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Antibacterial efficacy of aloe vera peel extract against streptococcus mutans and P. gingivalis: A preclinical experimental study

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Abstract--Background: The anti-inflammatory, antioxidant, antiviral, antibacterial, antitumor, antiarthritic, and anti-diabetic properties of aloe vera has been well documented in the literature. Considering its antibacterial properties evaluating its benefits as mouth rinse in the oral diseases may facilitate the patients in getting cheaper and safer herbal remedy to clean their oral cavities. Objective: To evaluate the Antibacterial efficacy of aloe vera peel extract against Streptococcus mutans and P. gingivalis. Methodology: It was a preclinical experimental study conducted at Baqai Medical and Dental College Karachi, from February to March 2023. Calculated sample size was n =60. The participants were given an envelope for group randomization.

S. mutant samples and *P. gingivalis* were incubated on appropriate media. The Aloe Vera leaves were washed and gel was removed the peels were shed dried and lastly was grinded to powder form. Ethanol (70%) were used for soaking for 15 days with intermittent shaking. The extract was diluted in distilled water in 1:4 (Extract: Distilled water) concentration. Study participants were divided into three groups (negative control, positive control, aloe vera peel extract rinse group) Diluted aloe vera peel extract was given to experimental group for rinses, distilled water was given to negative control group and positive controls were given a standard commercially available mouth rinse. Next sample of plaque was collected after two hours to observe the effects of aloe vera peel extract on bacterial colonies. Results: The paired analysis showed significant (p-value <0.05) decrease in number of colonies in positive control group (conventional rinse) and aloe vera peel extract group. The intra group comparison of negative and positive control showed significant difference in number of colonies and same was observed with the aloe vera peel extract rinse. Conclusion: The aloe vera peel extract have comparable antibacterial activities as that of commercially available mouth rinse.

Keywords--Antibacterial efficacy, aloe vera peel extract, *Streptococcus mutans*, *P. gingivalis*.

Introduction

Aloe vera, from the *Liliaceae* family, is a well-known plant and has been used for centuries in medicine for the treatment of various ailments ¹. Its name has been derived from the Arabic word “*Alloeh*” which means a shining and bitter substance ². Aloe vera is a cactus-like plant that grows widely in hot and dry regions around the globe ³. Various recent studies have reported its anti-inflammatory, antioxidant, antiviral, antibacterial, antitumor, antiarthritic, and anti-diabetic properties ^{4,5}. Additionally, Aloe vera has also been found curative in gastrointestinal disorders, constipation and immune deficiency problems ⁶. Reportedly, 75 bioactive compounds are present in Aloe vera leaves possessing different variety of remedial properties ⁷. Bioactive saponins, phenols, sterols, anthraquinones and lignin are the major compounds of Aloe vera ⁸. Various vitamins like vitamin A, vitamin C, vitamin E and vitamin B₁₂ are also widely present in Aloe vera leaves ⁹. Minerals like magnesium, iron, calcium, sodium, potassium and zinc are also part of the bioactive machinery of Aloe vera leaves ¹⁰.

Recent literature has reported that various oral diseases, including dental caries, periodontitis and gingivitis, have a significant global burden of disease. Numerous other systemic illnesses, like as cardiovascular conditions, have also been linked to periodontal infections ¹¹. The initiation and progress of dental caries or gingivitis depends on various oral pathogens and the progression of these diseases ultimately leads to a worse oral pathology, that is periodontitis ¹². *Streptococcus mutans* and *lactobacilli* are two examples of gram-positive bacteria that are primarily responsible for the initiation of dental decay ¹³. While multiple gram-negative anaerobic bacteria, such as *Porphyromonas gingivalis*,

Porphyromonas endodontalis, and *Tannerella forsythia*, are linked to the chronic progression of periodontal disease ¹⁴. Collectively, these bacteria cause enamel decay and inflammation of gingiva along with supporting structures that eventually leads to periodontitis ¹⁵.

In our study, we have evaluated the effects of Aloe vera peel extract on the growing colonies of *S. mutans* and *P. gingivalis*. One of the many distinct kinds of bacteria that can be discovered in the oral cavity and the most prevalent and virulent contributor to the oral microbiome is *Streptococcus mutans* ¹⁶. While *P. gingivalis* has the capacity of biofilm formation over gingival epithelial cells and calcified hard tissues, and *P. gingivalis* is described as a "keystone" pathogen in periodontitis ¹⁷. That is why it is important to maintain a good oral hygiene and prevent growth of these bacteria by brushing, flossing and using antibacterial mouthwashes ¹⁸. Various commercially available mouth rinses contain chlorhexidine as their major antibacterial compound ¹⁹. Chlorhexidine has a very good antibacterial activity but has also multiple adverse effects that leads to loss of compliance ²⁰. Brown or yellow staining of the teeth, a metallic taste after washing, dry mouth, irritation of the oral mucosa, and occasionally allergic responses are also recorded as side effects of using chlorhexidine mouthwash ^{21, 22}. Thus, there is a need of newer antibacterial agent that have equal or better antibacterial activity as compared to chlorhexidine, but should have least adverse effects. This will reduce various adverse effects and will improve patient compliance.

Methodology

It was a preclinical experimental study conducted at Baqai Medical and Dental College Karachi., from February to March 2023. Calculated sample size was n =60. Consecutive sampling technique was used to recruit the participants. The participants were given an envelope for group randomization. Film of plaque from labial surface of teeth of study participants was collected on sterile strips that was transported to laboratory for culture in sterile containers. For culture *S. mutant* samples were inoculated in Columbia Agar with 5% sheep blood and incubated for 48 h at 37 °C and increased level of CO₂. *P. gingivalis* were grown in wilkins-Chalgren anaerobic broth under anaerobic conditions of 5% CO₂, 10% H₂, and 85% N₂ at 37 °C. All bacteria were sub-cultured twice and were grown to the early stationary phase. Aloe Vera were purchased from local market of Karachi and authentication number i.e. Specimen voucher 10052 was allotted. The leaves were washed and gel was removed the peels were shed dried and lastly was grinded to powder form. The powdered peel was soaked in 2500mL of 70% ethanol for 15 days with intermittent shaking. After 15 days the filtrate was filtered with Whatman filter paper (number 1) that was further processed at 60°C by using water bath. The mixture was than dried at 50°C until a well concentrated extract was produced on rotary evaporator. The extract was kept in an airtight bottle and stored in a refrigerator till usage. The extract was diluted in distilled water in 1:4 (Extract: Distilled water) concentration. Study participants were instructed to not brush their teeth before sampling. Study participants were divided into three groups (negative control, positive control, aloe vera peel extract rinse group) each group had 20 participants. Diluted aloe vera peel extract was given to experimental group for rinses, distilled water was given to negative control group

and positive controls were given a standard commercially available mouth rinse. Next sample of plaque was collected after two hours to observe the effects of aloe vera peel extract on bacterial colonies. ANOVA followed by post hoc tukey's test was applied to identify the inter and intra group comparison and Paired t test was applied as test of significance for pre and post experimental comparison, <0.05 p-value was considered as significant at 95% confidence interval.

Results

There were sixty participants in the study 39 (65%) were males and 21 (35%) were females the mean age of participants was 29 ± 2.31 . On asking about brushing habit 49 (81.6%) participants responded that they brush their teeth daily. The growth of colonies was calculated on growth media plates in samples collected prior to rinsing and samples that were taken after the rinsing. The pre and post samples showed significant (p-value <0.05) decrease in number of colonies in positive control group (conventional rinse) and aloe vera peel extract group as shown in table 1. The intra group comparison of negative and positive control showed significant difference in number of colonies and same was observed with the aloe vera peel extract rinse. However, the positive control and aloe vera peel extract comparison was insignificant. Table 2 shows the intra group comparison of experiment.

Table. 1 Paired t test analysis showing the number of colonies before and after intervention

	Negative Control	Positive control	Aloe vera peel extract
<i>Streptococcus Mutans</i>			
Before	$8.3 \pm 2.1 \times 10^4$	$9.3 \pm 2 \times 10^4$	$9.76 \pm 2.2 \times 10^4$
After	$7.9 \pm 1.3 \times 10^4$	$5.23 \pm 1 \times 10^4$	$5.14 \pm 1.6 \times 10^4$
P value	0.831	0.002*	0.001*
<i>P. gingivalis</i>			
Before	$19.2 \pm 3.1 \times 10^3$	$18.32 \pm 2.75 \times 10^3$	$17.23 \pm 3.62 \times 10^3$
After	$17.45 \pm 1.2 \times 10^3$	$11.71 \pm 3.35 \times 10^3$	$10.23 \pm 1.56 \times 10^3$
P value	0.538	0.041*	0.001*
*significant p-value			

Table 2. ANOVA followed by post hoc tukeys` s Analysis on post interventional results

Groups wise comparison <i>Streptococcus Mutans</i>				p- value
Negative control	$7.9 \pm 1.3 \times 10^4$	Positive control	$5.23 \pm 1 \times 10^4$	0.003*
Negative control	$7.9 \pm 1.3 \times 10^4$	Aloevera peel extract	$5.14 \pm 1.6 \times 10^4$	0.001*
Positive control	$5.23 \pm 1 \times 10^4$	Aloevera peel extract	$5.14 \pm 1.6 \times 10^4$	1.000
Groups wise comparison <i>P. gingivalis</i>				
Negative control	$17.45 \pm 1.2 \times 10^3$	Positive control	$11.71 \pm 3.35 \times 10^3$	0.021*
Negative control	$17.45 \pm 1.2 \times 10^3$	Aloevera peel extract	$10.23 \pm 1.56 \times 10^3$	0.001*
Positive control	$11.71 \pm 3.35 \times 10^3$	Aloevera peel extract	$10.23 \pm 1.56 \times 10^3$	0.036*

*significant p-value

Discussion

Due to its numerous health advantages, including its emollient, anti-inflammatory, purgative, anti-microbial, aphrodisiac, antifungal, and antioxidant properties, aloe vera attracts the interest of the food business²³. As previously mentioned, aloe vera has multiple constituents that have various medicinal effects which were reported in previous literature. One of the most active compounds of Aloe vera is *aloin* that is a type of anthraquinone²⁴. Reportedly, its metabolite aloe-emodin anthraquinone has been associated with several adverse effects like carcinogenicity, genotoxicity and nephrotoxicity. Besides this, few other alkaloids and polysaccharides are reported to cause hepatotoxicity and male infertility, respectively²⁵. A study conducted by Jun wu et al. reported no significant subacute toxicity, acute toxicity and genotoxicity up to a concentration of 3330mg/kg BW when treated with Aloe vera soft capsules²⁶. Another study reported no hepatotoxicity and nephrotoxicity when treated with 500mg/kg BW aqueous extract of Aloe vera gel²⁷.

Our results have reported a remarkable antibacterial activity against *S. mutans* and *P. gingivalis* when treated with conventional mouthwash. A significant decrease in bacterial colonies was observed upon treatment with chlorhexidine containing standard mouthwash. Similar results were obtained in another study that showed potential bactericidal activity against various oral pathogens for subgingival use in the treatment of chronic periodontitis²⁸. A notable suppression was observed in *S. mutans* colonies when treated with chlorhexidine mouthwash and ethanolic licorice root extract, in a study done by Malvania et al.²⁹. Various other studies have also reported antibacterial activity of chlorhexidine containing mouthwashes against *S. mutans*³⁰⁻³³. *P. gingivalis* growth was also suppressed in various studies when treated with chlorhexidine³⁴⁻³⁶. Therefore, in our study we have compared Aloe vera peel extract with already proven antibacterial agent that is chlorhexidine containing mouthwash.

The Aloe vera peel extract group showed a significant decline in the number of bacterial colonies that showed its remarkable antibacterial activity. Our results also showed a significant difference between Aloe vera peel extract group and non-treated group where it showed a high number of *S. mutans* and *P. gingivalis* colonies. A previous in vitro study that was done to evaluate the antibacterial and antifungal properties of Aloe vera plant has reported a remarkable bactericidal and antifungal effects of both Aloe vera root and leaves extracts against various bacteria and fungi ³⁷. Growth of *E. Coli* and *K. Pneumoniae* was also suppressed in another study when treated with Aloe vera extract ³⁸.

Our results have reported comparable and non-significant antibacterial activity between Aloe vera peel extract group and chlorhexidine mouthwash group. In our results the maximum antibacterial activity was observed in our herbal-treated group with Aloe vera. A similar study compared the antibacterial activity of standard chlorhexidine containing mouthwash with different herbs like Aloe vera, green tea and chamomile. Contrary to our results, this study showed that maximum antibacterial activity was possessed by chlorhexidine containing mouthwash when compared with other herbal groups including Aloe vera ³⁹. Similar to our results, another study has reported a notable antibacterial effect of Aloe vera gel against *S. mutans* and *Lactobacilli* ⁴⁰. Based on the antibacterial and anti-inflammatory properties, the natural ingredients propolis, aloe vera, green tea, cranberry, calendula, myrrh, and salvia showed a great potential to be utilized as oral hygiene products ⁴¹. Regarding the discoloration of acrylic dentures due to chlorhexidine, a study has reported that Aloe vera is a better cleaning and antibacterial agent that can be used by patients with acrylic dentures ⁴². Another study reported a decrease in plaque index and bleeding on probing score without any adverse effects when treated with Aloe vera mouthwash ⁴³. Similar results were obtained when chlorhexidine and Aloe vera mouthwashes were compared. Both solutions reduced gingival inflammation, gingival bleeding and plaque accumulation in orthodontic patients ⁴³.

The previous literature and our results conclude that chlorhexidine despite being a gold standard antimicrobial agent, can be replaced by herbal mouthwashes like Aloe vera in patients that are seeking for an indigenous, chemical-free and patient-friendly oral hygiene.

Conclusion

The aloe vera peel extract have comparable antibacterial activities as that of commercially available mouth rinse. Further studies should be conducted to validate its role as antibacterial mouth rinse.

References

1. Saleem A, Naureen I, Naeem M, Murad HS, Maqsood S, Tasleem G. Aloe vera gel effect on skin and pharmacological properties. *Sch Int J Anat Physiol*. 2022;5(1):1-8.
2. Kumar A, Zechariah J, Ramchandra S, Tripathi P. Study on Aloe vera leaves farming in Rajasthan. *Pharma Innov J*. 2022;11(5S):289-91.

3. Otiemo PO. A review on antiplasmodial potential and quantification of aloin and aloe-emodin in aloe vera. 2022.
4. Akbari B, Baghaei-Yazdi N, Bahmaie M, Mahdavi Abhari F. The role of plant-derived natural antioxidants in reduction of oxidative stress. *BioFactors*. 2022;48(3):611-33.
5. Prasad S, Srivastava SK. Oxidative stress and cancer: antioxidative role of Ayurvedic plants. *Cancer: Elsevier*; 2021. p. 301-10.
6. Qadeer M, Itrat N, Iftikhar N, Ahmed U. Evaluation of Hypoglycemic Impact of Aloe Vera in Diabetic Subjects. vol. 2019;4:835-8.
7. Martínez-Burgos WJ, Serra JL, Marsiglia F RM, Montoya P, Sarmiento-Vásquez Z, Marin O, et al. Aloe vera: From ancient knowledge to the patent and innovation landscape—A review. *South African Journal of Botany*. 2022.
8. Ebrahim AA, Elnesr SS, Abdel-Mageed M, Aly M. Nutritional significance of aloe vera (*Aloe barbadensis* Miller) and its beneficial impact on poultry. *World's Poultry Science Journal*. 2020;76(4):803-14.
9. Hagar S. Effect of the Infestation by *Myzus persicae* (Sulzer) and *Tetranychus urticae* Koch on the Internal Components of Aloe Vera Cactus, *Aloe barbadensis* (Miller). *Egyptian Academic Journal of Biological Sciences A, Entomology*. 2021;14(4):117-22.
10. Said F, Rahmawati I, Ningsih NS. Analysis of Metabolite Levels, Secondary Minerals and Aloe Vera Formulation from Kalimantan Indonesia. *Medical Laboratory Technology Journal*. 2022;8(1):27-34.
11. Hugo FN, Bailey JA, Stein C, Cunha ARd, Iser BPM, Malta DC, et al. Prevalence, incidence, and years-lived with disability due to oral disorders in Brazil: an analysis of the Global Burden of Disease Study 2019. *Revista da Sociedade Brasileira de Medicina Tropical*. 2022;55:e0284-2021.
12. Nyvad B, Takahashi N. Integrated hypothesis of dental caries and periodontal diseases. *Journal of oral microbiology*. 2020;12(1):1710953.
13. Mallya P, Mallya S. Microbiology and clinical implications of dental caries—a review. *J Evol Med Dent Sci*. 2020;9:3670-5.
14. Ji S, Kook J-K, Park S-N, Lim YK, Choi GH, Jung J-S. Characteristics of the salivary microbiota in periodontal diseases and potential roles of individual bacterial species to predict the severity of periodontal disease. *Microbiology Spectrum*. 2023;11(3):e04327-22.
15. Adelfio M, Martin-Moldes Z, Erndt-Marino J, Tozzi L, Duncan MJ, Hasturk H, et al. Three-Dimensional Humanized Model of the Periodontal Gingival Pocket to Study Oral Microbiome. *Advanced Science*. 2023;10(12):2205473.
16. Amissah F, Andey T, Ahlschwede KM. Nanotechnology-based therapies for the prevention and treatment of *Streptococcus mutans*-derived dental caries. *Journal of oral biosciences*. 2021;63(4):327-36.
17. Kugaji MS, Kumbar VM, Peram MR, Patil S, Bhat KG, Diwan PV. Effect of Resveratrol on biofilm formation and virulence factor gene expression of *Porphyromonas gingivalis* in periodontal disease. *Apmis*. 2019;127(4):187-95.
18. Patil SS, Yadav AR, Chopade A, Mohite S. Design, development and evaluation of herbal mouthwash for antibacterial potency against oral bacteria. *Journal of University of Shanghai for Science and Technology*. 2020;22(11):881-98.1137.
19. Bescos R, Ashworth A, Cutler C, Brookes ZL, Belfield L, Rodiles A, et al. Effects of Chlorhexidine mouthwash on the oral microbiome. *Scientific reports*. 2020;10(1):5254.

20. Lorusso F, Tartaglia G, Inchingolo F, Scarano A. Early Response and Clinical Efficacy of a Mouthwash Containing Chlorhexidine, Anti Discoloration System, Polyvinylpyrrolidone/Vinyl Acetate and Sodium DNA in Periodontitis Model: A Triple-Blind Randomized Controlled Clinical Trial. *Dentistry Journal*. 2022;10(6):101.
21. Bagaria A, Garg V, Kumar K. *Oral Hygiene The Pulse of the Practice: DENTOMED PUBLICATION HOUSE; 2022.*
22. Deus FP, Ouanounou A. Chlorhexidine in dentistry: pharmacology, uses, and adverse effects. *International dental journal*. 2022;72(3):269-77.
23. Sonawane SK, Gokhale JS, Mulla MZ, Kandur VR, Patil S. A comprehensive overview of functional and rheological properties of aloe vera and its application in foods. *Journal of Food Science and Technology*. 2021;58:1217-26.
24. Sadiq U, Gill H, Chandrapala J. Temperature and pH Stability of Anthraquinones from Native Aloe vera Gel, Spray-Dried and Freeze-Dried Aloe vera Powders during Storage. *Foods*. 2022;11(11):1613.
25. Jangra A, Sharma G, Sihag S, Chhokar V. The dark side of miracle plant-Aloe vera: A review. *Molecular Biology Reports*. 2022;49(6):5029-40.
26. Wu J, Zhang Y, Lv Z, Yu P, Shi W. Safety evaluation of Aloe vera soft capsule in acute, subacute toxicity and genotoxicity study. *PloS one*. 2021;16(3):e0249356.
27. Abderrahmane F, Addou S, Labadie I, Kheroua O. Acute toxicity of the aqueous extract of Aloe barbadensis miller gel. *Bangladesh Journal of Scientific and Industrial Research*. 2023;58(1):19-34.
28. Polizzi E, Teti G, Bova F, Pantaleo G, Gastaldi G, Cappara P, et al. Antibacterial properties and side effects of chlorhexidine-based mouthwashes. A prospective, randomized clinical study. *Journal of Osseointegration*. 2020;12(1):2-7.
29. Malvania EA, Sharma AS, Sheth SA, Rathod S, Chovatia NR, Kachwala MS. In vitro analysis of licorice (*glycyrrhiza glabra*) root extract activity on streptococcus mutans in comparison to chlorhexidine and fluoride mouthwash. *J Contemp Dent Pract*. 2019;20(12):1389-94.
30. Badri SM, Felemban EH, Alnajjar GK, Alotaibi FM, Aljahdali ST, Maher YA, et al. Effectiveness of probiotic lozenges and Chlorhexidine mouthwash on plaque index, salivary pH, and Streptococcus mutans count among school children in Makkah, Saudi Arabia. *The Saudi Dental Journal*. 2021;33(7):635-41.
31. Mensitieri F, Caggiano M, Gaudino G, Charlier B, Coglianesi A, Amato A, et al. In Vitro Evaluation of Antibacterial and Antibiofilm Activity of Different Chlorhexidine-Containing Mouthwash Formulations against Streptococcus mutans. *Applied Sciences*. 2023;13(13):7531.
32. Gowtham A, Badakar C, Hugar S, Gokhale N, Davalbhakta R, Joshi R. Comparative evaluation of antibacterial efficacy of green coffee bean extract mouthwash and chlorhexidine mouthwash against Streptococcus mutans and Lactobacilli spp.-An in vitro study. *Indian Journal of Health Sciences and Biomedical Research*. 2020;13(2):147-.
33. Mehdipour A, Ehsani A, Samadi N, Ehsani M, Sharifinejad N. The antimicrobial and antibiofilm effects of three herbal extracts on Streptococcus mutans compared with Chlorhexidine 0.2%(in vitro study). *Journal of Medicine and Life*. 2022;15(4):526.

34. Joseph RA, Sabarish R, Muthukumar S, Bhat K, Balaji S. Comparative Evaluation on the effect of Herbal mouthwash on Putative Periodontal Pathogens–In vitro study. *Research Journal of Pharmacy and Technology*. 2023;16(1):97-102.
35. Yavagal PC, Rajeshwar SS. Antibacterial Efficacy of Trachyspermum ammi Oil Against Porphyromonas gingivalis, Fusobacterium nucleatum, and Aggregatibacter actinomycetemcomitans: An: In vitro: Study. *Journal of Indian Association of Public Health Dentistry*. 2022;20(4):398-402.
36. Barrak I, Baráth Z, Tián T, Venkei A, Gajdács M, Urbán E, et al. Effects of different decontaminating solutions used for the treatment of peri-implantitis on the growth of Porphyromonas gingivalis-an in vitro study. *Acta Microbiologica et Immunologica Hungarica*. 2021;68(1):40-7.
37. Danish P, Ali Q, Hafeez M, Malik A. Antifungal and antibacterial activity of aloe vera plant extract. *Biological and Clinical Sciences Research Journal*. 2020;2020(1).
38. Jokhio U, Buriro RS, Bughio S, Soomro SA, Arain MB, Soomro AG, et al. 4. An in-vitro antibacterial activity of Aloe vera and gentamicin against Escherichia coli and Klebsiella pneumoniae isolates from mastitis milk samples, from Tandojam, Sindh Pakistan. *Pure and Applied Biology (PAB)*. 2021;11(2):411-20.
39. Naghsh N, Moghareabed A, Nematnejad M, Yaghini J, Sadeghi SM. A comparative evaluation of the antimicrobial effect of chamomile, Aloe vera-green tea, and chlorhexidine mouthwashes on some oral bacterial species. *Dental Research Journal*. 2023;20(1):70.
40. Hajiahmadi M, Faghri J, Salehi Z, Heidari F. Comparative evaluation of antibacterial effect of propolis and aloe vera, xylitol, and Cpp-Acp gels on Streptococcus mutans and Lactobacillus in vitro. *International journal of dentistry*. 2021;2021.
41. Figueiredo LC, Figueiredo NF, Cruz DFd, Baccelli GT, Sarachini GE, Bueno MR, et al. Propolis, Aloe Vera, Green Tea, Cranberry, Calendula, Myrrha and Salvia Properties against Periodontal Microorganisms. *Microorganisms*. 2022;10(11):2172.
42. Mahboub F, Nourizadeh A, Izadpanah A. The Comparison of Color Stability of Aloe Vera Gel and Chlorhexidine Solution on Acrylic Teeth. *International Journal of Dentistry*. 2022;2022.
43. Kamath DG, Nadimpalli H, Nayak SU, Rajendran V, Natarajan S. Comparison of antiplaque and anti-gingivitis effects of aloe vera mouthwash with chlorhexidine in fixed orthodontic patients—A randomized controlled trial. *International Journal of Dental Hygiene*. 2023;21(1):211-8.