Probiotics in the Management of Gingivitis and Periodontitis. A Review

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In the management of intestinal health problems, the targeted use of probiotic microorganisms is a common therapeutic measure with a long-standing tradition. In clinical dentistry however, probiotics-based therapy is still a rather new and developing field, whose usefulness for the control of gingivitis and periodontitis has been questioned by recent meta-analyses and systematic reviews. The purpose of the subsequent descriptive review is to provide an introduction to the concept of probiotic microorganisms and their multifaceted health-promoting interactions with the human host and microbial competitors, followed by a detailed comparison of the results of available controlled clinical trials assessing the use of probiotics in the control of gingival and periodontal inflammations. It aims at contributing to a deeper understanding of the unique capabilities of probiotics to resolve chronic plaque-induced inflammation even in the absence of mechanical plaque control and will discuss how possible misconceptions about the rationale for using probiotics may have led to the present controversies about their usefulness as a therapeutic option.

Keywords: probiotics, gingivitis, periodontitis, review, periodontal therapy

THE HUMAN MICROBIOME

The skin and all mucosal surfaces of the human body are colonized by a multitude of different microbial species. According to current estimates (1) their cumulative number (3.0×10^{13}) roughly matches the number of human body cells. Due to the enormous species richness of colonizing microorganisms their combined genome has been recently calculated to contain a total of more than 45 million different microbial genes (2). It vastly outnumbers the human genome, containing, depending on the definition, only about 20.000–100.000 different genes (3), which has contributed to the understanding, that host-microbiota interactions are an integral and essential part of human physiology (4). While the concept of antisepsis pioneered by Louis Pasteur and Robert Koch certainly laid the foundation for the control of infectious diseases and saved the lives of millions of people, it narrowed the focus of scientific efforts for many decades mainly to the exploration of bacterial pathogens, which in a healthy host are either absent or forming only a minute portion of the resident microbiota. This is particularly true in clinical dentistry, where the significance of the oral microbiota still is mostly reduced to being a health threat, exemplified by the fact, that therapeutic concepts based on the indiscriminate elimination of oral bacterial biofilms continue to dominate even the most recent evidence-based guidelines for the management of caries and periodontal disease (5–7).
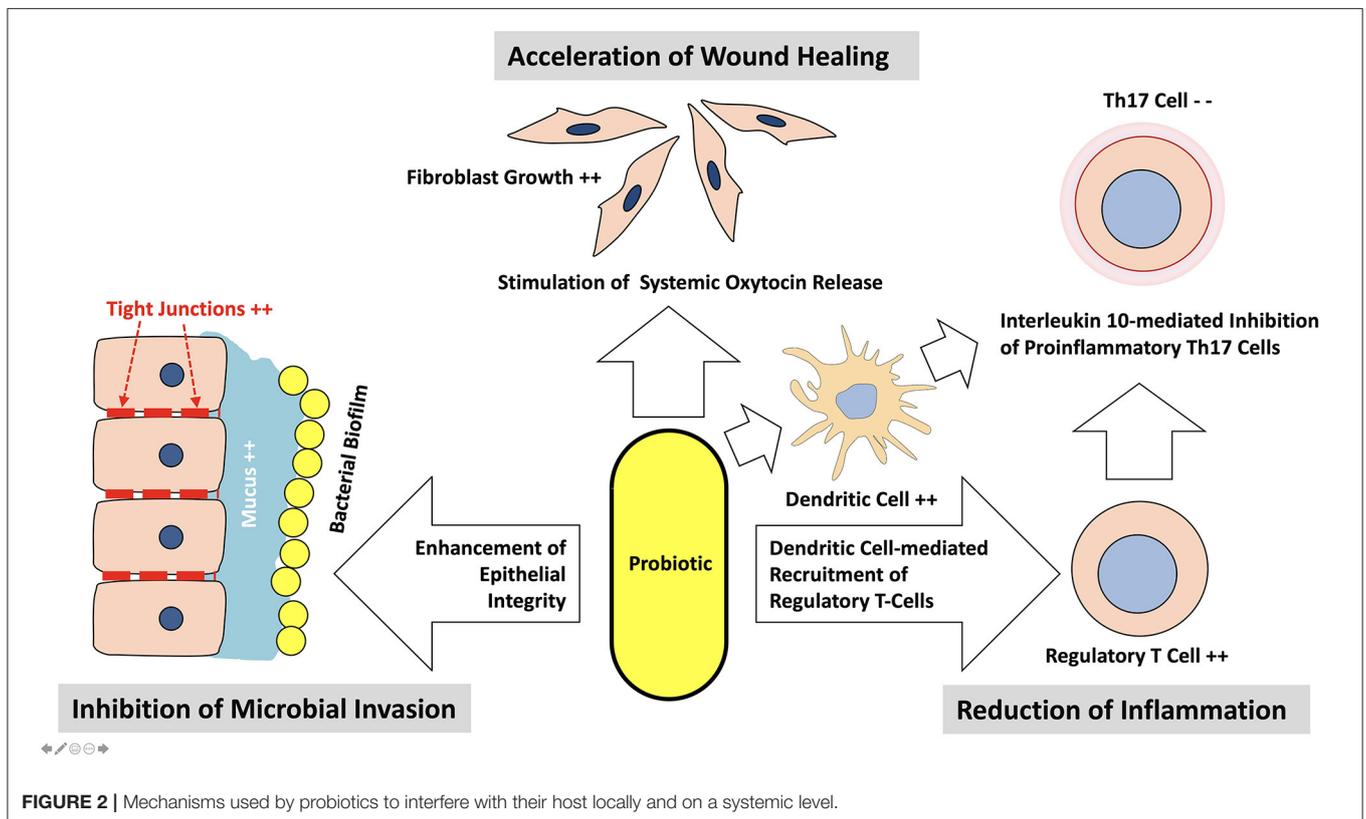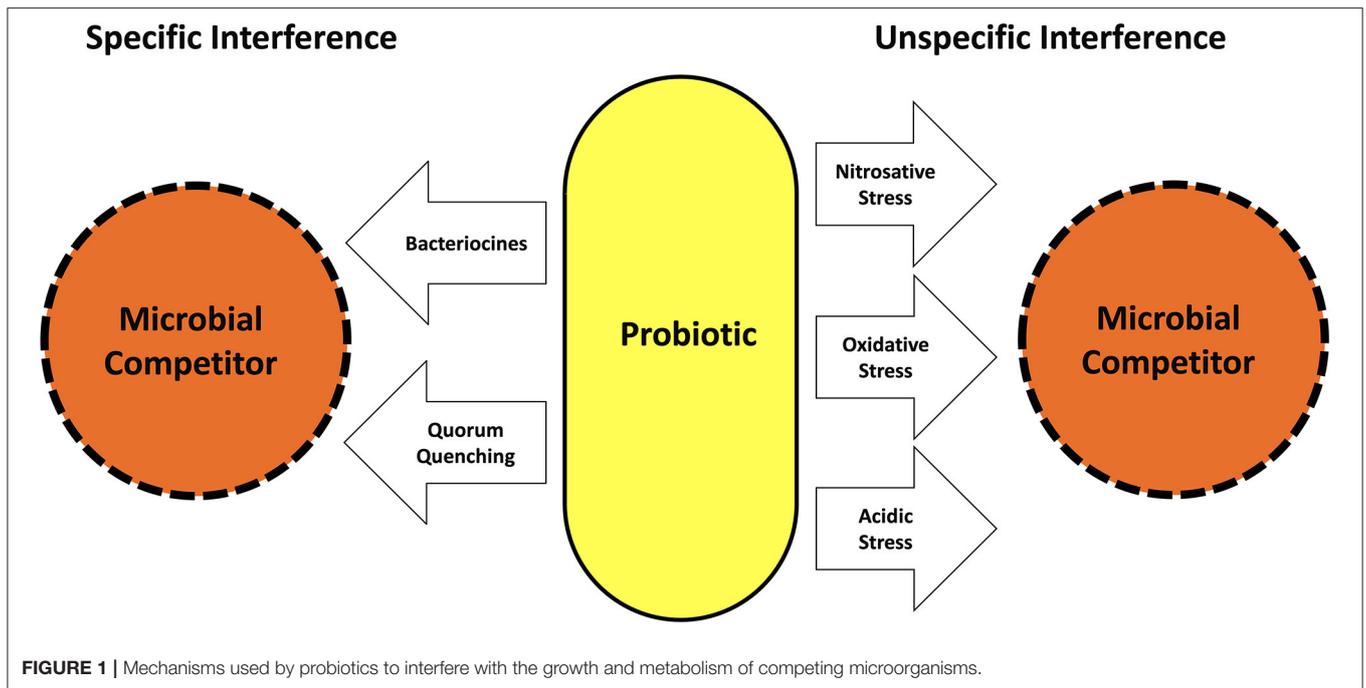
Microorganisms With a Beneficial Impact on the Health of Their Host (Probiotics)

Nevertheless, in some Asian countries like Japan, the beneficial impact of the consumption of certain bacterially fermented foods on human health has been known already as experiential knowledge for centuries. Nobel laureate Eli Metchnikoff was the first modern-day scientist, who investigated the nature of beneficial bacteria systematically (8). Noting that Bulgarian peasants lived longer than those in other countries, despite comparably miserable living conditions, he traced down the possible reason for the observed longevity to the consumption of yogurt, which was very popular in Bulgaria and attributed its health-promoting effect to its rich content of the bacterial species *Lactobacillus delbrueckii. subsp. bulgaricus*. Ever since a whole variety of beneficial bacteria and yeasts, mostly associated with fermented food components of the human diet has been identified and is nowadays summarized by the term "probiotics." According to a definition set by the World Health Organization in 2001 (9), probiotics are microorganisms, which survive the passage through the acidity of the stomach alive, and when consumed in adequate amounts, confer a health benefit on the host. This definition has been met so far by a series of microorganisms among them e.g., various strains of *Lactobacillus spp.*, *Streptococcus spp., Bifidobacterium spp., Bacillus spp.* as well as the *Escherichia coli* strain Nissle 1917 or the yeast *Saccharomyces boulardii* (10). Their health-promoting probiotic properties may originate from a local interference with competing members of the human microbiota as well as by a direct contact to cellular structures of the mucosal immune system of the host, which may not only elicit a local but also a systemic response modulating inflammatory reactions even in distant parts of the body. As schematically depicted in **Figure 1**, probiotics are able to inhibit other microorganisms competing for the same space and/or substrates by interfering with their metabolism and/or living conditions. This may occur by a variety of measures, like a change in local pH or the concentration of reactive oxygen or nitrogen species (ROS; RN) via the synthesis of metabolites like organic acids, peroxide or nitric oxide (11, 12) as well as by the synthesis and release of so called bacteriocines, specific peptides, able to selectively kill competing bacteria or to change the spectrum of their metabolism (13–15). Some probiotics are also capable to successfully impair the biofilm formation of their microbial competitors by various mechanisms including the quenching of their quorum sensing communication (16, 17). Many probiotic bacteria and particularly probiotic lactobacilli display the ability to attach to the mucus lining of the gut and oral epithelia (18), which brings them in close proximity to the body cells. Their health-promoting impact therefore may also arise from a selective interference with immunocompetent host cells like dendritic cells, T-cells or generic epithelial cells at the interface between the lining mucosa and the opposing microbiota (see **Figure 2**). This may result in an improved epithelial barrier function due to a stimulated increase in the thickness of the protective mucus layer as well as due to reestablishing the tightness of damaged interepithelial tight junctions (19, 20).

Moreover, the consumption of probiotic *Lactobacillus reuteri* strains showed to enhance tissue repair of defined skin wounds in mice as well as in human volunteers despite not being physically in direct contact with the wound, by increasing the plasma level of the neuropeptide oxytocin (21, 22), accompanied by a downregulation of blood corticosterone levels and a reduction in the number of circulating blood neutrophils. In an *in vitro* experiment the exposure of mouse bone marrow dendritic cells (DC) to *L. reuteri* supernatants or sterile *L.reuteri*-lysates promoted DC maturation and upregulated the secretion of anti-inflammatory interleukin-10 (23). In a rat model of necrotizing enterocolitis, which is also a life-threatening condition in the intestine of premature human infants, the addition of *L.reuteri* DSM 17938 to the diet of the newborn rats resulted in a downregulation of enteric inflammation by an increased migration of Foxp3(+) regulatory T-cells to the inflamed parts of the gut (24). Immunomodulatory properties however are not limited to specific *L. reuteri* strains but have also been identified in many other commonly used probiotics (25). While various probiotic bacteria readily adhere to the intestinal and oral epithelia after ingestion, it must be noted, that most of them are unable to colonize their new host permanently (26). Clinical trials have shown that the number of detectable probiotic bacteria rapidly decreases within days to few weeks after the cessation of their consumption, making their continuously repeated intake for the maintenance of the beneficial effects associated with them a necessity (27). Although most therapeutically used probiotic bacteria have been initially retrieved from the resident microbiota of healthy human individuals, they may not readily fit into the microbial spectrum of other human hosts due to inhibition by microbial competitors from the resident host microbiota, an insufficient genetic adaption to the new host or, probably most important, due to a lack of suitable growth substrates provided by the daily diet of their actual host, as indicated by studies evaluating the impact of different food elements on the growth and the diversity of the human microbiota (28, 29).

Safety Aspects of Probiotics Consumption

Given the wide array of known metabolic interferences with the human microbiota and the impact on the physiology of the host as described before, potential adverse effects associated with an indiscriminate, medically unsupervised consumption of probiotics need to be considered. They include the induction of bacterial translocation, bacteremia, sepsis, bacterial dysbiosis and the conferral of bacterial resistance genes, further complicated by the development of systemic health problems induced by these conditions (30). Cumulative evidence from the literature nevertheless, proves an overall excellent safety record for the long-term consumption of probiotics by healthy adults and children (31–33). There are however various case reports of bacteremia, fungemia or bacterial dysbiosis or other adverse events. It has to be noted, that the vast majority of them occurred in individuals, who were concomitantly affected by severe chronic systemic disease or compromised immunity and/or frailty (32, 34–37), conditions also frequently associated

FIGURE 1 | Mechanisms used by probiotics to interfere with the growth and metabolism of competing microorganisms.

FIGURE 2 | Mechanisms used by probiotics to interfere with their host locally and on a systemic level.

with bacterial dysbioses of the human microbiome (38) and the development of severe periodontitis (39–42). The validity of a further, much-noticed case report blaming the development of D-lactic acidosis-induced "brainfogginess" to the consumption of a mix of lactobacilli and bifidobacteria, *Streptococcus thermophilus* and some other unspecified bacterial species (43) has been challenged by other experts, as all commonly used probiotic lactobacilli and bifidobacteria are known to exclusively synthesize

L-lactic acid (44). In a clinical study, assessing urinary D-lactic acid concentrations in healthy newborns, who consumed a *L.reuteri* DSM 17938-containing infant formula during the first 28 days of their life, the addition of the probiotic to their diet did not initiate or facilitate the colonizing of D-lactic acid synthesizing bacteria in the developing intestinal microbiota of the newborns, compared to controls, consuming an identical infant formula being void of probiotics (45). Another widespread misunderstanding is the assumption, that probiotics may only be consumed for a limited time. This may have arisen because the duration of probiotic consumption reported in clinical trials is usually only weeks or a few months. However, all commercially available probiotic products must have approval from the relevant regulatory authorities as food supplements, which includes the safety of indefinite, long-term consumption (see next section legal and commercial aspects). In summary the consumption of probiotics may be regarded safe for systemically healthy persons, but it may be advisable to use only probiotic products whose safety profile for the intended application purpose has been verified by *in vitro* assessments and clinical trials (30).

Legal and Commercial Aspects of Probiotics

In the European Union and the United States probiotics are not legally considered medicines but food supplements. Their market admission therefore needs to comply with the safety regulations of the European Food Safety Authority (EFSA) or the US Food and Drug Administration (FDA) for a food or food supplement, but it is not dependent on the verified proof of any health benefit. Currently there is a multitude of commercial probiotic food supplements freely available, which all meet EFSA and/or FDA safety standards, but which may not have any evidence for a health-promoting impact supported by the results of randomized controlled clinical trials. As a matter of fact, in 2011 EFSA did no longer allow the promotion of any health claim for any commercial probiotic product sold in Europe, due to a general lack of scientific evidence meeting the efficacy standards set by EFSA (EC regulations #1924/2006, # 1169/2011). It must be noted in this context, that the extent and the specificity of the health-promoting efficacy of probiotics show huge variations even within different strains of the same species. Therefore, the consumption of two seemingly comparable probiotic food supplements containing different strains of the same bacterial species may show profound differences regarding their impact on the microbiota as well as on the health status of their host (30, 46). Also, the indiscriminate use of probiotic multi-species food supplements with possibly opposing properties may not be advisable due to the arising complexity of interactions (30).

PROBIOTICS IN THE MANAGEMENT OF ORAL HEALTH PROBLEMS

Although the vast majority of available scientific evidence regarding the therapeutic usefulness of probiotics has originated so far from the field of intestinal health research, there is now a steadily growing number of randomized controlled clinical trials (RCT) dedicated to the evaluation of probiotics in the management of oral health problems and in particular gingivitis and periodontitis. The subsequent descriptive analysis is based on the results of a total of 36 available RCTs, evaluating the use of probiotics in the control of gingivitis or periodontitis. They were published between 2009 and 2021 and were all retrieved from the PubMed database. Their main characteristics are schematically listed in **Tables 1**, **2**.

Probiotics Evaluated in Oral Trials

The most frequently evaluated probiotic, used in 17 of the reviewed 36 RCTs, was a combination of two probiotic *Lactobacillus reuteri* strains (DSM17938; ATCC PTA 5289) (49–54, 67–76), followed by various probiotic *Lactobacillus rhamnosus* strains (60–62, 79–81), probiotic strains of *Lactobacillus brevis* (56, 57, 77), *Bifidobacterium animalis subsp. lactis* (48, 64, 65), *Lactobacillus casei* Shirota (58, 59), *Weissella cibaria* (63), *Lactobacillus plantarum* (78) and combinations of probiotic *Bacillus subtilis, B. megaterium* und *B. pumilus* (47), *Streptococcus oralis, S. uberis* and *S. rattus* (82) as well as *Streptococcus thermophilus, Enterococcus faecium, Bifidobacterium longum,* and *B. animalis subsp. lactis* strains in conjunction with prebiotic fructooligosaccharides (55).

Application, Dosage and Duration of Consumption

In most trials the probiotic bacteria were consumed as an ingredient of a lozenge, but also the administration via a probiotic yogurt (58, 59) or as an ingredient of toothpaste (47) has been evaluated. The reported probiotics dosage administered in a single application mostly varied between 10^8 and 10^9 colony forming units (CFU), with a bandwidth ranging from as low as 3×10^3 CFU (57) up to 6.5×10^9 CFU (58). The number of daily intakes varied between $1 \times$ daily to $4 \times$ daily, with $2 \times$ daily being the most frequently specified consumption scheme. Total duration of consumption varied from 21 to 84 days.

Probiotics in the Management of Chronic Gingivitis and Experimental Gingivitis
Evaluated Study Cohorts

The impact of probiotics administration in the management of chronic and experimental gingivitis has been assessed in a broad range of different cohorts, like adolescents (48, 61), adults (56, 57, 59, 62), dental students (49, 58, 63), pregnant women (52), smokers (55), navy sailors on duty (54), diabetics (53), or frail elderly (51).

Evaluated Study Outcomes

Evaluated study outcomes were mostly clinical indices of gingival inflammation like gingival index (GI) and bleeding on probing (BoP), often supplemented by the documentation of plaque accumulation and an analysis of the composition of the gingivitis-associated microbiota. Study participants were either affected by established chronic gingivitis of varying severity or were subjected to the development of experimental gingivitis after cessation of personal oral hygiene (48, 50, 56, 58, 59).

TABLE 1 | Randomized controlled trials evaluating the use of probiotics in the management of gingivitis.

References	Type of investigation	Assessed condition	No. subjects	No. groups	PMPR	Hygiene instructions	Assessed probiotics	Application mode	Dosage (CFU)	Frequency/ day	Duration of intake	Observation time	Assessed Outcomes	Primary outcome	Outcomes significantly improved by consumption of probiotic
Alkaya et al. (47)	RCT	Gingivitis	30	2	Yes	Yes	*Bacillus subtilis, Bacillus megaterium, Bacillus pumilus*	Toothpaste	5×10^7	2 x daily	56 days	56 days	PI, GI, PPD, BoP	PI, GI	None
Kuru et al. (48)	RCT	Experimental gingivitis	51	2	Yes	Yes	*Bifidobacterium.animalis.subsp. lactis* DN-173010	Yogurt	1×10^8	1 x daily	28 days	33 days	GCF volume, PI, GI PPD, BoP	Not specified	PI, GI, BoP, GCF volume
Iniesta et al. (49)	RCT Cross-over[#]	Gingivitis	40	2	Yes	No	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	1 x daily	28 days	28 days	GI, PI	Not specified	None
Hallström et al. (50)	RCT Cross-over	Experimental gingivitis	18	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 x daily	21 days	21 days	PI, GI, BoP	Not specified	None
Kraft-Bodi et al. (51)	RCT	Gingivitis candidiasis	215	2	No	No	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 x daily	84 days	84 days	Candida count PI, BoP	Candida count	Candida count
Schlagenhauf et al. (52)	RCT	Gingivitis pregnancy	45	2	No	No	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 x daily	49 days	49 days	GI, PI	GI	GI, PI
Sabatini et al. (53)	RCT No Placebo	Gingivitis diabetes	80	2	No	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 x daily	30 days	30 days	PI, BoP	Not specified	PI, BoP
Schlagenhauf et al. (54)	RCT	Gingivitis	72	2	No	No	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 x daily	42 days	42 days	BoP, PI, GI, PPD, CAL	BoP	BoP, PI, GI, PPD, CAL
Ercan et al. (55)	RCT	Gingivitis smoking	80	4	Yes	Yes	*Lactobacillus acidophilus, Bifidobacterium animalis.subsp. lactis, Enterococcus faecium, Streptococcus thermophilus, Bifidobacterium longum*	Lozenge	1.4×10^8	1 x daily	30 days	30 days	GCF interleukins PI, GI	GCF interleukins	None
Lee et al. (56)	RCT	Experimental gingivitis	30	2	Yes	Yes	*Lactobacillus brevis* CD2	Lozenge	1×10^9	3 x daily	14 days	14 days	GI, PI, BoP, PPD GCF volume	Not specified	None
Montero et al. (57)	RCT	Gingivitis	52	2	Yes	Yes	*Lactobacillus brevis, Lactobacillus plantarum* CECT 7481 *Pediococcus acidilactici* CECT 8633	Lozenge	3×10^3	2 x daily	42 days	42 days	GI, PI	GI	high GI scores
Staab et al. (58)	RCT	Experimental gingivitis	50	2	No	No	*Lactobacillus casei* Shirota	Milk drink	6.5×10^9	1 daily	56 days	60 days	API, PI, PBI serum marker	Not specified	None
Slawik et al. (59)	RCT	Experimental gingivitis	28	2	No	No	*Lactobacillus casei* Shirota	Milk drink	6.5×10^9	1 x daily	28 days	14 days*	PI, GI, BoP GCF volume	Not specified	BoP, GCF volume
Toiviainen et al. (60)	RCT	Gingivitis	60	2	No	No	*Lactobacillus rhamnosus* GG *Bifidobacterium.animalis.subsp. lactis* BB-12	Lozenge	4×10^9	4 x daily	28 days	28 days	Mutans Streptococci PI, GI	Mutans Streptococci	PI, GI
Alanzi et al. (61)	RCT	Gingivitis	108	2	No	Yes	*Lactobacillus rhamnosus* GG *Bifidobacterium.animalis.subsp. lactis* BB-12	Lozenge	2×10^8	2 x daily	28 days	28 days	GI, PI	GI	GI
Keller et al. (62)	RCT	Gingivitis	47	2	No	Yes	*Lactobacillus rhamnosus* PB01 DSM14869 *Lactobacillus curvatus* EB10 DSM32307	Lozenge	1×10^8	2 × daily	28 days	42 days	BoP, PI GCF cytokines	Not specified	None
Kang et al. (63)	RCT	Gingivitis	68	2	Yes	No	*Weissella cibaria* CMU	Lozenge	1×10^8	1 × daily	56 days	56 days	PPD, BoP PI, GI	Not specified	None

API, Approximal Plaque Index; BoP, Bleeding on Probing; CAL, Clinical Attachment Level; GI, Gingival Index; GCF, Gingival Crevicular Fluid; PI, Plaque Index; PPD, Probing Pocket Depth.

[#]*Due to s a significant order-of-use effect only data of the first cross-over period included in the analysis.*

Unsupervised consumption of the probiotic started 14 days before baseline.

TABLE 2 | Randomized controlled clinical trials evaluating the use of probiotics in the management of periodontitis.

References	Type of investigation	Assessed condition	No. subjects	No. groups	Professional scaling	Hygiene instructions	Assessed probiotics	Application mode	Frequency/ day	Frequency	Duration of Intake	Observation Time	Assessed Outcomes	Primary Outcome	Outcomes significantly improved by consumption of probiotic
Invernici et al. (64)	RCT	Periodontitis	41	2	Yes	Yes	*Bifidobacterium animalis. subsp. lactis* HN019	Lozenge	1×10^9	2 x daily	30 days	90 days	CAL, PI, BoP, PPD	CAL	CAL, PPD
Invernici et al. (65)	RCT	Periodontitis	30	2	Yes	Yes	*Bifidobacterium animalis. subsp. lactis* HN019	Lozenge	1×10^9	2 × daily	30 days	90 days	PI, GBI, Adhesion	PI, GBI	PI, GBI
Vivekananda et al. (66)	RCT	Periodontitis	30	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	21 days	42 days	PI, GI, GBI, PPD, CAL	not specified	PI, GI, GBI, CAL PPD
Teughels et al. (67)	RCT	Periodontitis	30	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	84 days	84 days	PPD, CAL	PPD	PPD, CAL
Vicario et al. (68)	RCT	Mild periodontitis	20	2	No	No	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	1 × daily	30 days	30 days	PPD, PI, BoP	not specified	PI, PPD, BoP
Ince et al. (69)	RCT	Periodontitis	30	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	21 days	360 days	PPD, PI, GI, BOP	PPD	PPD, PI, GI, BOP
Tekce et al. (70)	RCT	Periodontitis	40	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	21 days	360 days	PPD, PI, GI, BoP	PPD	PPD, PI, GI, BoP
Theodoro et al. (71)	RCT	Periodontitis smoking	34	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	21 days	90 days	PPD, BoP, CAL	PPD	BoP
Pelekos et al. (72)	RCT	Periodontitis	41	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily @	28 days	180 days	PPD, CAL, BoP	PPD	None
Grusovin et al. (73)	RCT	Periodontitis aftercare	20	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	84 days	360 days	PPD, CAL, BoP	PPD	PPD, CAL, BoP
Vohra et al. (74)	RCT	Periodontitis smokeless tobacco	127	4	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	21 days	180 days	PPD, PI, BoP, CAL	PPD	None
Laleman et al. (75)	RCT	Periodontitis residual pockets	39	2	Yes	Yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	84 days	168 days	PPD, CAL, PI, BoP	PPD	PPD
Pelekos et al. (76)	RCT*	Periodontitis	40	2	Yes	yes	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily @	28 days	180 days	PPD, CAL	PPD	CAL in pockets $\geq$ 5mm
Schlagenhauf et al. (54)	RCT[#]	Periodontitis smoking	35	2	No	No	*Lactobacillus reuteri* DSM 17938/ATCC PTA 5289	Lozenge	2×10^8	2 × daily	42 days	42 days	BoP, CAL, PPD	BoP	BoP, CAL, PPD
Pudgar et al. (77)	RCT	Periodontitis	40	2	Yes	Yes	*Lactobacillus brevis* CECT7489 *Lactobacillus plantarum* CECT7481	Lozenge	2×10^9	1 × daily	84 days	84 days	BoP, PPD, PI, CAL, GBI	PD>4mm +BoP	BoP[§]
Iwasaki et al. (78)	RCT	Periodontitis aftercare	39	2	Yes	Yes	*Lactobacillus plantarum* L-137 (heat-killed)	Capsule	10 mg	1 × daily	84 days	84 days	PPD, PI, GI, BoP	PPD > 4 mm	PPD $\geq$ 4 mm
Yuki et al. (79)	RCT	Periodontitis Intellect. Disability	23	2	No	No	*Lactobacillus rhamnosus* L8020	Milk yogurt	1.5×10^9	1 × daily	90 days	120 days	PMA, GI, PPD	not specified	PMA
Morales et al. (80)	RCT	Periodontitis	28	2	Yes	Yes	*Lactobacillus rhamnosus* SP-1	Sachet	2×10^7	1 × daily	84 days	336 days	CAL, PI, BoP, PPD	CAL	None
Morales et al. (81)	RCT	Periodontitis	47	3	Yes	Yes	*Lactobacillus rhamnosus* SP-1	Sachet	2×10^7	1 × daily	84 days	270 days	CAL, PPD, BoP	CAL	None
Laleman et al. (82)	RCT	Periodontitis	48	2	Yes	Yes	*Streptococcus oralis* KJ3 *Streptococcus uberis* KJ2 *Streptococcus rattus* JH145	Lozenge	3×10^8	2 × daily	84 days	168 days	PPD, CAL, BoP, PI, GI	PPD	None

BoP, Bleeding on Probing; CAL, Clinical Attachment Level; GI, Gingival Index; GBI, Gingival Bleeding Index; PPD, Probing Pocket Depth; PMA, Papillary-Marginal-Attached-Index; PI, Plaque Index.
*Subanalysis of RCT published 2019 by the same authors. [#]Subanalysis of of 35 study participants out of a total of 72 displaying a PSR score $\geq$ 3 at baseline. [§]Significantly negative impact on reduction of pockets > 4 mm.
@Study participants instructed to consume the lozenge prior to brushing their teeth.

Therapeutic Strategies

The administration of the probiotic was either used as an adjunct to established measures of mechanical plaque control, i.e., oral hygiene training and/or professional mechanical plaque removal (PMPR) (47–50, 53, 55–57, 61–63), or was the only intervention performed (51, 52, 54, 58–60).

Results

Both therapeutic approaches led to diverging results. In some of the studies the administration of the probiotic as an adjunct to efficacious mechanical plaque control significantly enhanced the reduction of gingival inflammation (48, 60), while in others only minor or no significant benefits were observed (47, 49). In study cohorts with inadequate mechanical plaque control or in experimental gingivitis trials with cessation of oral hygiene measures, the results also were ambiguous. Kuru et al. (48) for example observed no difference in bacterial plaque mass or extent of gingival inflammation between study patients consuming a *Bifidobacterium lactis subsp. animalis* probiotic and the placebo-consuming controls during the first 4 weeks of the trial, while they were still maintaining an efficacious level of personal oral hygiene. After a 5-day cessation of personal hygiene efforts however, the resulting increase in plaque mass, gingival index and bleeding on probing scores observed in the probiotic-consuming group was significantly lower than in the placebo-consuming controls. This was in contrast to a preceding study by Hallstrom et al. (50), who failed to detect any inhibitive impact of the consumption of a *L.reuteri* DSM 17938/ATCC PTA 5289 probiotic on the development of experimental gingivitis after the cessation of personal plaque control in healthy young volunteers with efficacious personal oral hygiene. However, in two other randomized clinical trials, evaluating the consumption of the identical *L.reuteri* probiotic by pregnant women (52) and navy sailors (54) with poor oral hygiene efficacy and evident clinical signs of established chronic gingivitis, its intake was accompanied by a significant and pronounced decrease of gingival inflammation when compared to the placebo-consuming controls, despite the lack of any concomitant attempt to improve oral hygiene efficacy.

Probiotics in the Management of Periodontitis

Evaluated Study Cohorts

The impact of probiotics administration in the management of periodontitis has also been evaluated in a wider spectrum of different patients, like systemically inconspicuous non-smokers (64, 65, 67, 68, 70), smokers (54, 71), smokeless tobacco users (74), periodontal recall patients (73, 78) or individuals with intellectual disabilities (79). They were also affected by diverging periodontal conditions like previously untreated periodontitis of varying severity, periodontitis aftercare (73, 78) or residual deep periodontal pockets (75).

Evaluated Study Outcomes

In 14 out of 20 RCTs assessed in this review the reduction of pocket depth was the primary study outcome (66–78, 82), followed by the gain of clinical attachment (64, 80, 81) and the

reduction of markers of inflammation, like gingival bleeding or bleeding on probing (54, 65, 79).

Therapeutic Strategies

Predominantly the consumption of the probiotic was used as an adjunct to professional mechanical debridement and oral hygiene training. Only three trials evaluated the administration of the probiotic as the only therapeutic measure (54, 68, 79).

Results

Similar to the situation in the gingivitis studies the results of the periodontitis trials do not display a uniform and consistent pattern. The majority of RCTs using the *L. reuteri* DSM 17938/ATCC PTA 5289 probiotic as an adjunct to mechanical biofilm and calculus removal reported a significant enhancement of pocket closure by the consumption of the probiotic and/or the reduction of periodontal inflammation compared to the intake of a placebo (54, 66–70, 73, 75, 76). Nevertheless, there are some others studies, which either completely failed to detect any significant additional benefit of its use (72) or found only a very limited clinical usefulness in smokers and smokeless tobacco users (71, 74). In the case of the trial by Pelekos et al. (72) this might be possibly attributable to the reported administration scheme. The study participants were instructed to brush their teeth immediately after consuming the lozenge, which might have severely reduced dosage and contact time of the probiotic.

The adjunctive consumption of the other previously mentioned probiotics also led to diverging results. While studies evaluating the impact of *L. rhamnosus* SP-1 (80, 81) or a mixture of different probiotic strains of *Streptococcus oralis, S. uberis* and *S. rattus* (82) in untreated periodontitis patients failed to verify any additional clinical benefit of their regular consumption after scaling and root planing, the intake of *Bifidobacterium lactis* HN019-containing lozenges by contrast was accompanied by a significantly enhanced pocket closure and attachment gain as well as a reduction of periodontal inflammation when compared to the consumption of a placebo (64, 65). Furthermore, also the 12-week consumption of lozenges containing heat-killed *Lactobacillus plantarum* L-137 fragments, known to enhance Th1 helper cell-related immune functions in humans (83), resulted in a significant reduction of residual pocket depth in periodontal recall patients (78).

Meta-Analyses and Systematic Reviews

Confronted with this variety of partially contradictory results, conducting a meta-analysis or a systematic review of all previously described RCTs seems to be the most reasonable way to still be able to draw valid conclusions. A meta-analysis however is not a panacea for the analysis of any complex research question and may also only provide blurred answers when challenged with the task to compare study results being based on substantially diverging study protocols, patient cohorts and administered probiotics, as illustrated by the heterogeneity in the study protocols of the various trials listed in **Tables 1**, **2**. Given the unique pattern of interferences with the host response and the competing microbiota associated with the use of each specific probiotic strain, it appears to be problematic to pool

results obtained by the use of different probiotics in a meta-analysis. It is therefore not surprising, that the conclusions of currently available meta-analyses and systematic reviews on the use of probiotics in the control of gingivitis (84–86) or periodontitis (87–90) are not clear-cut, but vary considerably from "no additional benefit for pocket reduction" (87), to "limited, short term benefits in attachment gain or pocket closure of deeper pockets" (88–90) and "limited benefit for the reduction of gingival inflammation" (84–86).

Controversies Over the Benefits of Probiotics—A Problem of Misunderstanding the Rationale for Their Use?

An often-neglected aspect in the discussion about the benefits of probiotics in the management of gingivitis and periodontitis is the rationale for their use. As stated before, even the most recent guidelines for the therapy of gingivitis and periodontitis are still dominated by the principle of strict mechanical plaque control due to its proven efficacy in clinical practice. It is therefore no coincidence, that the authors of most probiotic trials used the probiotic only as an adjunct to those established mechanical plaque removal measures. Adding the consumption of a probiotic to properly performed mechanical plaque control in complying and systemically healthy patients however promises only little, if any additional short-term benefit to an already very efficacious therapeutic strategy. Efficacious however may not be confounded with cause-related, as the primary driving force for the development of inflammation-promoting oral bacterial dysbiosis is not insufficient mechanical plaque control (91). The actual and unique potential of probiotics for reducing inflammation on a systemic level becomes much more evident in gingivitis and periodontitis patients where the concept of strict mechanical plaque control fails or may not be applicable and in particular in those, who are concomitantly are also affected by altered systemic inflammatory responses for example heavy smokers, patients with systemic chronic-inflammatory diseases but also pregnant women (92). This is clearly reflected by the positive results of some aforementioned trials where the probiotic was given as the only therapeutic measure to individuals with established chronic gingivitis or periodontitis and habitual poor oral hygiene (52, 54, 79). While the impact of mechanical plaque control on plaque-associated inflammation is always strictly local, many commonly used probiotics are known to be capable to downregulate inflammation also on a systemic level as described before (25). An elevated level of systemic inflammation, however, is suspected to be a major factor for

the initiation of periodontitis-promoting bacterial dysbiosis, as suggested by the significantly elevated incidence of periodontal disease in patients suffering from systemic chronic inflammatory conditions like diabetes type 2, rheumatoid arthritis or chronic kidney disease (39–41). As there is evidence from RCTs that the regular consumption of probiotics may also significantly improve the outcome of therapy in these chronic diseases (93–95), their administration may have the potential to become an integral part of innovative, more cause-directed concepts for the management of gingivitis and periodontitis aiming at the primary prevention of bacterial dysbiosis by controlling inflammation not only locally, but also on a systemic level.

Future Directions

As a first future step however it seems to be mandatory, to rectify the evident current lack of commonly agreed study protocols and evaluation criteria for the assessment of the usefulness of probiotics administration in the control of gingivitis and periodontitis by a consensus conference with a broad participation of experts in the fields of periodontology, microbiology, immunology as well as gastroenterology and other subspecialties of internal medicine. This would provide the indispensable foundation for gaining sound and standardized study data from intervention trials, which would subsequently allow the performance of valid and meaningful comparisons and analyses.

CONCLUSIONS

At the present time the use of probiotics in the management of gingivitis and periodontitis is still a young and rapidly developing field with many unanswered questions including a lack of generally agreed recommendations regarding the selection of the appropriate probiotic, as well as the dosage and the duration of its use. Nevertheless, the use of probiotics with an efficacy record proven by randomized controlled clinical trials, may already represent a valuable addition to available therapy options, especially in clinical situations in which established concepts based on plaque control cannot be implemented with the required effectiveness.

AUTHOR CONTRIBUTIONS

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