

Linking Research to Clinical Practice

Probiotics and Oral Health

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The purpose of Linking Research to Clinical Practice is to present evidence based information to clinical dental hygienists so that they can make informed decisions regarding patient treatment and recommendations. Each issue will feature a different topic area of importance to clinical dental hygienists with A BOTTOM LINE to translate the research findings into clinical application.

Twetman S, Keller MK. Probiotics for caries prevention and control. *Adv Dent Res.* 2012;28(2):98-102.

Objective: Modulation of the microbiota for restoring and maintaining health is a growing issue in medical science. A search for relevant clinical trials on the use of probiotic bacteria as a potential and clinically applicable anti-caries measure was performed.

Methods: According to predetermined criteria, papers were selected and key data on study design, sample size, intervention, duration and results were extracted.

Results: Two animal and 19 human studies were retrieved. Most studies were short-term and restricted to microbiological endpoints, and only 3 human studies reported a caries endpoint. A high degree of heterogeneity among the included investigations hampered the analysis. Significant reductions of mutans streptococci in saliva or plaque following daily intake of probiotic lactobacilli or bifidobacteria were reported in 12 out of 19 papers, whereas 3 reported an increase of lactobacilli. Three caries trials in preschool children and the elderly demonstrated prevented fractions between 21 and 75% following regular intakes of milk supplemented with *L. rhamnosus*. No adverse effects or potential risks were reported.

Conclusions: The currently available literature does not exclude the possibility that probiotic bacteria can interfere with the oral biofilm, but any clinical recommendation would be premature. Large-scale clinical studies with orally derived specific anti-caries candidates are still lacking.

Commentary

An increased interest in use of probiotics to foster oral health has been fueled by the marketing of new products, consumer interest in possible preventive and health maintenance benefits, and research to investigate accuracy of claims and effectiveness in oral health care. Concern about the development of resistant strains to antibiotics is also a factor leading to the emergence of new approaches to combating bacterial infections. A joint statement by the United Nations Food and Agricultural Organization and the World Health Organization included the most recent definition of probiotics as, "Live microorganisms which when administered in adequate amounts confer a health benefit on the host."¹ Unfortunately, the term has been used loosely, not always applied to live bacteria in adequate doses to benefit health or to those bacterial species and genera that have been shown scientifically to confer a health benefit. Studies to validate contents and effectiveness of various probiotics are needed. Because dental caries and periodontal disease are caused by different bacteria, different probiotics might be needed to combat those oral diseases.

Regulation of probiotics by the Food and Drug Administration differs from that of antimicrobials, dentifrices or mouthrinses that make therapeutic claims (e.g. anti-gingivitis or anti-caries). The regulation of claims made for probiotics depends upon their intended use and categorization, such as a dietary supplement, so the burden of proof falls with the FDA after marketing of the product. In the case of other therapeutic oral health products, the burden of proof is the responsibility of the sponsor of the product, so research results

are needed to substantiate claims before marketing. This difference requires the oral health professional's diligence in considering the evidence related to the claims made regarding the effectiveness of probiotics in prevention and treatment of oral diseases or maintenance of oral health.

Studies of clinical oral health benefits of probiotics are uncommon. This study focused on the use of probiotic bacteria as an anti-caries measure. It is a review of the literature related specifically to that purpose; therefore, the authors established criteria to guide what published research articles would be included. They decided to include both in vitro (laboratory) and in vivo (live humans) studies with any caries-related outcome measure, such as reductions in mutans streptococci or lactobacilli, known etiologic species in caries, in saliva or oral biofilm or reduced clinical caries in subjects over time. This literature review cannot be classified as a systematic review, the highest level of evidence, because it did not formally evaluate quality of studies included or limit its inclusion criteria to the highest quality of research – randomized clinical trials (RCTs). For that reason, there was a great deal of variability in research findings included in this review. Two animal and 19 human studies were found, most of which were short-term and restricted to microbiological endpoints. Only 3 of the 19 human studies reported a caries endpoint. A reduction in microbes associated with caries cannot be assumed to result in reduced clinical caries unless caries are evaluated clinically over time. Caries clinical trials generally are at least 3 years in length due to the time it takes to develop new carious lesions that can be measured clinically. These longitudinal RCTs are expensive to conduct, so related outcome measures are studied first to test whether there is promise for a particular intervention such as probiotics.

Results showed statistically significant reductions of mutans streptococci (MS) in saliva or plaque at the end of probiotic use reported in 12 of the 19 human studies, of which only 1 was conducted longer than 3 weeks. Probiotics tested included *L. rhamnosus* GG, Lactobacilli mix, bifidobacteria, *L. reuteri* (2 strains) and *L. rhamnosus* LB21, and combinations thereof. Regrowth of caries etiologic bacteria after probiotic usage was not measured in any of these studies. Interestingly, the authors note, lactobacilli only was reported to have increased in 3 studies after daily intake of the probiotic, lactobacilli. The mode of delivery, for example various dairy products versus tablets or lozenges, did not seem to impact the findings. In addition to being short term, most of the stud-

ies also had small sample sizes and did not control and/or define dosages used. Dosages of specific strains of probiotics needed to have beneficial health effects are critical for effectiveness. These weaknesses in study design make definitive conclusions impossible.

If one focuses on the 3 RCTs that were identified in this literature review, findings and clinical endpoints are similar. All investigated strains of *L. rhamnosus* (GG or LB21) that were delivered in milk. One study showed no statistically significant reduction in caries in preschool children after 7 months. The other study of early childhood caries (ECC) showed a significant reduction in ECC after use of milk supplemented with *L. rhamnosus* and 2.5 ppm fluoride, but the effects of the 2 interventions could not be separated, so it is unknown whether the probiotic, fluoride or both affected the outcome of reduced caries. The third study evaluated the effects of probiotics and fluoride on root caries in 4 groups of elderly adults. Findings indicated root caries reversal in all groups compared to the control group with the greatest effect in the probiotic/fluoride group. Perhaps the probiotic bacteria can be considered as an adjunct to fluoride in prevention and control of the caries process, although further study is needed before such a claim can be accurately made with dental hygiene clients who inquire about using probiotics to prevent dental caries. None of the studies reported significant side effects of the probiotics studied. As the authors indicated, there is nothing in the literature to negate the possibility that probiotic bacteria can interfere with the oral biofilm, but any clinical recommendation would be premature. Large-scale RCTs with specific candidates for anti-caries probiotics are lacking.

van Essche M, Loozen G, Godts C, et al. Bacterial antagonism against periodontopathogens. J Periodontol. 2012. [Epub ahead of print].

Background: The aim of the current study was to compare the prevalence of commensal bacteria, with beneficial properties, for healthy and diseased individuals, and additionally to examine the inhibitory effect of some commercial dietary probiotics on periodontopathogens comparing this inhibitory effect with that of orally derived beneficial bacteria.

Methods: Subgingival plaque samples from 35 patients (healthy and periodontitis patients) were analyzed. Growth inhibition of the periodontal pathogens *Porphyromonas gingivalis*, *Prevotella intermedia*, *Fusobacterium nucleatum* and *Aggre-*

Aggregatibacter actinomycetemcomitans was examined using the agar overlay technique and agar well diffusion method. The quantification of the inhibitory effect was checked with the agar well diffusion method.

Results: Using the agar overlay technique the prevalence of strains antagonistic towards *P. gingivalis*, *A. actinomycetemcomitans* and *F. nucleatum* was found to be higher in healthy individuals than in individuals suffering from periodontitis. This could not be validated by the agar well diffusion assay. Compared with the antagonistic activity of the isolated strains, the probiotic strains overall showed a stronger inhibition of the periodontal pathogens.

Conclusion: It was shown that some oral bacteria can cause antagonism towards periodontopathogens and these observations underline the therapeutic potential of applications that stimulate oral health by the application of beneficial effector strains.

Commentary

In periodontal diseases, there is an increase in the quantity of plaque and a shift in bacterial composition towards requisite anaerobic and proteolytic bacteria, many of which are Gram-negative. The host damage that occurs during the disease process is caused by the combined activities of subgingival biofilms and the host responses to these diverse bacterial inhabitants. Limited knowledge is available regarding the effect of probiotics on biofilm-related periodontitis. The oral microbiota is complex and dental biofilms are considered to be difficult therapeutic targets. The current view on the etiology of plaque-related periodontal inflammation considers 3 factors that determine whether disease will develop: a susceptible host, the presence of pathogenic species and the reduction or absence of supposed beneficial bacteria.² The complexity of the etiology, initiation and progression of inflammatory periodontal disease lies at the root of the failure of many previous approaches to eradicate or definitively control the disease, such as local and systemic antimicrobial therapies. Oral probiotics represent a current approach to combat periodontal pathogens by introducing "so called" beneficial bacteria that may have the ability to prevent colonization of pathogenic bacteria in the oral biofilm.

The purpose of this study was two-fold. The first aim was to compare the prevalence of commensal bacteria, with beneficial properties, for healthy and diseased individuals. Commensal bacteria

have a symbiotic relationship in which one species is benefited while the other is unaffected. These researchers wanted to know how many of these bacteria with beneficial properties were present in individuals with periodontal health in comparison to those with periodontal disease. Previous research has shown that periodontally healthy sites have greater numbers of endogenous beneficial species than diseased sites. The second aim was to examine the inhibitory effect of selected commercial dietary probiotics on periodontopathogens by comparing it with that of orally-derived beneficial bacteria. In other words, the goal was to evaluate if dietary probiotics available on the market inhibited or hindered periodontopathogens, pathogenic bacteria identified as capable of producing periodontal disease, and compare those products to beneficial bacteria derived from the oral cavity. Commensal bacteria have been shown to have a beneficial effect on the host response and the growth and colonization of periodontal pathogens in plaque biofilm.

The most common probiotic strains belong to the genera *Lactobacillus* and *Bifidobacterium*; however, probiotic strains have been isolated from several species within each of these genera. The *Lactobacillus* species from which probiotic strains have been isolated include *L. acidophilus*, *L. johnsonii*, *L. casei*, *L. rhamnosus*, *L. gasseri* and *L. reuteri*. Similarly, the *Bifidobacterium* strains include *B. bifidum*, *B. longum* and *B. infantis*.² Dietary *Lactobacillus* strains are most commonly found in milk products such as yogurt, fermented milk (e.g. kefir, buttermilk, acidophilus milk) or cheese with active cultures. *Bifidobacterium* is also found in fermented milk products, as well as fermented teas, such as kombucha, and cultured vegetables like sauerkraut. The *Lactobacillus* strains tested in this study included *L. fermentum* 8900 LMG, *L. casei* Shirota YACULT, *L. casei* Actemel, *L. casei* ACTT-393, *L. paracasei* L 07-21, *L. rhamnosus* Hansen 1968 and *L. rhamnosus* GG.

Subgingival plaque samples were taken from 35 patients (healthy and periodontitis patients) and analyzed for growth inhibition of periodontal pathogens (i.e. *Porphyromonas gingivalis*, *Prevotella intermedia*, *Fusobacterium nucleatum* and *Aggregatibacter actinomycetemcomitans*). Each sample was examined using the agar overlay technique which allows for production of homogeneous bacteria within a thin layer of agar across the surface of an agar plate and the agar well diffusion method to determine the sensitivity of the microbes to the probiotic. The extent of the inhibitory effect also was checked with the agar well diffusion method. Results of the agar over-

lay test showed that the prevalence of isolated strains antagonistic to the periodontal pathogens was greater in samples from healthy individuals; however, this effect could not be verified through the agar well diffusion method. The inhibitory effect of the probiotic strains was greater than the antagonistic effect of the isolated strains indicating that “beneficial” oral bacteria can cause antagonism towards periodontopathogens.

The authors explained that, theoretically, restoring reduced numbers of beneficial bacteria via probiotics might be of interest in the treatment of plaque-related periodontal diseases. Probiotics might not only suppress the emergence of endogenous pathogens (within the host) or prevent the superinfection with exogenous pathogens (from external sources), they might also protect us through the promotion of a beneficial host response. Some oral bacteria act as antagonists to periodontopathogens and inhibit their growth. Probiotics can, easily and with little side effects, reduce the level of indigenous oral microbes, thus they can provide more sites for colonization by probiotic bacteria. This mechanism of action is similar to gastrointestinal and urogenital applications, and these similarities represent an interesting advance in knowledge related to oral health-care.²

Although these findings contribute toward an understanding of the potential inhibitory effect of probiotics, the role of beneficial bacteria in preventing the emergence of pathogenic species and oral health remains unknown. There is a need for additional research to clarify the role of the oral beneficial microbiota, to identify beneficial bacteria and to provide a foundation for large-scale studies on the usefulness of probiotics to maintain or improve oral health. In the meantime, it is premature to inform our patients that probiotics can prevent or cure periodontal disease.

The Bottom Line

There has been a rapid increase of studies published in the literature about probiotics and oral health within the past decade. Clinicians and consumers are encouraged to continue to read new research findings to determine the exact species, dosages and delivery mechanisms that are effective in prevention and control of oral diseases such as dental caries and periodontal disease.

Each of these studies examined the effect of probiotics on oral health, specifically dental caries and periodontal disease. Probiotics have the potential to offer a new mechanism for prevention

of these oral diseases by boosting the beneficial oral immune response and by interfering with the growth and colonization of pathogens. Results add to the body of knowledge about probiotics in the prevention and treatment of these oral diseases; however, they do not provide evidence of the effectiveness of probiotics in combating dental caries or periodontal disease.

Based on the findings of these 2 studies, the following conclusions can be drawn:

- Probiotics have been shown to have a positive effect on the oral immune response and inhibition of pathogens associated with periodontal disease.
- Because probiotics seem to affect the colonization of periodontal pathogens, it is logical to assume their potential lies in the regrowth of plaque following its removal by self-care or professional therapy rather than with decreasing the effects of established periodontopathogens in oral biofilm.
- Future large-scale clinical studies are needed to make clinical recommendations for probiotics as anti-caries agents. Probiotic bacteria might be considered as an adjunct to fluoride in prevention and control of the caries process, although further study is needed before such a claim can be accurately made with dental hygiene clients who inquire about using probiotics to prevent dental caries. Certainly, the use of probiotics in lieu of fluoride therapy should be discouraged.
- Some oral probiotics on the market might make exaggerated claims, and these claims are not monitored by the Federal Trade Commission for probiotics as they are for other dental therapeutic products like dentifrices and mouthrinses containing fluoride or antimicrobials. As a result, dental hygienists need to read research related to the benefits of probiotics in relation to oral health care.
- Probiotics are safe for use by our patients when used as instructed as these studies and others have shown no significant side effects.

Summary

Dental hygienists are preventive professionals responsible for advising their patients and the public about the effects of oral care products and natural interventions. The recent increase in consumer and professional interest in the potential effects of probiotics on oral and systemic health further emphasizes the relationship between oral and systemic health, especially as related to the host immune response and growth of pathogens

in the oral biofilm. Probiotics may reduce the colonization of oral bacteria, similarly to their effect in the gastrointestinal tract, but such an effect would most likely have an impact for regrowth of bacteria after self-care, dental hygiene care, nonsurgical or surgical periodontal therapy rather than with biofilm that is firmly established. Clinical recommendations for probiotics as anti-caries or as periodontal disease therapeutic agents are premature.

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