Review Article

Role of *Streptococcus mutants* Biofilm in Dental Caries

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Abstract

Dental caries is a common chronic disease affecting tooth of many subjects at different ages due to biofilm-mediated oral bacterial infection. *Streptococcus mutans* is regarded as a pivotal cariogenic bacterium responsible for dental caries and plaque formation. It produces exopolysaccharides on the surface of the tooth promoting the colonization of cariogenic bacteria and dental biofilm development. Formation of this biofilm can be prevented by using different antimicrobial agents such as BlueM mouthwash, polypyrrole, natural flavonoids, and endocannabinoid anandamide.

Keywords: Biofilm, dental, mutants, *Streptococcus*

Oral cavity including many diseases such as gingivitis, periodontitis, and dental caries prevalent around the world and people usually suffer from it during their lifetime. These diseases effect individual well-being and oral health. Tooth decay is a multifactorial disease that many factors predispose to it like a high sugar diet, poor oral hygiene practice, disruption of the ecological and biological balance of the oral cavity which either increase or decrease in the number of certain microbiota in the configuration of the microbiota might cause dysbiosis leads to certain oral and systemic diseases.[1] According to the ecology hypothesis in the development of dental caries can be divided into three reversible stages, the first one is dynamic stabilization stage, the second is acid production stage, and the last one is aciduria stage.[2] The pH in the biofilm does not alter due to the metabolic reactions of the different microbial community that remain in the state of equilibrium and there is a small different in the local oral bacterial competitiveness. The occlusal surface is favourable to bacterial adhesion, the sulcus-fosse system, long eruption period, decrease of oral mechanical function, and the metabolic activity in the mouth can become unbalanced. These occlusal surfaces can encourage the build-up of biofilms, which can also develop into cariogenic biofilms, Gtf-derived dextran helps to accumulate bacterial cells. Excessive intake of carbohydrates leads to a more acidic plaque microbiota, increases the acidity of nonproteobacteria, boosted bacterial-fungal sugar metabolism when *Candida albicans* is symbiotic with *Streptococcus mutans* in a sucrose-rich environment.[3] Carbohydrate rich diet enrich acidogenicity plaque microbiota like *S. mutans*, *Lactobacillus*, *Actinomyces* spp. and *Bifidobacterium* which contribute to the low pH plaques and the weak organic acids produced from fermentation carbohydrate metabolism contributes to the demineralization of tooth enamel.[4] In addition to that, carboxyl groups of acidic proline-rich polypeptides help the attaching of oral microbial groups in cell membranes and increase the colonization of tooth surfaces.[5] Other factor like type of saliva components that contributes to biofilm proliferation and formation while colostrum plays the opposite role, and 3'-sialolactose significantly reduces the formation of biofilms.[6] Genetic factors also influence caries predisposition and facilitate sucrose sweetness favorite. Genetic factors constitute about 65% of the individuals with dental caries. Genetic host susceptibility to this disease like mutation in the genes coding for organic matrix materials leads to abnormal protein production, mineralization defects, bacterial adhesion, resistance to acidic environments, and tooth sensitivity. For example, the SPP1 gene that codes for protein contributes to tissue mineralization and remodelling and also

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alteration in genes that affect the susceptibility to dental caries via alteration in the immune responses.[7,8]

Enamel is controlled by the interaction of many organic matrix molecules, and mutations in the genes encoding these organic matrix molecules can lead to the production of abnormal proteins leading to mineralization defects, disturbing bacterial adhesion, increasing tooth sensitivity, and ending in dental caries. Dental caries is associated with loci 13q31.1 and 14q24.3 and resistance to caries is related with loci 5q13.3, 14q11.2, and Xq27.1.[9] Genetic variation in genes like the carbonic anhydrase (CA) VI gene (rs17032907) genetic variant and a haplotype of CA VI (ACA) caused an increase in caries and deficiency of transcriptional repressor GATA binding 1 (TRPS1), a transcription factor involved in tooth development increasing the likelihood of mineral loss under acidic circumstances.[10] Altered immune response is also affected by genetic factors like polymorphisms in the major histocompatibility complex (human leukocyte antigens) might lead to altered immune responses toward the oral bacterial colonization and increased susceptibility to dental caries such as HLA-DR4, HLA-DR3, tumour necrosis factor alpha genes might increase the risk of childhood dental caries.[11] This leads to stimulating the production of pro-inflammatory cytokines such as salivary interleukin-1β and distal-less homeobo × 3 (DLX3), which is important in odontogenesis, and DLX4, which is expressed in dental pulp cells, might play an important role in the dental caries.[12] Actinin alpha 2 involved in tissue-forming cells during tooth enamel formation and had a role in caries susceptibility. Dentin extracellular matrix secretory calcium-binding phosphoprotein genes (SPP1) encoding secreted phosphoprotein 1, MEPE encoding matrix extracellular phosphoglycoprotein, IBSP encoding integrin binding sialoprotein, DMP1 encoding dentin matrix acidic phosphoprotein 1, and DSPP encoding Dentin Sialophosphoprotein involved in mineralized and remodelling that involved in biomineralization.[13] Other factors like dental plaques, environmental factor like decrease fluoride contact, and the composition of oral microbiota (bacteria, viruses, fungi, and other microorganisms) which is a critical factors in the development of this disease that developed after sugar, carbohydrates, and candy sweets consumption that stimulates *S. mutans* secrets many virulence factors through many pathways (sucrose and nonsucrose) pathways with aid of normal flora in the mouth leading to dysbiosis and biofilm formation ending with dental caries.[14] The World Health Organization reports that about half of the population complains of diseases of the oral cavity and about two billion have tooth decay.[15] Dental caries disease (tooth decay or cavity) depends on diet and biofilm formation by cariogenic bacteria on tooth surfaces.[16] The main bacteria is *S. mutans* which was first defined by James Kilian Clarke in 1924 as a Gram-positive coccus, facultatively anaerobic that is frequently found in the human oral cavity causing tooth decay.[17,18] These bacteria live in a sticky biofilm on the surface of the tooth that produced from insoluble glucan with a high concentration of sucrose. In addition, they produce acid from sugar metabolism, bacteriocin, and fructan.[19] Biofilms is an extracellular matrix consists of exopolysaccharides, lipids, proteins, and eDNA that released from cell death or bacterial lysis which act as attachment and adhesive factor among bacteria.[20] Sucrose synthesizes extracellular polysaccharides by glucosyltransferase enzyme that promotes adhesion to tooth surface and acetylation of this enzyme regulates the formation of biofilm and virulence of *S. mutans*.[21,22] Many regulatory mechanisms control the synthesis of biofilm-like c-di-AMP, Ap4A, transcription factors such as EpsR, RcrR, StsR, AhrC, FruR, small RNA for example sRNA0426, srn92532, and srn133489, quorum-sensing signaling system as LuxS, and enzymes including Dex, YidC, CopZ, EzrA, lmrB, SprV, RecA, PdxR, MurI.[23] An enzyme Vick (sensor histidine kinase) is important in regulating genes associated with extracellular polysaccharides and antisense *vicK* RNA (AS*vicK*) bound with *vicK* into double-stranded RNA (dsRNA).[24] Other factors that regulate metabolic pathways and biofilm formation in cariogenic *S. mutans* is lysine lactylation which expressed in the greater number of lactylated sites and proteins.[25] Hence, frequent consumption of sucrose which is the most common dietary carbohydrate that cause an increase in the dental caries occurrence via the action of *S. mutans* in association of poor oral hygiene, socioeconomic status of the patient, hypofunction of the salivary glands, and genetic factors, all play an important role in dental caries.[26] Consumption of some drugs like methamphetamine in drugs abuser persons leads to increased microbial dental diseases like painful gum inflammation and tooth decay ending with loss of teeth. This drug decreases the secretion of saliva, enhances bacterial growth, enhances drinking of sugary drinks, frequent tooth grinding, and absence of common oral hygiene and cleanness. All these will stimulate *S. mutans* growth, tooth adhesion, and biofilm formation on the surface of the tooth through stimulation of *S. mutans* glycosyltransferases enzymes, low PH and lactic acid production. Hence, frequent use of oral rinse with chlorhexidine (CHX) suggestively reduces tooth colonization and decreases dental caries.[27] Other cariogenic bacteria are *Streptococcus sobrinus* that form cariogenic environment with other bacteria such as *Streptococcus salivarius* and *Streptococcus thermophilus*.[28] In addition, *Streptococcus sanguinis* was found in the dentition connective tissue dysplasia and biofilm microbiome that produced hydrogen peroxide that alters the pH of the mouth and increases the growth of cariogenic acid resistance bacteria, and it is the main cause of infective endocarditis.[29] Moreover, *lactobacilli* aid the attachment of bacteria to the surface of the tooth and help in plaque formation, *Streptococcus pyogenes* forms a complex of protein that help in the growth of bacteria, *Streptococcus gordonii* regulates PH by the production of alkali in the mouth and adheres with other bacteria to form plaque like *Streptococcus constellatus* and *Streptococcus pharyngitis*.[30]

Fungi also had a part in plaque formation and called cariogenic fungi like *C. albicans* that interact with other cariogenic

bacteria to form symbiosis and enhance plaque formation by changing the sensitivity of the teeth and penetrate tissues through adherence to specific receptors. It also induces expression of virulence genes by bacteria and enhance the toxicity of the biofilm.[31]

Treatment of Dental Caries

It should be known that a radical treatment for dental caries has not been start but these biofilms were simply removed by regularly tooth and water washing or using BlueM mouthwash that had antimicrobial activity via decrease expression of gbpA gene of *S. mutans* and control biofilm formation.[32] In addition to that using polypyrrole which is an organic conductive polymer formed from pyrrole ring is effective in removing streptococci biofilms.[33] Deletion of dexA gene can modulate biofilm and decrease cariogenicity and could use as probiotic treatment.[34] Moreover, using natural flavonoids like Orientin-2"-O-β-L-galactoside, vitexin, and orientin are antibacterial and inhibit biofilm formation through decrease levels of transcription of *spaP*, *srtA*, *brpA*, *gtfB* and *luxS* genes of *S. mutans*.[35] Other material that used as antibacterial and anti-biofilm formation is natural bioactive lipid material known as endocannabinoid anandamide that decrease thickness of biofilm and growth of bacteria which used as therapeutic agent.[36] Several risk modifiers have been determined to decrease dental caries formation for example fluoride which had an important role in prevention demineralization since 1940s.[37] It's action through formation fluorapatite crystals, remineralising, and antibacterial effect of *S. mutans* and prevent acid formation by these bacteria.[38] The main useful effect of fluoride is during erupted teeth and fluoride intake more than the optimal levels leads to a condition known as dental fluorosis that characterized by decrease mineralization of the tooth, enamel and dentin destruction, and plaque accumulation on the teeth.[39] Hence, fluoride in the normal drinking water act as anticariogenic factor irrespective of the severity of fluorosis and salivary levels of *S. mutans*.[40] One of the important factors in dental caries is bacterial cause, so bacteriotherapy (probiotics) should be one of the important methods in prevention this disease and exert its effect on the bacteria that habitat the mouth as part of gastrointestinal system playing a vital role in preserving oral health.[41] Probiotics are live microorganisms such as strains of *Lactobacillus* and *Bifidobacteria* that act against oral pathogens that cause dental caries and periodontal diseases via different mechanisms via direct and indirect methods. Regarding direct mechanisms that include production of antimicrobial substances that inhibit bacterial growth for example bacteriocins, organic acids, ammonia, fatty acids, and hydrogen peroxide that decrease the number of pathogenic bacteria in the human intestine and in the oral cavity and prevent colonization of these bacteria while, direct action on plaque and biofilm formation through competing and intervening with bacteria to bacteria attachments, competing with oral microorganisms, and involvement in the binding of oral microorganisms to proteins and biofilm formation while the indirect mechanisms which include regulation of mucosal permeability, modulation of the immune system function, oral colonization by less pathogenic bacteria, and effect on local immunity in the oral cavity via nonimmunologic defense mechanisms [Figure 1].[42,43]

For example, *Lactiplantibacillus plantarum* Ln4 successfully inhibited biofilm formation by antimicrobial and antibiofilm effect against *S. mutans* KCTC 5124 and used in the industry as a probiotic to prevent and improve oral health and hygiene.[44]

In addition, nanoparticles were also used in the treatment to restore eubiosis and homeostasis, interfere with bacterial metabolism, inhibit biofilm formation, decrease demineralization like graphene oxide's nanoparticles which had antimicrobial properties and inhibit the growth of dental pathogenic bacteria like *S. mutans* through anti-adherence activity and bactericidal effect.[45,46] Other nanoparticles that supplement to antibiotics are nanobacterial which fill the deficiency of antibiotics due to antibacterial resistance and its good stability.[47]

In addition, there is organic and inorganic nanoparticles like chitosan nanoparticles that used for rinsing root canal, plaque removal, and inhibition reattachment of bacteria and other nanoparticles are quaternary polyethyleneimine which binds proteins and lipid layers in bacterial cell walls and membrane inhibiting the exchange of different substances ending in bacterial death.[48,49]

Other method of treatment using bacterial ecology and quorum sensing which is interbacterial communication systems that permit bacteria to spread data around their environment, metabolism, and other survival information through extracellular signal transmission like pheromones peptides (autoinducer-2 and competence-stimulating peptide) that makes a change in gene expression and series of responses in the body and biofilm formation.[50] Regarding quorum system of *S. mutans*, it had a short hydrophobic peptide/Rgg quorum sensing system that controls a precise biosynthetic manipulator with the radical-SAM (S-adenosyl-l-methionine) (RaS) enzyme and forms a ribosomally synthesized and posttranslationally modified peptides.[51] *S. mutans* coordinates genetic transformation through two peptide pheromones CSP and comX inducing peptide. This quorum system plays an important role in environment conditions of bacteria like oral biofilm formation, genetic capacity, acid production, bacterial virulence activity, and plaque production.[52] Others used many antibiotics and antimicrobial peptides like GH12 that inhibit biofilm formation, inhibit dental caries, inhibits the virulence factors of *S. mutans*, reduces EPS synthesis, and increase the ecological competitiveness of *S. sanguinis* and *S. gordonii*.[53] KSL is another antimicrobial peptide act against different oral bacteria and fungi, inhibit biofilms formation. Cyclic bacteriocins such as Bac8c has a stronger antibacterial activity and reduce cell viability in biofilms formed in BioFlux systems.[54] The specifically targeted antimicrobial peptide

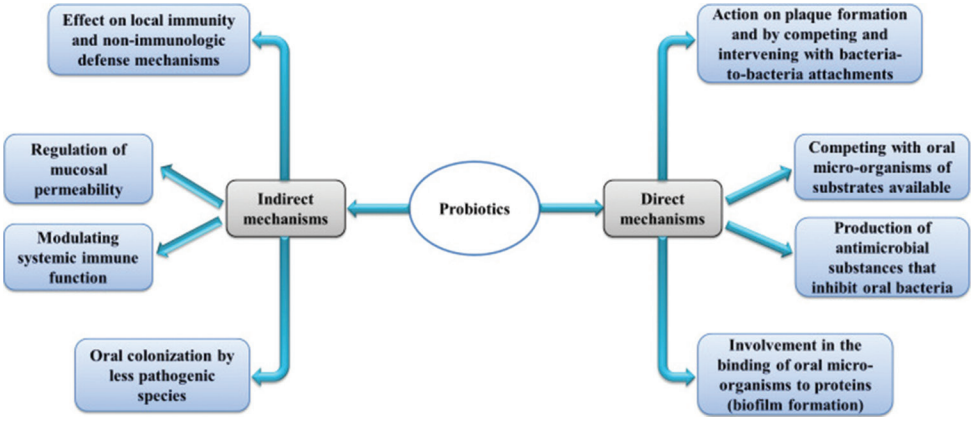

Figure 1: Mechanisms of action of probiotics[43]

which is a synthetic fusion peptide formed from the targeting domain of *S. mutans* known as (C16) and the killing domain of the broad-spectrum antibacterial peptide G2.[55] Another material like chrysophsin-1 which is a cationic bactericidal peptide against Gram-positive and Gram-negative bacteria and pHly-1 which is a pH- and lipid-dependent AMP in conformational transitions that able in killing the acidic oral pathogen *S. mutans* under acidic circumstances more successfully than CHX.[46]

Vaccines against Dental Caries

Vaccines against dental caries are classified as DNA vaccines, subunit vaccines, and mucosal vaccines. Subunit vaccines used part of the genome responsible for the production of Ag I/II or gtf or GBP and intranasal mucosal vaccines induced the production of secretory immunoglobulin A (IgA) through using nasal drops or spray which is more effective and decrease *S. mutans* biofilm production which is more safe and protective. The last one is DNA vaccine that used nanoparticles by combining anionic liposomes into chitosan/DNA complexes that increasing cellular uptake and this vaccine deliver anti-caries DNA vaccine pGJA-P/VAX into the mucosa of the nasal cavity and induces a salivary IgA antibody production and decrease the enamel and dentin damage after infection with *S. mutans*.[56,57] Other type of nasal immunization is fusing *S. mutans* rPAc with C-terminus of *Escherichia coli*-derived flagellin that induce systemic and local mucosal antibody responses which used as preventive and therapeutic measures. Thus, DNA vaccine has become a trend in dental caries vaccine due to its safety, stable antigenic protein expression and antigenicity.[56,58]

At the end, researchers should find a new antigenic virulence genes or proteins for developing a new vaccine with adjuvant and new delivery method.

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Conflicts of interest

There are no conflicts of interest.

References

1. Chen B, Wang Z, Wang J, Su X, Yang J, Zhang Q, *et al*. The oral microbiome profile and biomarker in Chinese type 2 diabetes mellitus patients. Endocrine 2020;68:564-72.
2. Takahashi N. Chapter 11: Future perspectives in the study of dental caries. Monogr Oral Sci 2023;31:221-33.
3. Farkash Y, Feldman M, Ginsburg I, Steinberg D, Shalish M. Polyphenols inhibit *Candida albicans* and *Streptococcus mutans* biofilm formation. Dent J (Basel) 2019;7:42.
4. Neves BG, Stipp RN, Bezerra DD, Guedes SF, Rodrigues LK. Quantitative analysis of biofilm bacteria according to different stages of early childhood caries. Arch Oral Biol 2018;96:155-61.
5. Hay DI, Carlson ER, Schluckebier SK, Moreno EC, Schlesinger DH. Inhibition of calcium phosphate precipitation by human salivary acidic proline-rich proteins: Structure-activity relationships. Calcif Tissue Int 1987;40:126-32.
6. Dukanovic Rikvold P, Skov Hansen LB, Meyer RL, Jørgensen MR, Tiwari MK, Schlafer S. The effect of enzymatic treatment with mutanase, Beta-glucanase, and DNase on a saliva-derived biofilm model. Caries Res 2024;58:68-76.
7. Tulek A, Mulic A, Runningen M, Lillemo J, Utheim TP, Khan Q, *et al*. Genetic aspects of dental erosive wear and dental caries. Int J Dent 2021;2021:5566733.
8. Hasan A, Roome T, Wahid M, Ansari SA, Akhtar H, Jilani SN, *et al.* Gene expression analysis of toll like receptor 2 and 4, Dectin-1, Osteopontin and inflammatory cytokines in human dental pulp *ex-vivo*. BMC Oral Health 2022;22:563.
9. Vieira AR. Genetics and caries: Prospects. Braz Oral Res 2012;26 Suppl 1:7-9.
10. Li ZQ, Hu XP, Zhou JY, Xie XD, Zhang JM. Genetic polymorphisms in the carbonic anhydrase VI gene and dental caries susceptibility. Genet Mol Res 2015;14:5986-93.
11. Opal S, Garg S, Jain J, Walia I. Genetic factors affecting dental caries risk. Aust Dent J 2015;60:2-11.
12. Hu XP, Song TZ, Zhu YY, Wu LL, Zhang X, Zhou JY, *et al.* Association of ENAM, TUFT1, MMP13, IL1B, IL10 and IL1RN gene polymorphism and dental caries susceptibility in Chinese children. J Int Med Res 2019;47:1696-704.
13. Eckert S, Feingold E, Cooper M, Vanyukov MM, Maher BS, Slayton RL, *et al.* Variants on chromosome 4q21 near PKD2 and SIBLINGs are associated with dental caries. J Hum Genet 2017;62:491-6.
14. Dashper SG, Mitchell HL, Lê Cao KA, Carpenter L, Gussy MG, Calache H, *et al.* Temporal development of the oral microbiome and prediction of early childhood caries. Sci Rep 2019;9:19732.
15. Campus G, Cocco F, Ottolenghi L, Cagetti MG. Comparison of ICDAS, CAST, Nyvad's Criteria, and WHO-DMFT for Caries Detection in a Sample of Italian Schoolchildren. Int J Environ Res Public Health 2019;16:4120.
16. Loesche WJ. Role of *Streptococcus mutans* in human dental decay. Microbiol Rev 1986;50:353-80.

17. Thomas VJ, Rose FD. Ethnic differences in the experience of pain. Soc Sci Med 1991;32:1063-6.
18. Nicolas GG, Lavoie MC. *Streptococcus mutans* and oral streptococci in dental plaque. Can J Microbiol 2011;57:1-20.
19. Banas JA. Virulence properties of *Streptococcus mutans*. Front Biosci 2004;9:1267-77.
20. Kawarai T, Narisawa N, Suzuki Y, Nagasawa R, Senpuku H. *Streptococcus mutans* biofilm formation is dependent on extracellular DNA in primary low pH conditions. J Oral Biosc 2016;58:55-61.
21. Ma Q, Pan Y, Chen Y, Yu S, Huang J, Liu Y, *et al*. Acetylation of glucosyltransferases regulates *Streptococcus mutans* biofilm formation and virulence. PLoS Pathog 2021;17:e1010134.
22. Selwitz RH, Ismail AI, Pitts NB. Dental caries. Lancet 2007;369:51-9.
23. Zheng T, Jing M, Gong T, Yan J, Wang X, Xu M, *et al.* Regulatory mechanisms of exopolysaccharide synthesis and biofilm formation in *Streptococcus mutans*. J Oral Microbiol 2023;15:2225257.
24. Sun Y, Chen H, Xu M, He L, Mao H, Yang S, *et al.* Exopolysaccharides metabolism and cariogenesis of *Streptococcus mutans* biofilm regulated by antisense Vick RNA. J Oral Microbiol 2023;15:2204250.
25. Li Z, Gong T, Wu Q, Zhang Y, Zheng X, Li Y, *et al*. Lysine lactylation regulates metabolic pathways and biofilm formation in *Streptococcus mutans*. Sci Signal 2023;16:eadg1849.
26. Tungare S, Paranjpe AG. Diet and nutrition to prevent dental problems. In: StatPearls. Treasure Island (FL): StatPearls Publishing; 2023.
27. Lee HH, Sudhakara P, Desai S, Miranda K, Martinez LR. Understanding the basis of METH mouth using a rodent model of methamphetamine injection, sugar consumption, and *Streptococcus mutans* infection. mBio 2021;12:e03534-20.
28. Li XW, Wang YF, Jiang WT, Zhang LL. Effects of antimicrobial peptide GH12 on the morphology and composition of cariogenic three-species biofilm. Hua Xi Kou Qiang Yi Xue Za Zhi 2021;39:188-94.
29. Vaknin M, Steinberg D, Featherstone JD, Feuerstein O. Exposure of *Streptococcus mutans* and *Streptococcus sanguinis* to blue light in an oral biofilm model. Lasers Med Sci 2020;35:709-18.
30. Chaudhary S, Dhiman A, Patidar A, Malhotra H, Talukdar S, Dilawari R, *et al.* Moonlighting glyceraldehyde e-3-phosphate dehydrogenase (GAPDH) modulates protein aggregation. Biochim Biophys Acta Mol Basis Dis 2021;1867:166202.
31. Xiao J, Moon Y, Li L, Rustchenko E, Wakabayashi H, Zhao X, *et al*. *Candida albicans* carriage in children with severe early childhood caries (S-ECC) and maternal relatedness. PLoS One 2016;11:e0164242.
32. Santos VC, Maquera-Huacho PM, Imbriani MJ, Minhaco VM, Spolidorio DM. Effects of BlueM® against *Streptococcus mutans* biofilm and its virulence gene expression. Braz Dent J 2023;34:19-28.
33. Senpuku H, Tuna EB, Nagasawa R, Nakao R, Ohnishi M. The inhibitory effects of polypyrrole on the biofilm formation of *Streptococcus mutans*. PLoS One 2019;14:e0225584.
34. Yan Y, Hailun H, Fenghui Y, Pingting L, Lei L, Zhili Z, *et al*. *Streptococcus mutans* dexA affects exopolysaccharides production and biofilm homeostasis. Mol Oral Microbiol 2023;38:134-44.
35. Yan L, Zhang S, Zhou X, Tian S. Anti-biofilm and bacteriostatic effects of three flavonoid compounds on *Streptococcus mutans*. Biofouling 2023;39:245-56.
36. Wolfson G, Sionov RV, Smoum R, Korem M, Polacheck I, Steinberg D. Anti-bacterial and anti-biofilm activities of anandamide against the cariogenic *Streptococcus mutans*. Int J Mol Sci 2023;24:6177.
37. Costa Sde M, Abreu MH, Vargas AM, Vasconcelos M, Ferreira e Ferreira E, Castilho LS. Dental caries and endemic dental fluorosis in rural communities, minas gerais, Brazil. Rev Bras Epidemiol 2013;16:1021-8.
38. Buzalaf MA, Pessan JP, Honório HM, Ten Cate JM. Mechanisms of action of fluoride for caries control. Monogr Oral Sci 2011;22:97-114.
39. González-González R, Bologna-Molina R, Molina-Frechero N. Editorial: Fluoride exposure, dental fluorosis, and health. Front Oral Health 2023;4:1256495.
40. Ravuru N, Reginald BA, Reddy BS, Samatha M. Relationship between dental fluorosis, dental caries and salivary levels of *Streptococcus mutans*. J Oral Maxillofac Pathol 2023;27:603.
41. Gruner D, Paris S, Schwendicke F. Probiotics for managing caries and periodontitis: Systematic review and meta-analysis. J Dent 2016;48:16-25.
42. Cheng Y, Liu J, Ling Z. Short-chain fatty acids-producing probiotics: A novel source of psychobiotics. Crit Rev Food Sci Nutr 2022;62:7929-59.
43. Homayouni Rad A, Pourjafar H, Mirzakhani E. A comprehensive review of the application of probiotics and postbiotics in oral health. Front Cell Infect Microbiol 2023;13:1120995.
44. Jang HJ, Kim JH, Lee NK, Paik HD. Probiotic *Lactobacillus plantarum* Ln4 showing antimicrobial and antibiofilm effect against *Streptococcus mutans* KCTC 5124 causing dental caries. J Microbiol Biotechnol 2024;34:116-22.
45. Mohammed RK, Ibrahim AA. The anti-adherence activity and bactericidal effect of GO against *Streptococcus mutans* from Iraqi dental patients. Odontology 2023;111:863-9.
46. Zhang P, Wu S, Li J, Bu X, Dong X, Chen N, *et al.* Dual-sensitive antibacterial peptide nanoparticles prevent dental caries. Theranostics 2022;12:4818-33.
47. Besinis A, De Peralta T, Handy RD. The antibacterial effects of silver, titanium dioxide and silica dioxide nanoparticles compared to the dental disinfectant chlorhexidine on *Streptococcus mutans* using a suite of bioassays. Nanotoxicology 2014;8:1-16.
48. Nehra P, Chauhan RP, Garg N, Verma K. Antibacterial and antifungal activity of chitosan coated iron oxide nanoparticles. Br J Biomed Sci 2018;75:13-8.
49. Makvandi P, Jamaledin R, Jabbari M, Nikfarjam N, Borzacchiello A. Antibacterial quaternary ammonium compounds in dental materials: A systematic review. Dent Mater 2018;34:851-67.
50. Zhu Y, Wang Y, Zhang S, Li J, Li X, Ying Y, *et al.* Association of polymicrobial interactions with dental caries development and prevention. Front Microbiol 2023;14:1162380.
51. Rued BE, Covington BC, Bushin LB, Szewczyk G, Laczkovich I, Seyedsayamdost MR, *et al*. Erratum for rued *et al*., "Quorum sensing in *Streptococcus mutans* regulates production of tryglysin, a novel RaS-RiPP antimicrobial compound." mBio 2021;12:e0256021.
52. Acet Ö, Erdönmez D, Acet BÖ, Odabaşı M. N-acyl homoserine lactone molecules assisted quorum sensing: Effects consequences and monitoring of bacteria talking in real life. Arch Microbiol 2021;203:3739-49.
53. Jiang W, Wang Y, Luo J, Li X, Zhou X, Li W, *et al.* Effects of antimicrobial peptide GH12 on the cariogenic properties and composition of a cariogenic multispecies biofilm. Appl Environ Microbiol 2018;84:e01423-18.
54. Ding Y, Wang W, Fan M, Tong Z, Kuang R, Jiang W, *et al.* Antimicrobial and anti-biofilm effect of Bac8c on major bacteria associated with dental caries and *Streptococcus mutans* biofilms. Peptides 2014;52:61-7.
55. Philip N, Suneja B, Walsh LJ. Ecological approaches to dental caries prevention: Paradigm shift or shibboleth? Caries Res 2018;52:153-65.
56. Patel M. Dental caries vaccine: Are we there yet? Lett Appl Microbiol 2020;70:2-12.
57. Chen L, Zhu J, Li Y, Lu J, Gao L, Xu H, *et al*. Enhanced nasal mucosal delivery and immunogenicity of anti-caries DNA vaccine through incorporation of anionic liposomes in chitosan/DNA complexes. PLoS One 2013;8:e71953.
58. Bao R, Yang JY, Sun Y, Zhou DH, Yang Y, Li YM, *et al.* Flagellin-PAc fusion protein inhibits progression of established caries. J Dent Res 2015;94:955-60.