regarding case and control definitions and an existence of confounding factors that may affect the study results.

Conclusions

Due to the heterogeneous results found in the present study, it can be concluded that the current scientific evidence is insufficient to support the efficiency of probiotics upon peri-implant health.

KEY-WORDS

Probiotics; Peri-implantitis; Dental implant; Peri-implant mucositis; Peri-implant diseases; Peri-implant therapy; Periodontal Health

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INDEX OF ABBREVIATONS

BOP	Bleeding on probing
BOPI	Bleeding on probing at the implant level
CHX	Chlorhexidine gluconate
CCL5 C-C	Chemokine ligand 5
FMPI	Full mouth plaque index
FMBOP	Full mouth bleeding on probing
FMBS	Full mouth bleeding score
FMPS	Full mouth plaque score
GCF	Gingival crevicular fluid
GI	Gingival index
GM-CSF	Granulocyte macrophage-colony stimulating factor
IL-1β	Interleukin-1β
IL-17	Interleukin 17
IL-10	Interleukin 10
IL-1RA	Interleukin-1 receptor antagonist
IL-4	Interleukin 4
IL-6	Interleukin 6
IL-8	Interleukin 8
IFN-γ	Interferon gamma
MD	Mechanical debridement
Mo	Months
MSBI	Modified sulcus bleeding index

MGI	Modified gingival index
MMPs	Matrix metalloproteinases
PI	Plaque index
PII	Plaque index at the implant level
PPD	Probing pocket depth
PPDI	Probing pocket depth at the implant level
PICF	Peri-implant crevicular fluid volume
PT	Probiotic therapy
TNF-α	Tumor necrosis factor-alpha
TIMPs	Tissue inhibitors of metalloproteinases
Th1	T helper type 1
Th2	T helper type 2
Th17	T helper type 17
Wk	Weeks

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INTRODUCTION

Periodontal biofilm is an organized consortium of bacteria surrounded by a complex matrix aggregate formed by polysaccharides and proteins. Biofilm adheres to the teeth surface and is considered a determining factor for caries and periodontal disease (1).

In health, biofilms are well-balanced with the host defences and no damage occurs. The unbalance between the periodontal microbiota and the host immune system leads to a dysbiosis. As a result, predominantly gram-positive, non-mobile, aerobic and facultative health-compatible microbiota shifts to essentially gram-negative, mobile and anaerobic disease-related microbiota (2). In susceptible individuals, chronic inflammation due to dysbiotic biofilms leads to progressive tooth attachment loss (for example periodontitis) and ultimately tooth loss.

Dental implants are used to replace missing teeth and have gained a lot of popularity considering their numerous advantages over other possibilities for oral rehabilitation. However and inevitably, periodontal biofilms will colonize any prosthetic solution for dental replacement, leaving us with a new version of and old problem, as we see happen in peri-implantitis (3). Infection is one of the factors responsible for dental implant failure and the microbiota traditionally associated with dental implant failure is the same as that associated with periodontitis (2).

Biofilm-related peri-implant diseases can be divided into peri-implant mucositis and peri-implantitis. Peri-implant mucositis is defined as a reversible inflammatory reaction of the soft tissues surrounding endosseous implants, without loss of supporting bone (4-8). Clinical features of peri-implant mucositis include bleeding on probing (BOP) and erythema. Swelling and/or suppuration may also be present (9). BOP with a slight force (<0.25N) is a key criterion for the diagnosis (4).

Is the accumulation and consequent adhesion of the bacterial plaque to the surface of the dental implant that causes the inflammatory host response, which when left untreated extends and can invade the supporting alveolar bone - and peri-implantitis appears - ultimately leading to failure and irreversible loss of the dental

implant (10, 11). In this way, it is considered that dental plaque, combined with poor oral hygiene, is the major etiological factor for the occurrence of peri-implant diseases (9). However, the individual response to the bacterial accumulation varies between patients. Research has shown that there is an increased risk for patients diagnosed with severe periodontitis, with poor plaque control and when the monitoring and evaluation after dental implant placement is poor. There are also reports that smoking and systemic diseases (as diabetes mellitus) are potential risk factors for peri-implant mucositis and peri-implantitis (8, 9).

Treatment of peri-implant mucositis may include all procedures that decrease the amount of periodontal microorganisms, such as self-performed oral hygiene, non-surgical mechanical debridement (MD) and MD combined with antimicrobial agents (chlorhexidine (CHX), triclosan and/or hydrogen peroxide) and/or topic and systemic antibiotics (6, 8).

According to a recent systematic review, the incidence of peri-implant mucositis and peri-implantitis is 19-65% and 1-47%, respectively, whereas the mean prevalence of peri-implant mucositis and peri-implantitis is 43% and 22%, respectively (12).

Although experimental studies in humans have shown that non-surgical MD is efficient in resolving peri-implant mucositis, the role of the complementary antiseptic solutions, as well as the use of antibiotics, remains inconclusive (5). Regarding to peri-implantitis, both the use of local antiseptics and the use of antibiotics can improve clinical parameters, although they do not fully resolve the peri-implant disease, requiring the application of surgical techniques, concomitantly (5, 13). Concerning to the use of antibiotics, the increase bacterial resistances worldwide and the misuse of antibiotics raise global public health concerns that cannot be neglected, propelling us to look for other equally effective treatment alternatives (12, 14-17).

The dysbiosis concepts support a new perspective about periodontal/peri-implant infections where the goal is no longer eliminate periodontal microbiota, but instead, driving the biofilm to a health-compatible status. The use of probiotics seams tailored to this purpose and several studies have been conducted. (12)

In recent years, many definitions have been assigned to probiotics. The World Health Organization defines them as "*live microorganisms that when administered in adequate amounts, confer a health benefit on the host*" (4, 6, 7, 11-13, 15, 16, 18-32).

Originally, probiotics were introduced by Lilly & Stillwell in 1965 as "*substances produced by microorganisms which promote growth of other microorganisms*" (18, 20). The use of probiotics in oral microbiota was recently suggested but, in order to take place, the survival of harmless bacteria on oral environment is required just like their firm adherence to saliva coated surfaces in order to colonize, proliferate and exert their effect by inhibiting oral pathogens. (10, 14). These harmless bacteria are easily accessible to the population, existing in products for daily consumption (mainly fermented milks), food supplements in tablet forms or in soft drinks (14).

Although the mechanisms of action of probiotics are not completely understood, studies have shown that probiotics' action can be observed in the oral biofilm modification, alteration of bacteria colonization, decrease of cytokines inflammatory levels, reduction of pathogenic microorganisms and improvement of periodontal clinical signs (13, 19, 33-35). However, probiotics' success has been demonstrated to depend on the applied bacterial strain (s), microbial concentration, targeted disease and number of species (11).

Within the mechanisms of action of probiotics, they are able to inhibit periodontal pathogens by producing antimicrobial defence agents (such as bacteriocins, lactic acid and hydrogen peroxide) and have the potential to neutralize the pH of the oral cavity, preventing the formation of organized bacterial plaque and thus prevent oral diseases (10, 18, 26). They also can produce antiviral and antifungal agents, which suppress oral pathogens (10).

Other advantages of probiotics' use are the increase of innate immunity and the modulation of pathogen-induced inflammation by producing anti-inflammatory cytokines (TNF-α (Tumor necrosis factor-alpha), IL-1β (Interleukin-1β), IL-17 (Interleukin-17) and activating T helper cells - Th1 (T helper type 1); Th2 (T helper type 2) and Th17 (T helper type 17) or T regulatory cells that induces IL-10 (Interleukin-10) production which is an anti-inflammatory cytokine) (15, 26, 28, 29, 33, 35, 36).

Probiotics can be grouped into moulds, yeasts or bacteria (predominantly) and exist in four known species: **a)** Lactic acid producing bacteria (LAB): *Lactobacillus*, *Bifidobacterium*, *Streptococcus*; **b)** Non lactic acid producing bacterial species: *Bacillus*, *Propionibacterium*; **c)** Non-pathogenic yeasts: *Saccharomyces*; **d)** Non spore forming and non-flagellated rod or coccobacilli (37).

In order to be successful, probiotics must possess certain features to culminate in measurable beneficial effects on hosts' health. For instance, they must be non-virulent and non-pathogenic, stable under most conditions, stay metabolically active and viable, capable of stimulating the immune system and producing antimicrobial substances (26, 28).

According to the literature, *Lactobacillus* and *Bifidobacteria* are two of the known probiotics capable of changing the oral microbiota composition, exerting anti-inflammatory and antimicrobial properties, acting positively on gingivitis and periodontitis (10, 11, 38). Recent studies have pointed to the potential role of strains of *Lactobacilli* in improving clinical and microbiological parameters and reducing the inflammatory response associated with these oral diseases (6, 7, 25).

Regarding the use of these harmless bacteria for the treatment of peri-implant diseases, there are not yet many studies that evaluate their effects (6, 7, 25).

The aim of the present systematic review is to clarify the probiotics' role in the treatment of peri-implant diseases, namely peri-implant mucositis and peri-implantitis. With this work we hope to help clinicians in their decision-making process about therapeutic options to tackle peri-implant diseases.

MATERIALS AND METHODS

Focused question

The main items for conducting and reporting systematic reviews and meta-analyzes (PRISMA) (39) were followed to answer the focused (PICO) question: "The use of probiotics to treat peri-implant diseases improve the disease outcome when compared with conventional treatments?"

Search strategy

To prepare this review, three researches were conducted using MEDLINE via PubMed, Scopus (Elsevier) and Web of Science (Clarivate analytics) online databases. The databases were searched up to 25th of November, 2019.

Studies were searched using the following keywords: “Probiotics”; "Peri-implantitis"; "Dental implant"; “Peri-implant mucositis”; “Peri-implant diseases”; “Peri-implant therapy”; "Periodontal Health", combined with the Boolean connector "AND"; with the truncation connector (*) and the proximity operators (“” and ()), used whenever appropriate.

The search terms used in the databases were: “Peri-implant * AND probiotics”; “Dental Implant AND Probiotics”; “Peri-implant Mucositis” AND Probiotics ”; ”Peri-implant diseases AND Probiotics”; “Peri-implant * therapy AND probiotics”; “Periodontal health and Probiotics”. Of these keywords, only “peri-implantitis”; “Dental Implant” and “Probiotics” are MeSH (Medical Subject Headings) terms.

Selection criteria

As inclusion criteria, all studies that evaluate the application of probiotics in oral implants were found eligible. We only include articles applied in humans and written in

Portuguese, English or Spanish. Reasons for exclusion were: non-randomized controlled trial articles, pilot studies, reviews, letters and case reports.

Screening methods and data extraction

The titles and abstracts of studies that meet the inclusion and exclusion criteria were evaluated and selected. Any disagreement was resolved after discussion and analysis by a third reviewer (J.P.). Data were extracted from included studies by two independent reviewers (A.M., L.M.) and the following parameters were recorded: author/ year; study design; clinical parameters; sample size; smoking habits; definition of the peri-implant condition; probiotic bacterial strain/ vehicle and regimen; treatment; follow-up; study outcome; adverse events and funding.

Risk of bias in individual studies/ quality appraisal

The assessment of the risk of bias in the included studies was carried out independently by each reviewer (AM, LM) according to the *Scottish Intercollegiate Guidelines Network* (SIGN 50) (40), by completing the checklist for Randomized Controlled Trials (Checklist in annex), following the levels of evidence and degrees of recommendation of the SIGN methodology (Supplemental Table I and II in annex).

The internal validity of each article was scored in 10 questions: "*1. The study addresses an appropriate and clearly focused question? 2. The assignment of subjects to treatment groups is randomized? 3. An adequate concealment method is used? 4. Subjects and investigators are kept 'blind' about treatment allocation? 5. The treatment and control groups are similar at the start of the trial? 6. The only difference between groups is the treatment under investigation? 7. All relevant outcomes are measured in a standard, valid and reliable way? 8. What percentage of the individuals or clusters recruited into each treatment arm of the study dropped out before the study was completed? 9. All the subjects are analyzed in the groups to which they were randomly*

allocated (often referred to as intention to treat analysis)? and, 10. Where the study is carried out at more than one site, results are comparable for all sites?" (40).

For each question we use each of the following items to describe how well it was identified in the study: "*1. Well covered; 2. Adequately addressed; 3. Poorly addressed; 4. Not addressed (for example not mentioned, or indicates that this aspect of study design was ignored); 5. Not reported (i.e. mentioned, but insufficient detail to allow assessment to be made);and, 6. Not applicable*" (40).

The general evaluation of the work, classifying the methodological quality of the study, is based on the answers to the 10 questions, using the following coding system: "*high quality (**++**): If all or most of the criteria have been met; low risk of bias (**+**): If only some of the criteria have been met; and, high risk of bias (**-**): If few or no criteria have been met*" (40).

The code marked in this section, together with the type of study, will decide the level of evidence that this study provides.

Any disagreements were resolved after discussion and analysis.

Statistical analysis

No meta-analysis could be carried out caused by the methodological heterogeneity in the included studies. For that reason, the outcomes are described as a narrative review.

RESULTS

Study selection and flow diagram

In the initial electronic search, 84 studies were located in Pubmed, 87 in Scopus, and 139 in Web of Science (310 papers). After removing 107 duplicate studies, 149 were rejected after the initial screening of titles. By reading the abstract, 46 studies were rejected. The remaining 8 studies fulfilled the inclusion criteria and the full texts were obtained and analyzed. After full text reading, 2 studies (19, 30) were excluded because they were pilot studies. Consequently, a total of 6 articles were considered in this systematic review: Peña M. *et al.* 2019 (4), Alqahtani F. *et al.* 2019 (7), Fernández A. *et al.* 2015 (25), Laleman I. *et al.* 2020 (41), Galofré M. *et al.* 2018 (5), Hallström H. *et al.* 2016 (6).

The PRISMA flow diagram (Figure 1) illustrates the entire selection process.

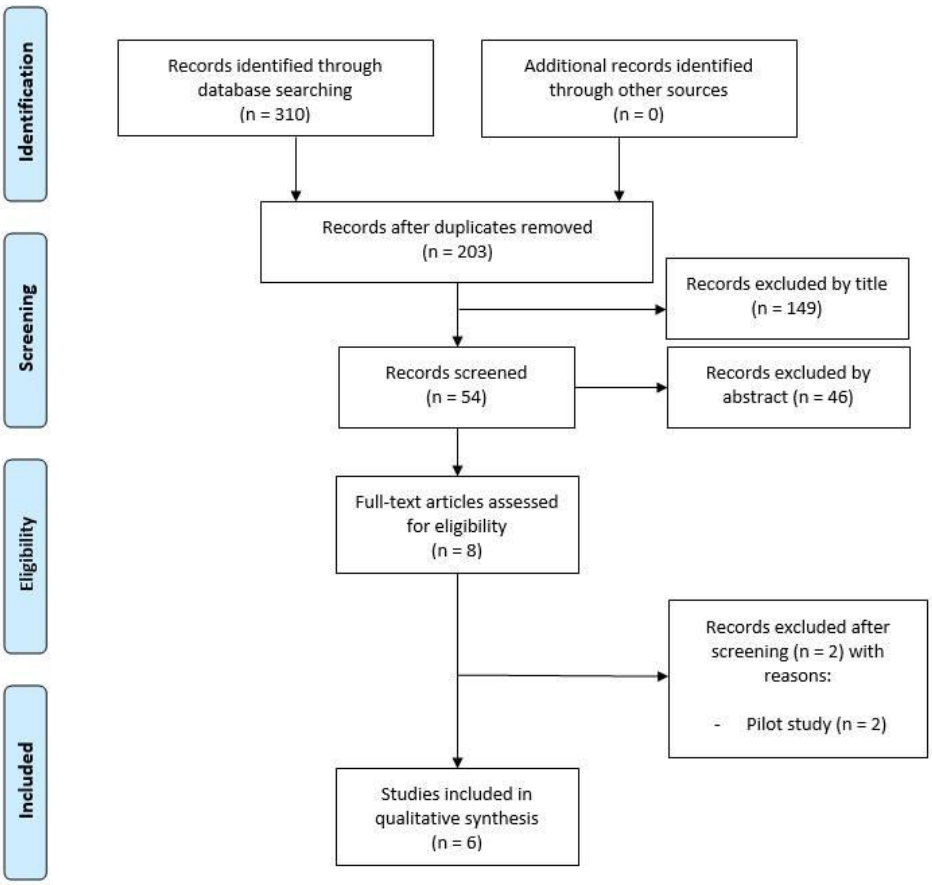

Figure 1 | Study selection process.

Characteristics of included studies

A total of 6 articles were considered for this review. Findings in terms of author, study type and study groups, sample, clinical parameters assessed in each study, existence of smoking or non-smoking patients, peri-implant diagnostic criteria, probiotic administered, duration of treatment and follow-up, presence of adverse events and funding are given in Table II.

Treatment modalities

In all 6 studies (4-7, 25, 41), all patients underwent supportive periodontal therapy, namely, non-surgical MD complemented with oral hygiene instructions - In all clinical trials, all participants underwent supragingival prophylaxis with ultrasounds. 3 studies, Galofré M. *et al.* 2018 (5), Hallström H. *et al.* 2016 (6), Laleman I. *et al.* 2020 (41), additionally, used titanium curettes to perform manual instrumentation of peri-implant pockets.

In 2 studies, Peña M. *et al.* (4) and Alqahtani F. *et al.* (7), in both test and control group, a MD was performed, and after that the patients were ordered to rinse with 15 ml of 0.21% of CHX for 1 minute, two times a day for 2 weeks (Wk).

Laleman I. *et al*. (41), performed a MD of the peri-implant sites using the Satelec P5 Newtron XS BLED (ACTEON® Médico-Dental Iberica S.A.U., Barcelona, Spain) with specific tips (PH1, PH2L and PH2RA), followed by hand instrumentation with titanium curettes. At last, the peri-implant pockets were subgingivally treated with the Air-N-GO Easy air polisher (ACTEON® Médico-Dental Iberica S.A.U., Barcelona, Spain).

Galofré M. *et al*. (5), carried out a supragingival prophylaxis in the mucositis implants and a subgingival non-surgical MD in the peri-implantitis implants using a NEWTRON® P5 ultrasonic generator (ACTEON® Médico-Dental Iberica S.A.U., Barcelona, Spain), carbon tips Ph1, Ph2L and Ph2R (ACTEON® Médico-Dental Iberica S.A.U.) and titanium curettes (Quirurgical Bontempi, Barcelona, Spain).

Quality and risk of bias assessment of selected studies

According to the level of evidence using SIGN criteria (Scottish Intercollegiate Guidelines Network) (40), 4 articles (4-6, 41) were coded with 1++ (very low risk of bias), 1 article (7) coded with 1+ of scientific evidence level (low risk of bias) and 1 article (25) coded with 1- (a high risk of bias) (Table I).

In 3 studies (4, 5, 7) there was no drop out and remaining 3 studies (6, 25, 41) had a drop out ranging from 6% to 17%. All of 6 studies (4-7, 25, 41) were randomized trials, but 1 study (25) used an inadequate randomization system (no computer generated allocation).

SIGN scoring system: methodology checklist for RCT

Study	1.1	1.2	1.3	1.4	1.5	1.6	1.7	1.8	1.9	1.10	2.1
Peña M. *et al. (4)*	AA	WC	WC	WC	WC	WC	WC	0%	WC	NA	1++
Alqahtani F. *et al. (7)*	AA	WC	NA	PA	WC	WC	WC	0%	WC	NA	1+
Fernández A. *et al*. (25)	PA	PA	AA	AA	WC	WC	WC	15%	AA	NA	1-
Laleman I. et al. (41)	AA	AA	WC	AA	WC	WC	WC	17%	WC	NA	1++
Galofré M. et al. (5)	AA	WC	WC	WC	WC	WC	WC	0%	WC	NA	1++
Hallström H. *et al.* (6)	WC	WC	WC	WC	WC	WC	WC	6%	WC	NA	1++

Table I | Risk of bias assessment of included studies.

Categories assessed are as follows: 1.1) Clear question; 1.2) Randomization; 1.3) Concealment method; 1.4) Blinding; 1.5) Groups similarity at baseline; 1.6) Treatment under investigation; 1.7) Standard, valid and reliable way; 1.8) Drop-out rate; 1.9) Intention to treat analysis; 1.10) Comparable results for all sites; 2.1) Overall bias rating.

WC, Well covered; **AA,** Adequately addressed; **PA,** Poorly addressed; **NA,** Not addressed; **NR,** Not reported; **NA,** Not applicable.

++, High quality; **+,** Low risk of bias; **-,** High risk of bias.

Main outcomes of the studies

- Plaque Index (PI)

Only 3 studies (7, 25, 41) showed significant differences between groups during follow-up visits.

In the Laleman I. *et al.* (41) study, PI (%) at baseline was 15% and decreased to 3% at 12 weeks Wk and to 2% at 24 Wk of follow-up. Regarding to the control group, the mean PI at baseline was 8% after 12 Wk the percentage of PI increased to 11%, and after 24 Wk, the PI was 7%. Thus, the reduction in PI was better during follow-up visits in the test group than in the placebo group ($p<.001$ at 12 Wk and $p=.002$ at 24 Wk).

Alqahtani F. *et al*. (7) showed that among the group of smoking patients, there was no statistically significant difference in follow-up visits between those who underwent non-surgical MD plus probiotic therapy (PT) and among those who only underwent non-surgical MD. About patients who never smoked, the PI percentage at baseline was 48% and after 3 months (Mo) of treatment it decreased to 10%, for those submitted to non-surgical MD plus PT ($p<.05$) and only to 24% for those who only underwent non-surgical MD. This difference is clearly statistically significant for the first group mentioned. In spite of this, at 6 Mo of follow-up, there were no differences between the groups that underwent both treatment protocols.

Fernández A. *et al*. (25) showed that after the administration of the PT, in both group A (patients without peri-implant disease) and B (patients with peri-implant mucositis), the mean PI decreased 0.59 and 0.74 points, respectively, and remained stable with placebo in both groups - the difference between the both groups was statistically significant ($p=0.001$ in group A and $p=0.035$ in group B).

Hallström H. *et al.* (6) reported a general improvement in both test and placebo group (with over 50% reduced) at the follow-up compared with baseline, but no significant differences were observed between groups.

Galofré M. *et al.* (5) mention a decrease in PI in the probiotic group as in the placebo group between baseline and 30 days in patients with mucositis (13% vs 7.1%) and peri-implantitis (13% vs 7.7%) and improved until 90 days. No significant differences were noted between the probiotic and placebo groups ($p>.05$). Regarding to the plaque index at the implant level (PII), it decreased after 30 days of receiving PT than placebo in implants with mucositis (36.4% vs 9.1%) and in implants with peri-implantitis (54.5% vs 27.3%), reaching no significant statistical differences between the groups.

Peña M. *et al.* (4) observed that both groups showed a comparable clinical improvement parameters at follow-up. At 15 days, a significant improvement was noted at the implant site (48% and 60% implants were no plaque, in the control group and test group, respectively.) At the end of the 3 Mo of follow-up, an absence of plaque over time was announced in 32% of the implants in the control group and in 44% of the test group. Thus, no significant differences were observed between the control and the test group.

- Bleeding on Probing (BOP)

In only 2 studies (5, 7), statistically significant differences were observed between groups during follow-up visits.

Galofré M. *et al*. (5) demonstrated that there are statistically significant differences between the test and control groups. After 30 days of intervention, the BOP was statistically significant for the test group of patients with mucositis compared to the control group (27% vs 8%, $p=0.031$). In patients with peri-implantitis, the BOP decreased 20% in the test group as compared to 8% decreased in the control group - not being statistically significant between groups. 90 days after the study intervention, BOP remained stable and improved in both control and test groups, being statistically significant in the test group of patients with mucositis ($p=.024$).

Regarding to bleeding on probing at the implant level (BOPI) on patients with mucositis and peri-implantitis, test group showed a significant improvement for

implants with peri-implantitis between baseline and for 30 days (p=.018) to 90 days (p=.045).

Alqahtani F. *et al.* (7), reported that among smoking patients, no statistically significant differences were observed even after 3 or 6 Mo of follow-up. In relation to never smoking patients, at 3 Mo of follow-up, the percentage of BOP decreased significantly in individuals submitted to non-surgical MD and PT in relation to those who only did non-surgical MD (p<.05). However, after 6 Mo of follow-up, there were no statistically significant differences between the two treatment protocols.

Laleman I. *et al.* (41), reported that the BOP decreased from 87% to 59% (p<.001) for the test group and from 87% to 53% (p<.001) for the control group at follow-up visits - not showing results statistically significant between the test and control groups.

Hallström H. *et al.* (6), described an improvement in both study groups (with over 50% reduced) at the follow-ups compared with baseline, but no significant differences were displayed both in probiotic and the control group.

Peña M. *et al.* (4) demonstrate that both groups showed a comparable clinical parameters at follow-up. Following administration of 0.12% CHX, BOP reduced in 52% and 44% of the patients in the control group and the test group, respectively. At 135 days, 32% of implants in both studies group BOP was solved. No significant differences intergroup were noted for these characteristics.

- Probing Pocket Depth (PPD)

Laleman I. *et al.* (41) reported that after 12 and 24 Wk of follow-up, PPD decreased from 5.17 mm to 4.13 mm, respectively, in the test group (p<.001). In the control group, after 12 and 24 Wk, PPD decreased from 5.45 mm to 4.30 mm and 4.18 mm (p<.001), respectively. Thus, both treatment groups showed improvement in terms of follow-up visits, with no statistically different differences between them.

Hallström H. *et al.* (6) indicated that the change in PPD ranged from 0.7-1.2 mm in both study groups compared to baseline ($p<0.05$), but no significative intergroup differences were reached.

Galofré M. *et al.* (5), demonstrated that after 30 days of intervention, the mean probing pocket depth at the implant level (PPDI) went from 3.84 mm at baseline to 3.58 mm in patients with peri-implant mucositis and from 5.07 mm to 4.55 mm in patients with peri-implantitis, both in the group where the probiotic was administered. The PPDI of the mucositis implants improved by 0.23 mm in the probiotic group and worsened by 0.15 mm in the control group between 30 and 90 days - being statistically significant ($p=.000$). In patients with peri-implantitis, the PPDI improved by 0.02 mm in the probiotic group and worsened by 0.05 mm in the control group between 30 and 90 days - not being statistically significant. At 90 days, implants with mucositis and peri-implantitis treated with probiotic had a reduction of 0.5 mm and those treated with placebo exhibit a decrease of 0.2 mm, being statistically significant in the peri-implantitis group ($p=.036$).

Alqahtani F. *et al*. (7) revealed that among cigarette-smokers, there was no statistically significant differences at all-time intervals for MD with and without adjunct PT. In never smoking patients, during 3 Mo of follow-up, the PPD at baseline was 3.5 mm and decreased to 0.9 mm in the MD+PT group ($p<.05$) and to 2 mm ($p>.05$) in the MD group. However, at 6 Mo of follow-up, no statistically significant differences were found between the patients who underwent non-surgical MD with or without adjunct probiotic.

Peña M. *et al.* (4) indicated that at 15 days of CHX rinse, PPD decreased in both groups, being only significant ($p=0.010$) in the control group (with a decreased of 0.24 mm). Up to 135 days, PPD remained constant, reaching a statistically significant reduction of 0.34 mm ($p=0.005$) in the control group and a reduction of 0.21 mm in the test group ($p=0.220$).

Fernández A. *et al*. (25) reported that in group A (patients without peri-implant disease), the mean PPD decreased from 2.72 mm to 2.56 mm after oral intake of the probiotic. After placebo, the mean PPD increased from 2.55 mm to 2.82 mm. In group B

(patients without peri-implant mucositis), the mean PPD decreased from 3.55 mm to 2.46 mm after PT and following placebo, the mean PPD increased from 2.47 mm to 2.65 mm - showing statistically significant differences between the test and control groups (p=0.001).

- Concentrations of the selected cyto- and chemokines in gingival crevicular fluid (GCF)

Hallström H. *et al.* (6) reported that the concentrations of the selected cyto- and chemokines in GCF (Interleukin-1 receptor antagonist (IL-1β), Interleukin 4 (IL-4), Interleukin 6 (IL-6), Interleukin 8 (IL-8), Interleukin-1 receptor antagonist (IL-1RA), Chemokine ligand 5 (CCL5), Interferon gamma (IFN-γ), Granulocyte macrophage-colony stimulating factor (GM-CSF), TNF-α, IL-17) was reduced in both groups compared with baseline (p<0.05), but there were no statistically significant intergroup differences. In contrast, Fernández A. *et al*. (25), showed that after oral ingestion of the probiotic, in both groups A (patients without peri-implant disease) and B (patients with peri-implant mucositis), the cytokine concentration values of IL-6 and IL-8 decreased (p=0.033 and p=0.013, respectively) - being better than the concentrations of these mediators after the administration of placebo (in both groups).

Author, year / Study characteristics	Peña M. *et al.* (4) 2019	Alqahtani F. *et al.* (7) 2019	Fernández A. *et al*. (25) 2015	Laleman I. et al. (41) 2020	Galofré M. et al. (5) 2018	Hallström H. *et al. (6)* 2016
Study design	A triple-blind parallel randomized clinical trial	A randomized clinical trial	A double-blind, placebo-controlled, prospective cross-over study	A double-blind, randmized, placebo-controlled study	Randomized, controlled, parallel-design, triple-blind prospective climical study	Double-blind randomized placebo-controlled trial
Clinical parameters	**FMPI, FMBOP, PI, BOP** and **PPD**	**PI, BOP** and **PPD**	**PICF, PI, PPD, MGI** and concentration of **IL-1β, IL-6, IL-8**	**FMBS, FMPS, BOP, MSBI, PI** and **PPD**	**PPDI, PII, PI, BOPI** and **BOP**	**PPD, PI, BOP** and **GCF**
N, gender and age (mean in years)	50 21♂; 29♀ 58.6	80 80♂ 35.6	34 15♂; 19♀ 62.4	19 9♂; 10♀ 66.7	44 23♂; 21♀ 60.0	49 18♂; 31♀ 58.5
Smoking	Yes	Yes	No	No	No	Yes
Definition of the peri-implant condition	Peri-implant mucositis was defined as the presence of BOP without radiographic signs of bone loss	Peri-implant mucositis was defined as the presence of BOP and PPD ≥ 3mm at ≥ 30% sites and radiographic crestal bone loss of up to 2mm	Peri-implant mucositis was defined as the presence of BOP without radiographic signs of bone loss. As healthy implants are considered all those with a PPD <4 mm, the absence of inflammation without radiographic signs of bone loss	Peri-impantitis was defined as the presence of inflammation; PPD ≥4 mm with radiological bone loss	Peri-implant mucositis was defined as an inflamed mucosa with BOP and/or suppuration without radiographic bone loss. Peri-implantitis was diagnosed as the presence of BOP and/or pus, PPD ≥5 mm and radiographic bone loss of ≥2 mm and/or ≥3 implant threads	Probing pocket depth ≥4 mm; bleeding and/or pus
Probiotic strain/ vehicle; regimen	*Lactobacillus reuteri* (ATCC PTA 5289 and DSM 17938) / tablets; 1 tablet at night	*Lactobacillus reuteri* (ATCC PTA 5289 and DSM 17938) / lozenges; 1 lozenge every 12 hours, twice a day	*Lactobacillus reuteri* (ATCC PTA 5289 and DSM 17938) / tablets; 1 tablet every 24h	*Lactobacillus reuteri* (ATCC PTA 5289 and DSM 17938) / lozenges; application of 5 drops around the implants with peri-implantitis	*Lactobacillus reuteri* (ATCC PTA 5289 and DSM 17938) / tablets; 1 tablet for 10 minutes, once daily at night	*Lactobacillus reuteri* (ATCC PTA 5289 and DSM 17938) / tablets; 1 tablet twice daily

Treatment	Day 0 - day 45	3/6 months	18 months	6/12/24 weeks	30 days	3 months
Follow-up	Day 45 - day 135				30/90 days	1, 2, 4, 12 and 26 weeks
Study outcome (Complete study in **Table III**)	Both groups showed a comparable clinical parameters at follow-up	At 6 months' follow-up, both groups showed a comparable clinical parameters	Test group showed a significant improvement in clinical parameters as compared to control group at follow-up	Test group only showed a significant improvement at **PI** and **FMPS** as compared to control group at follow-up	At 90 days of follow up, test group only found a significant improvement at **BOP, BOPI** and **PPDI**	Both groups showed a comparable clinical parameters at follow-up
Adverse Events	Not reported	Not reported	Not reported	Not reported	Not reported	Not reported
Funding	This study was funded by the Universitat de Catalunya and partially supported by a research grant from Sunstar Suisse S.A. (Etoy, Switzerland	King Saud University, Grant/Award Number: Vice Deanship of Scientific Research Chairs, King Saud University, Riyadh, Saudi Arabia	The placebo and probiotics where a kind contribution by Sunstar (Etoy, Switzerland) and BioGaia (Stockholm, Sweden)	This study was partially financially supported by BioGaia AB Sweden. Additional support came from grants from the KU Leuven (C24/17/086) and the FWO (G091218N)	Probiotics and placebo were kindly provided by Sunstar Suisse SA (Etoy, Switzerland) and BioGaia (Stockholm, Sweden)	The test and the placebo products were generously supplied by BioGaia AB, Lund, Sweden

Table II | General characteristics of included studies.

FMPI, Full mouth plaque index; **FMBOP**, Full mouth bleeding on probing; **PI**, Plaque Index; **PPD**, Probing pocket depth; **BOP**, Bleeding on probing; **PICF**, Peri-implant crevicular fluid volume; **MGI**, Modified gingival index; **IL-1β,** Interleukin-1β; **IL-6**, Interleukin-6; **IL-8**, Interleukin-8; **FMBS,** Full mouth bleeding score; **FMPS,** Full mouth plaque score; **MSBI,** Modified sulcus bleeding index; **PPDI**, Probing pocket depth at the implant level; **PII**, Plaque index at the implant level; **BOPI,** Bleeding on probing at the implant level; **GCF**, Gingival crevicular fluid.

Author, year	Study group		Clinical parameters evaluated	Findings	*p* value
	Test (n)	Control (n)			
Peña M. *et al.* (4) 2019	25 **CHX+MD+PT**	25 **CHX+MD+PBO**	FMPI; FMBOP	Decreased significantly in both groups.	NS
			PI; PPD; BOP	From baseline to 135 days, in both groups, these variables decreased.	NS
Alqahtani F. *et al.* (7) 2019	40 **CHX+MD+PT**	40 **CHX+MD+PBO**	PI; BOP; PPD (in CS)	No differences among MD with and without adjunct PT.	NS
			PI; BOP; PPD (in NS)	At 6-months' follow-up, no differences between groups were found.	NS
Fernández A. *et al.* (25) 2015	34 **MD+PT**	34 **MD+PBO**	PI; PPD; MGI; PICF	Were significantly better in test group compared to the placebo group.	p= 0.001
			IL-6; IL-8	PT led to a statistically significant decrease in these parameters' concentration (in some cases).	p= 0.033 (IL-6) p= 0.013 (IL-8)
			IL-1β	No intergroup differences could be noted.	NS
Laleman I. et al. (41) 2020	9 **MD+PT**	10 **MD+PBO**	BOP; MSBI; PPD	No intergroup differences could be noted.	NS
			PI; FMPS	Showed a significant improvement in test group.	p< 0.001
			FMBS	No intergroup differences could be noted.	NS
Galofré M. et al. (5) 2018	22 **MD+PT**	22 **MD+PBO**	PI; PII	No intergroup differences could be noted.	NS
			BOP; PPDI; BOPI	BOP was statistically significant in mucositis in PT; PPDI and BOPI were statistically significant in peri-implantitis in the PT.	p= 0.024 (BOP) p= 0.036 (PPDI) p= 0.045 (BOPI)
Hallström H. *et al. (6)* 2016	24 **MD+PT**	25 **MD+PBO**	PPD; PI; BOP; GCF	A general improvement of clinical parameters was seen in both groups at the follow-ups in comparison with baseline.	NS

Table III | Extended results of clinical outcome.

FMPI, Full mouth plaque index; **FMBOP**, Full mouth bleeding on probing; **PI**, Plaque Index; **PPD**, Probing pocket depth; **BOP**, Bleeding on probing; **CS,** Cigarette-smokers; **NS,** Never smokers; **PICF**, Peri-implant crevicular fluid volume; **MGI**, Modified gingival index; **IL-1β,** Interleukin-1β; **IL-6**, Interleukin-6; **IL-8**, Interleukin-8; **FMBS,** Full mouth bleeding score; **FMPS,** Full mouth plaque score; **MSBI,** Modified sulcus bleeding index; **PPDI**, Probing pocket depth at the implant level; **PII**, Plaque index at the implant level; **BOPI,** Bleeding on probing at the implant level; **GCF**, Gingival crevicular fluid; **CHX,** Chlorhexidine gluconate; **MD,** Mechanical debridement; **PT,** Probiotic therapy; **PBO,** Placebo; **NS,** Not significant.

DISCUSSION

In addition to all fields of action of probiotic therapy, from gastrointestinal pathologies to anticariogenic effects and infections caused by fungi (*Candida albicans*) (20, 42), these harmless bacteria have been the subject of recent research because they have the ability to control bacterial colonization, oral biofilm formation and the consequent control of oral infections, improving clinical patterns, such as decreased gingival bleeding, pocket depth and clinical attachment loss (10, 43).

The purpose of this work is to find out if bacteriotherapy in the form of probiotics has an additive and beneficial effect on the treatment of peri-implant diseases. With the results of this work we hope to help clinicians in their decision-making process about therapeutic options to tackle peri-implant diseases and, as far as author knowledge, this is the first systematic review on the topic.

Randomized controlled trials (RCTs) represent the gold standard for examining the success of medical interventions and are generally regarded as the most reliable source of scientific evidence (44). These studies are part of a set of scientific experiments carried out with the aim of determining an effective treatment for a particular disease, being used as instruments to verify the benefit or risk of medical interventions. These clinical trials are established as entirely important evidence bases with regard to clinical applicability and, therefore, a systematic and absolute assessment of methodological quality is essential, since depending on the level of quality, this can influence the scientific outcome and the interpretation of the research (44).

This review explored randomized, blinded clinical trials that evaluated the clinical effect of the probiotics in a population of patients with implants with peri-implant mucositis and peri-implantitis and only 6 studies (4-7, 25, 41) were selected.

Main Results

Half of the studies included in this systematic review showed a significant additional beneficial effect of probiotics on peri-implant diseases, although inconsistent.

Among these, Fernández A. *et al*. (25) showed an improvement in peri-implant health, with a significative reduction in clinical parameters (PI, PPD, MGI, PICF) and immunological parameters (IL-6, IL-8) with MD following oral PT when compared with MD alone, in patients with peri-implant mucositis. Next, Galofré M. *et al.* (5) also shown improvements in inflammatory parameters with significant reductions in bleeding scores in mucositis (BOP) and peri-implantitis (BOPI), as well as in PPDI. On the other hand, Laleman I. *et al*. (41) was unable to find significant reduction in bleeding scores (BOP, MSBI and FMBS) despite the significant improvement in plaque scores (PI and FMPS).

Previous studies have also shown a beneficial effect of probiotics in oral health. Teughels W. *et al*. (45) evaluated the benefit of *Lactobacillus reuteri* in 30 patients with chronic periodontitis and observed that the adjunctive use of *Lactobacillus reuteri* lozenges resulted in significant additional clinical improvements in the reduction of clinical parameters after 12 Wk of treatment, compared with the placebo group. Twetman S. *et al*. (46), in a double-blind placebo-controlled study evaluated the effect in clinical variables and inflammatory mediators the use of chewing gums containing *Lactobacillus reuteri*. The results of 42 healthy adults with moderate levels of gingival inflammation showed that crevicular fluid volume, TNF-α, IL-8 levels and BOP were significantly reduced during 4 Wk of intervention. Also, Vivekananda M.R*. et al.* (47) reported that *Lactobacillus reuteri* can be recommended during non-surgical therapy and in monitoring the periodontal supportive phase, providing a benefit additional at the clinical and at the level of pro-inflammatory markers.

Peri-implant crevicular fluid volume (PICF) composition can be potentially useful in determining the concentration of pro-inflammatory mediators that can interfere with the degradation of connective tissue collagen and alveolar bone (48). IL-1β and TNF-α are two cytokines that are related to the production of osteoblastic cells and the resorption of the alveolar bone. IL-1β control the degradation of extracellular matrix components of the plasminogen system and collagenase activity in inflammation and wound healing. TNF-α induces fibroblast apoptosis and reduction of the repair capacity of the peri-implant tissue (48). When the concentration of IL-1β is increased in the PICF, as well as, in the gingival tissue, it is associated with the existence of peri-implant and

peri-implantitis mucositis and is related to early marginal bone loss around endosseous implants (25). On the other hand, the presence of IL-6 and IL-8 is more associated with the severity of peri-implant diseases (25, 49).

Significant PI and FMPS values between the study groups can be explained by the existence of sample sizes small and for example, because different brands and types of implants were used for the study, as reported in this trial. Therefore, different implant surfaces lead to different responses from the adjacent support alveolar bone and research suggests that the roughness of the implant surface may interfere with the development of peri-implantitis (30). It is described in the literature that implants without a too rough surface do not facilitate the accumulation of bacterial biofilm and contribute to the health of peri-implant tissues (30). The properties of the surfaces of the implants influence the accumulation of plaque due to differences in surface free energy and surface roughness - in particularly for abutment surfaces it can be clinical relevance since these components pierce the soft tissue barrier (50).

Although the statistically significant results of these studies suggest that the application of probiotics may be beneficial to the treatment of peri-implant diseases, there is still no solid scientific evidence to validate this hypothesis since, all the remaining included studies (4, 6, 7) were unable to report consistent statistically significant differences between test and control, despite the positive results shown when a PT was added to the MD. Peña M. *et al.* (4) showed that both treatment protocols improve clinical parameters and the oral intake of the probiotic does not appear to show beneficial effects at either the clinical or microbiological level. Considering the results of this trial, non-surgical therapy combined with instructions for oral hygiene and administration of 0.12% CHX can reduce peri-implant inflammation. However, the absolute resolution of peri-implant mucositis will be difficult to achieved, being associated with residual bleeding on probing from 14.3% to 47.5%, according to Shwarz F. *et al.* (51). Furthermore, Hallström H. *et al.* (6) reported that the administration of the PT after 3 Mo does not promote improvements at the clinical, microbiological or inflammatory variables of peri-implant mucositis, compared to the use of placebo. Iniesta M. *et al.* (52) in a placebo-controlled, parallel study was conducted in 40 gingivitis subjects to investigate the effects of an orally administered

Lactobacillus reuteri or placebo, and no significant differences were found in PI or Gingival index (GI) during the study period between test and control groups.

In the study of Alqahatani F. *et al.* (7), non-surgical mechanical therapy combined with oral intake of *Lactobacillus reuteri* does not improve the inflammatory parameters of peri-implant tissues at the end of the study period, when compared to MD alone, in never-smoking patients. Regarding to smoking patients, there were no statistically significant differences in PI, BOP and PPD throughout the time that they underwent MD with or without *Lactobacillus reuteri.* Studies show that tobacco compromises soft tissue healing and the clinical outcome of oral therapeutic interventions. Romero A. *et al.* (53) identified that tobacco smoke affects gingival and periodontal healing and may influence the clinical statistical results of the studies. Tobacco smoke alters cell migration and the differentiation of myofibroblastics and alters the ability of gingival fibroblasts to form a collagen matrix, impairing tissue maturation during wound healing. In another previous study, Zhang M. *et al.* (54), reported that cigarette smoke may destroy the balance and alter the localization of the matrix metalloproteinases (MMPs) and tissue inhibitors of metalloproteinases (TIMPs) to promote the degradation of the extracellular matrix as seen in periodontal disease.

To summarize, there is inherent methodological heterogeneity in the included studies and factors such as the existence of groups of smoking (4, 6, 7) and non-smoking patients (5, 25, 41) may have affected the outcome of treatments. Also the dose and the mode of administration of the probiotic, as well as the current state of health of the patient and the diet adopted by him, the existence of different treatment periods with small sample sizes and still a limited evidence on this topic contributed to the discrepancies in the results found. Another factor to take into account is that most of these randomized clinical trials (4-7, 41) did not respond to the so-called "*Hawthorne effect*" - and thus, the improvement in clinical parameters may have been affected by patients' motivation to improve their behavior and oral hygiene because they knew they would be evaluated during the study.

Therefore, and based on the results of six included studies (4-7, 25, 41), this systematic review demonstrated inconclusive findings about the role of PT in the management of peri-implant mucositis and peri-implantitis. Furthermore, other peri-

implant inflammatory factors such as microbiological and immunological parameters should be explored in order to analyze the treatment outcome.

Limitations and Clinical Relevance

As a result of to the variety of measurement methodologies, there is a need for additional studies that use a similar and equal methodology, which would make the comparison of results in the form of meta-analysis - this being perhaps the major limitation of the present study. It is also relevant to note that all included studies used the same probiotic and, therefore, their results cannot be generalized to the other types of probiotics. In light of this, randomized controlled trials with homogeneous methodologies and long-term follow-up periods with proper exclusion of confounders and involving larger patients' samples with enough statistical power are needed to clarify and validate the research question.

In last few years, oral rehabilitation with dental implants is increasingly used to replace edentulous spaces. The biggest problem inherent in dental implants is the occurrence of peri-mucositis, which, when not treated in a timely manner, can lead to peri-implantitis and the loss and irreversible failure of the dental implant. The diagnosis and early treatment of peri-implant pathology is extremely important, preventing long-term complications and the maintenance of bone support and the health of peri-implant soft tissues. Complete oral hygiene instructions with a self-performed oral hygiene measures around implants and a personalized follow-up period supportive therapy program are the main measures for the prevention of peri-implant diseases (8). The dentist should adopt the therapy that is most beneficial to the patient's general health, taking into account the needs and risk factors in each patient. Probiotics have been shown to enhance humoral immune responses and act on a wide variety of cells to modulate the immune system towards anti-inflammatory action (55). In view of this, and as far as finding treatment protocols that allow us to replace the conventional treatments used to treat oral infections that are present in this type of oral pathologies, probiotics have been the target of scientific research to complement the action of conventional treatments or even endure alone in the disease.

CONCLUSION AND FUTURE PERSPECTIVES

Despite the heterogeneity of the results found, we can conclude that there is no solid scientific evidence to document the efficacy of probiotics in peri-implant diseases.

To date, there is still very limited scientific evidence that allows us to draw solid conclusions about the role of probiotics in the treatment of peri-implant disease.

This systematic review highlights the need for more investigations in this field with larger samples and extended follow-up periods using different strains and different probiotic quantities.

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APPENDIX

Levels of Evidence	Diagnosis
1++	High quality meta-analyses, systematic reviews of RCTs, or RCTs with a very low risk of bias
1+	Well conducted meta-analyses, systematic reviews, or RCTs with a low risk of bias
1-	Meta-analyses, systematic reviews, or RCTs with a high risk of bias
2++	High quality systematic reviews of case control or cohort studies High quality case control or cohort studies with a very low risk of confounding or bias and a high probability that the relationship is causal
2+	Well conducted case control or cohort studies with a low risk of confounding or bias and a moderate probability that the relationship is causal
2-	Case control or cohort studies with a high risk of confounding or bias and a significant risk that the relationship is not causal
3	Non-analytic studies, such as case reports, case series
4	Expert opinion

Supplemental Table I | Levels of scientific evidence SIGN (40).

Grafes of recomendation	Interpretation
A	At least one meta-analysis, systematic review, or RCT rated as 1++, and directly applicable to the target population; or a body of evidence consisting principally of studies rated as 1+, directly applicable to the target population, and demonstrating overall consistency of results
B	A body of evidence including studies rated as 2++, directly applicable to the target population, and demonstrating overall consistency of results; or extrapolated evidence from studies rated as 1++ or 1+
C	A body of evidence including studies rated as 2+, directly applicable to the target population and demonstrating overall consistency of results; or extrapolated evidence from studies rated as 2++
D	Evidence level 3 or 4; or extrapolated evidence from studies rated as 2+

Supplemental Table II | Grades of recommendation of SIGN Criteria (Scottish Intercollegiate Guidelines Network) (40).

Section/topic	#	Checklist item	Reported on page #
TITLE			
Title	1	Identify the report as a literature review.	II
ABSTRACT			
Structured summary	2	Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings;	IX
INTRODUCTION			
Rationale	3	Describe the rationale for the review in the context of what is already known about your topic.	1
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).	4
METHODS			
Eligibility criteria	5	Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication status) used as criteria for eligibility, giving rationale.	5
Information sources	6	Describe all information sources (e.g., databases with dates of coverage) in the search and date last searched.	5
Search	7	Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.	5
Study selection	8	State the process for selecting studies (i.e., screening, eligibility).	5
Risk of bias in individual studies	9	Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level).	6-7
Risk of bias across studies	10	Specify any assessment of risk of bias that may affect the cumulative evidence (e.g., publication bias, selective reporting within studies).	6-7
RESULTS			
Study selection	11	Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.	8
Study characteristics	12	For each study, present characteristics for which data were extracted (e.g., study size, PICOS, follow-up period) and provide the citations.	9

Section/topic	#	Checklist item	Reported on page #
Synthesis of results of individual studies	13	For all outcomes considered (benefits or harms), present, for each study: (a) summary of results and (b) relationship to other studies under review (e.g. agreements or disagreements in methods, sampling, data collection or findings).	9-18
DISCUSSION			
Summary of evidence	14	Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g., healthcare providers, users, and policy makers).	19-24
Limitations	15	Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research, reporting bias).	24
CONCLUSION			
Conclusions	16	Provide a general interpretation of the results in the context of other evidence, and implications for future research.	25

Adapted from: Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group (2009). Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA statement. *PLoS Medicine*, 6(6), e1000097. doi:10.1371/journal.pmed1000097

METHODOLOGY CHECKLIST 2: RANDOMISED CONTROLLED TRIALS			
Study identification (Include author, title, year of publication, journal title, pages)			
Guideline topic:		Key Question No:	
Checklist completed by:			
SECTION 1: INTERNAL VALIDITY			
In a well conducted RCT study...		***In this study this criterion is:***	
1.1	The study addresses an appropriate and clearly focused question.	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable
1.2	The assignment of subjects to treatment groups is randomised	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable
1.3	An adequate concealment method is used	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable
1.4	Subjects and investigators are kept 'blind' about treatment allocation	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable
1.5	The treatment and control groups are similar at the start of the trial	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable
1.6	The only difference between groups is the treatment under investigation	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable
1.7	All relevant outcomes are measured in a standard, valid and reliable way	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable
1.8	What percentage of the individuals or clusters recruited into each treatment arm of the study dropped out before the study was completed?		
1.9	All the subjects are analysed in the groups to which they were randomly allocated (often referred to as intention to treat analysis)	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable
1.10	Where the study is carried out at more than one site, results are comparable for all sites	Well covered Adequately addressed Poorly addressed	Not addressed Not reported Not applicable

SECTION 2: OVERALL ASSESSMENT OF THE STUDY		
2.1	How well was the study done to minimise bias? Code + +, +, or –	
2.2	If coded as +, or – what is the likely direction in which bias might affect the study results?	
2.3	Taking into account clinical considerations, your evaluation of the methodology used, and the statistical power of the study, are you certain that the overall effect is due to the study intervention?	
2.4	Are the results of this study directly applicable to the patient group targeted by this guideline?	
SECTION 3: DESCRIPTION OF THE STUDY ***(The following information is required to complete evidence tables facilitating cross-study comparisons. Please complete all sections for which information is available).*** **PLEASE PRINT CLEARLY**		
3.1	How many patients are included in this study? Please indicate number in each arm of the study, at the time the study began.	
3.2	What are the main characteristics of the patient population? *Include all relevant characteristics - e.g. age, sex, ethnic origin, comorbidity, disease status, community/hospital based*	
3.3	What intervention (treatment, procedure) is being investigated in this study? *List all interventions covered by the study.*	
3.4	What comparisons are made in the study? Are comparisons made between treatments, or between treatment and placebo / no treatment?	
3.5	How long are patients followed-up in the study? *Length of time patients are followed from beginning participation in the study. Note specified end points used to decide end of follow-up (e.g. death, complete cure). Note if follow-up period is shorter than originally planned.*	
3.6	What outcome measure(s) are used in the study? *List all outcomes that are used to assess effectiveness of the interventions used.*	
3.7	What size of effect is identified in the study? *List all measures of effect in the units used in the study - e.g. absolute or relative risk, NNT, etc. Include p values and any confidence intervals that are provided.*	
3.8	How was this study funded? *List all sources of funding quoted in the article, whether Government, voluntary sector, or industry.*	
3.9	Does this study help to answer your key question? *Summarise the main conclusions of the study and indicate how it relates to the key question.*	

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Declaração de Autoria de Trabalho

MONOGRAFIA DE INVESTIGAÇÃO/RELATÓRIO DE ATIVIDADE CLÍNICA

Declaro que o presente trabalho, no âmbito da Monografia de Investigação/Relatório de Atividade Clínica, integrado no Curso de Mestrado Integrado em Medicina Dentária da Faculdade de Medicina Dentária da Universidade do Porto, é da minha autoria e todas as fontes foram devidamente referenciadas.

Porto, 15 de maio de 2020.

Ana Isabel Carvalho Moreira

(Ana Isabel Carvalho Moreira)

U.PORTO

FACULDADE DE MEDICINA DENTÁRIA
UNIVERSIDADE DO PORTO

Parecer

Eu, Luzia da Conceição Martins Mendes Gonçalves, Professora Auxiliar Convidada da Faculdade de Medicina Dentária da Universidade do Porto (FMDUP), declaro que o Trabalho da Monografia desenvolvido pela estudante Ana Isabel Carvalho Moreira, do 5º ano do Curso de Mestrado Integrado em Medicina Dentária da FMDUP, subordinada ao tema "Os Probióticos e o Tratamento de doenças peri-implantares: uma revisão sistemática/Probiotics and the Treatment of Periimplant Diseases: a systematic review", se encontra e está de acordo com as regras estipuladas pela FMDUP.

Mais informo que o referido trabalho foi por mim conferido e se encontra em condições de ser apresentado e defendido em provas públicas.

Porto, 15 de maio de 2020.

(Luzia da Conceição Martins Mendes Gonçalves)

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UNIVERSIDADE DO PORTO

Parecer

Eu, José António Ferreira Lobo Pereira, Professor Auxiliar da Faculdade de Medicina Dentária da Universidade do Porto (FMDUP), declaro que o Trabalho da Monografia desenvolvido pela estudante Ana Isabel Carvalho Moreira, do 5º ano do Curso de Mestrado Integrado em Medicina Dentária da FMDUP, subordinada ao tema "Os Probióticos e o Tratamento de doenças peri-implantares: uma revisão sistemática/Probiotics and the Treatment of Periimplant Diseases: a systematic review", se encontra e está de acordo com as regras estipuladas pela FMDUP.

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Porto, 15 de maio de 2020.

(José António Ferreira Lobo Pereira)

Probiotics and the treatment of peri-implant diseases: a systematic review.

Ana Isabel Carvalho Moreira

2020