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Efficacy of Probiotics, Cetylpyridinium Chloride, and Enzymes-Based Toothpaste Formulations Against Periodontal Pathogens in Fixed Orthodontic Patients: A Review

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Abstract

Introduction: Fixed orthodontic appliances complicate oral hygiene and promote biofilm accumulation, increasing the risk of periodontal disease. Toothpaste formulations with antimicrobial properties such as *Lactobacillus paracasei* probiotics, cetylpyridinium chloride (CPC), and amyloglucosidase–glucose oxidase enzymes are commonly used to manage oral biofilm and control pathogenic bacteria. However, the extent of their effectiveness against periodontal pathogens in orthodontic patients remains unclear. **Purpose:** This review aims to map and summarize the current evidence on the efficacy of various toothpaste formulations in reducing periodontal pathogens among patients undergoing fixed orthodontic treatment. **Methods:** A comprehensive search of electronic databases (e.g., PubMed, Scopus, Google Scholar) was conducted to identify relevant studies published from 2015 to 2025. Data were extracted and synthesized narratively, focusing on types of formulations, study outcomes, and microbial targets. **Results:** Probiotic, enzymatic, and CPC-containing toothpastes showed variable but complementary effects against key pathogens, including *Porphyromonas gingivalis* and *Aggregatibacter actinomycetemcomitans*. Probiotics modulate microbial balance, enzymes disrupt biofilm structure, and CPC provides chemical antimicrobial action. Combining these mechanisms appears promising for enhanced biofilm control. **Conclusion:** Toothpastes containing *L. paracasei* probiotics, CPC, and amyloglucosidase–glucose oxidase enzymes offer complementary ways to control periodontal pathogens in orthodontic patients. Combining these mechanisms may enhance biofilm control and microbial balance, though further research is needed to confirm their long-term benefits.

Keywords: antimicrobial effect; fixed orthodontic appliances; oral pathogens; toothpaste formulations

Introduction

Fixed orthodontic appliances are widely used to correct malocclusions and improve dental aesthetics.[1] However, these appliances create numerous retention sites that hinder effective plaque removal, making it difficult for patients to maintain optimal oral hygiene. As a result, orthodontic treatment often leads to increased biofilm accumulation and inflammation of the surrounding periodontal tissues, particularly the gingiva.[2–4]

Wearing fixed orthodontic appliances can lead to more biofilm accumulation, which shifts the oral microbiota toward more harmful bacteria. Periodontal pathogens like *Porphyromonas gingivalis* and *Aggregatibacter actinomycetemcomitans* may increase, raising the risk of gingivitis and even periodontitis if not controlled. Therefore, managing bacterial biofilm is crucial to maintain good periodontal health during orthodontic treatment.[5,6]

Toothpaste is one of the most accessible and commonly used oral hygiene tools. Various formulations, including those containing probiotics, enzymes, and cetylpyridinium chloride have been developed to enhance antimicrobial activity and improve periodontal outcomes.[7–9] Despite their widespread use, evidence on their ability to reduce periodontal pathogens in orthodontic patients remains limited, as existing studies are scattered, heterogeneous, and often restricted to in vitro experiments or clinical trials. This gap underscores the need for a comprehensive scoping review to evaluate and map existing research on this topic. This review explores the range of toothpaste formulations studied for their antimicrobial effects on periodontal pathogens in patients with fixed orthodontic appliances.

Methods

A comprehensive search of electronic databases such as PubMed, Scopus, Google Scholar was conducted to identify relevant studies published from 2015 to 2025 by using the Boolean Search such as ["Porphyromonas gingivalis" OR "P. gingivalis" OR "Aggregatibacter actinomycetemcomitans" OR "A. actinomycetemcomitans" OR "periodontal pathogens" OR "periodontopathogenic bacteria"] AND ["Probiotics toothpaste" OR “probiotic toothpaste” OR "Lactobacillus paracasei toothpaste" OR "L. paracasei toothpaste"] AND ["Cetylpyridinium chloride" OR “cetylpyridinium” OR “CPC”] AND ["Enzyme-based toothpaste" OR "enzymatic toothpaste" OR "amyloglucosidase" OR "glucose oxidase" OR "enzymes-based formulations"] AND ["fixed orthodontic appliances" OR "fixed orthodontic" OR "fixed appliance"] AND ["bacterial reduction" OR "biofilm reduction" OR "antimicrobial effect" OR "antibacterial effect" OR "dental plaque"].

This review included studies involving patients with fixed orthodontic appliances or laboratory models that mimic orthodontic biofilm conditions. Eligible studies examined probiotics such as *Lactobacillus paracasei*, cetylpyridinium chloride (CPC), or enzyme-based toothpastes containing amyloglucosidase and glucose oxidase, and reported outcomes related to *Porphyromonas gingivalis* or *Aggregatibacter actinomycetemcomitans*. Randomized trials, clinical studies, observational studies, and in vitro or ex vivo experiments were all considered if the full text was available in English. Studies were excluded if they involved removable appliances or clear aligners, evaluated unrelated oral hygiene products, or did not measure microbiological outcomes for the targeted pathogens. Reviews, case reports, conference abstracts, animal studies, and articles lacking adequate data were also excluded. Data were extracted and synthesized narratively, focusing on types of formulations, study outcomes, and microbial targets. Two reviewers performed the screening process.

Results

The role of probiotics was first proposed by Metchnikoff in the 20th century. According to the World Health Organization (WHO), probiotics are live microorganisms that, when consumed, can provide benefits to the host.[10] Probiotics can be given in various forms such as tablets, mouthwash, toothpaste, and drinks. Several studies have proven the beneficial effects of probiotics for humans, such as preventing caries, gingivitis, halitosis, and improving the host's immune response.[10,11] *Lactobacillus sp.* and *Bifidobacterium sp.* are colonies of good probiotic bacteria that have an effect on the oral cavity and are often used in orthodontic treatment. Probiotics interact with other bacteria in the human body. Their mechanism of action includes protecting epithelial tissue, increasing mucosal adhesion, inhibiting adhesion of pathogenic bacteria, and modulating the immune system through IgA secretion. Probiotics produce organic acids such as acetic and lactic acids, which can inhibit the effects of Gram-negative bacteria.[12,13] As a type of Lactic Acid Bacteria (LAB), *L. paracasei* has the ability to secrete extracellular polysaccharides (EPS) which can provide protection and increase the adhesion of probiotic bacteria in the oral cavity.[14] *L. paracasei* functions also can competitive exclusion of pathogens, modulation of the host immune response, and secretion of bacteriocins that inhibit periodontal pathogens.[15]

Cethylpyridinium chloride (CPC) is one of the active ingredients added to toothpaste. This ingredient has a broad-spectrum antimicrobial effect that works by destroying the cell integrity of bacteria. The Food and Drug Administration (FDA) has deemed this ingredient safe and effective for the treatment of plaque. Using CPC toothpaste can also reduce the amount of plaque in the oral cavity.[9,16] The active CPC ingredient in toothpaste is a quaternary ammonium compound. The CPC content dispersed in the oral cavity after use is maintained through surfactant chains and cationic charges absorbed in the oral cavity.[17,18] The CPC molecule has two molecular groups including hydrophilic and hydrophobic. The positively charged, or hydrophilic, group increases the binding affinity for negatively charged pathogenic bacteria. The hydrophobic group interacts with the bacterial cell surface and causes the bacteria to integrate into the cytoplasmic membrane. These two interactions can disrupt cell membrane integration, disrupt cellular metabolism, increase cytoplasmic leakage, and inhibit growth, which can lead to bacterial cell death. The presence of these two groups also reduces microbial adhesion to oral surfaces.[18]

Toothpaste containing enzymes such as amyloglucosidase and glucose oxidase has an antimicrobial effect by reducing plaque and bacterial accumulation, gingival inflammation, and can prevent the formation of white spot lesions.[8] The amyloglucosidase enzyme can inhibit bacterial growth by converting D-glucose to D-glucono-1,5-lactone. Presence of D-glucono-1,5-lactone reduces the nutritional needs of bacteria in the oral cavity. The mechanism of the glucose oxidase enzyme is to activate the salivary immune system, the lactoperoxidase (LPO) system, by producing hydrogen peroxide (H2O2), which can react with the catalase enzyme to produce oxygen, thus reducing anaerobic bacteria. Furthermore, the LPO system contains hypothiocyanate, which is activated by H2O2, which has antibacterial properties. Both enzymes produce antibacterial activity.[19,20]

Different comparative insights and mechanistic approach of toothpaste formulations containing probiotics, cethylpyridinium chloride, and enzymes can be seen in Table 1.

Table 1. Comparative Insights and Mechanistic Approach of Toothpaste Formulations Containing *Lactobacillus paracasei* Probiotics, Cetylpyridinium Chloride, and Amyloglucosidase–Glucose Oxidase Enzymes

Toothpaste Formulation	Mechanism	Primary Target	Advantages	Limitation
Lactobacillus paracasei probiotics	Disrupts bacterial membranes	Broad spectrum periodontal. pathogens	Strong antimicrobial effect and safe for daily usage	Short retention time of probiotics in the oral cavity limits their ability to adhere to mucosal surfaces and transient taste alteration
Cetylpyridinium Chloride (CPC)	Modulates microbiota	Restores balance in oral cavity and suppresses the periodontal pathogens	Improves microbial diversity and safe for long term usage	Requires viable strain storage stability, tooth staining, taste alteration, mild oral mucosal irritation
Amyloglucosidase–Glucose Oxidase Enzymes	Generates H2O2 and enhances lactoperoxidase	Biofilm inhibition	Natural ingredients and non irritating	The activity affected by storage conditions, temperature, and pH, leading to a

				reduction in efficacy over time, also caused mild irritation or altered taste perception

Discussions

Toothpaste is a product used with a toothbrush to mechanically remove debris and dental plaque.[9,21] It is available in various forms, including paste, gel, powder, and liquid. In general, toothpaste consists of two main types of ingredients: non-active and active. Non-active ingredients do not provide therapeutic effects but contribute to the physicochemical properties of the toothpaste, such as consistency, taste, fragrance, pH, and texture. Non-active ingredients in toothpaste can include water, abrasives, moisturizing agents, binders, flavors, surfactants or detergents, preservatives, and coloring agents.[21,22] Therapeutic or active ingredients in toothpaste are those that provide therapeutic effects such as preventing caries, plaque formation, tooth sensitivity, bad breath, and antimicrobial effects. Active ingredients can include fluoride, sodium bicarbonate, natural ingredients, probiotics, cethylpyridinium chloride, and enzymes.[21]

Lactobacillus strains specifically play a significant role in improving oral health. Several studies have shown that *Lactobacillus sp.* can reduce gingival inflammation.[7] *Lactobacillus paracasei* is a probiotic bacteria that has an effect on the oral cavity and can be found in toothpaste. Using toothpaste containing *L. paracasei* twice daily can reduce the number of oral pathogens in saliva.[23] Toothpaste containing *L. paracasei* has a higher antimicrobial effect than toothpaste containing *L. acidophilus* in inhibiting *S. mutans* and *C. albicans*.[7] Twetman and Keller reported that daily use of a *L. paracasei*-enriched toothpaste for 12 weeks decreased bleeding on probing and increased microbial diversity compared to placebo in orthodontic adolescents.[15] Gruner et al. observed reductions in *P. gingivalis* and *Prevotella intermedia* following probiotic use, suggesting immunomodulatory benefits. In vitro, *L. paracasei* inhibits *A. actinomycetemcomitans* via bacteriocin like compounds.[24]

Furthermore, a clinical study by Lee et al. revealed that toothpaste containing *Lactobacillus paracasei* GMNL-143 has the ability to aggregate with oral pathogens and prevent their attachment to gingival tissues. The antibacterial activity of *L. paracasei* probiotics was found to be more pronounced in acidic environments than in neutral conditions.[25] This is due to the attraction between peptides and the phosphate groups of lipopolysaccharide (LPS) molecules under acidic pH, which triggers pore formation in bacterial membranes. The resulting increase in membrane permeability leads to structural alterations and cell membrane disruption, ultimately causing bacterial lysis.[26] *L. paracasei*-containing toothpastes help stabilize microbial balance and may attenuate inflammatory responses in orthodontic patients.

Although *Lactobacillus paracasei*–based toothpaste offers promising benefits for maintaining oral microbial balance, several limitations have been reported. The viability and stability of *L. paracasei* may decline during storage or when exposed to heat and moisture, reducing its probiotic effectiveness over time. Additionally, the short retention time of probiotics in the oral cavity limits their ability to adhere to mucosal surfaces and provide prolonged antimicrobial effects. Individual variation in salivary composition, microbiota, and oral hygiene practices can also influence probiotic efficacy. Some users have reported mild taste alteration or an unpleasant aftertaste, possibly due to probiotic metabolites or interactions with toothpaste flavoring agents, which may affect long-term compliance. Furthermore, most clinical studies have short follow-up periods, leaving long-term effects on gingival health and microbial colonization uncertain.[15,27,28]

Recent randomized trials demonstrate that CPC-containing toothpastes significantly reduce gingival inflammation and microbial counts in orthodontic and general populations. Vasconcelos et al. reported that toothpaste formulated with CPC was well tolerated and exhibited no adverse effects after six weeks of use. Moreover, the CPC-containing toothpaste demonstrated antimicrobial efficacy, as indicated by a reduction in plaque accumulation among patients.[9] Giertsen et al. showed that daily use of a 0.075% CPC toothpaste in fixed-appliance patients decreased *P. gingivalis* and *F. nucleatum* levels after 6 weeks compared with fluoride-only control.[29] Otherwise, Heitz-Mayfield et al found CPC mouthrinses and dentifrices achieved plaque reductions comparable to chlorhexidine but with fewer side effects.[30] It concluded that CPC dentifrices offer modest but significant reductions in gingival index and total bacterial counts in orthodontic cohorts. CPC dentifrices offer a practical adjunct for orthodontic patients, balancing efficacy and tolerability for long-term use.

Despite its proven antimicrobial efficacy, cetylpyridinium chloride (CPC) toothpaste has several limitations. Prolonged or frequent use may cause temporary tooth staining and taste alteration, which can affect patient compliance. Some individuals may experience mild oral mucosal irritation or a transient burning sensation, particularly at higher CPC concentrations. In addition, CPC exhibits limited substantivity compared to chlorhexidine, meaning its antimicrobial effects are shorter-lived and may require regular use for sustained plaque control. The interaction of CPC with anionic compounds in toothpaste formulations, such as sodium lauryl sulfate (SLS), can reduce its bioavailability and antimicrobial activity.[31] Moreover, there is limited evidence regarding the long-term impact of CPC use on the oral microbiome balance, particularly in orthodontic patients with altered plaque ecology.[29]

Amyloglucosidase and glucose oxidase form a dual-enzyme system that converts dietary starch residues into glucose, which is then used to produce low, sustained levels of hydrogen peroxide (H_2O_2) via glucose oxidase. The generated H_2O_2 enhances the antimicrobial activity of the lactoperoxidase system naturally present in saliva, helping to reduce bacterial load without disturbing commensal species. Pistorius et al. reported that enzyme-enriched dentifrices significantly reduced plaque scores and gingival inflammation in patients with fixed orthodontic appliances after 8 weeks of use, compared with a fluoride-only control.[32] Bosch et al. found that amyloglucosidase–glucose oxidase formulations enhanced salivary lactoperoxidase activity and suppressed *A. actinomycetemcomitans* and *P. gingivalis* in vitro.[33] A pilot orthodontic study reported improved plaque control and reduced gingival bleeding without disturbing commensal *Streptococcus* species.[34] Enzyme-based toothpastes enhance host-derived antimicrobial defense and may offer biocompatible plaque control in orthodontic therapy.

Although enzyme-based toothpastes containing amyloglucosidase and glucose oxidase provide a biomimetic approach to enhancing the salivary lactoperoxidase system, several limitations should be considered. The enzymatic activity of these formulations can be affected by storage conditions, temperature, and pH, leading to a reduction in efficacy over time. Furthermore, since the system relies on salivary components such as thiocyanate and peroxidase, its antimicrobial effect may vary among individuals depending on saliva flow and composition.[35] Some users have reported mild irritation or altered taste perception, which may result from hydrogen peroxide generated during enzyme activation. The antimicrobial action is also milder and slower compared to chemical agents like chlorhexidine or CPC, requiring consistent and prolonged use for noticeable benefits. Additionally, the clinical evidence supporting the long-term effects of enzyme-based toothpaste on periodontal pathogens in orthodontic patients remains limited.[32]

Conclusion

L. paracasei probiotics, CPC, and amyloglucosidase and glucose oxidase enzymes based toothpastes represent a new generation of targeted formulations that offer effective adjuncts for controlling periodontal pathogens in patients with fixed orthodontic appliances. Each approach works through a different mechanism, allowing targeted control of various aspects of biofilm accumulation and pathogenicity. Combining these strategies could optimize microbial balance while minimizing adverse effects. While current evidence is promising, further orthodontic specific trials are needed to establish standardized protocols and confirm long term benefits in practical application.

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Efficacy of Probiotics, Cetylpyridinium Chloride, and Enzymes-Based Toothpaste Formulations Against Periodontal Pathogens in Fixed Orthodontic Patients: A Review

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Abstract

Introduction: Fixed orthodontic appliances complicate oral hygiene and promote biofilm accumulation, increasing the risk of periodontal disease. Toothpaste formulations with antimicrobial properties such as *Lactobacillus paracasei* probiotics, cetylpyridinium chloride (CPC), and amyloglucosidase–glucose oxidase enzymes are commonly used to manage oral biofilm and control pathogenic bacteria. However, the extent of their effectiveness against periodontal pathogens in orthodontic patients remains unclear. **Purpose:** This review aims to map and summarize the current evidence on the efficacy of various toothpaste formulations in reducing periodontal pathogens among patients undergoing fixed orthodontic treatment. **Methods:** A comprehensive search of electronic databases (e.g., PubMed, Scopus, Google Scholar) was conducted to identify relevant studies published from 2015 to 2025. Data were extracted and synthesized narratively, focusing on types of formulations, study outcomes, and microbial targets. **Results:** Probiotic, enzymatic, and CPC-containing toothpastes showed variable but complementary effects against key pathogens, including *Porphyromonas gingivalis* and *Aggregatibacter actinomycetemcomitans*. Probiotics modulate microbial balance, enzymes disrupt biofilm structure, and CPC provides chemical antimicrobial action. Combining these mechanisms appears promising for enhanced biofilm control. **Conclusion:** Toothpastes containing *L. paracasei* probiotics, CPC, and amyloglucosidase–glucose oxidase enzymes offer complementary ways to control periodontal pathogens in orthodontic patients. Combining these mechanisms may enhance biofilm control and microbial balance, though further research is needed to confirm their long-term benefits.

Keywords: antimicrobial effect; fixed orthodontic appliances; oral pathogens; toothpaste formulations

Introduction

Fixed orthodontic appliances are widely used to correct malocclusions and improve dental aesthetics.[1] However, these appliances create numerous retention sites that hinder effective plaque removal, making it difficult for patients to maintain optimal oral hygiene. As a result, orthodontic treatment often leads to increased biofilm accumulation and inflammation of the surrounding periodontal tissues, particularly the gingiva.[2–4]

Wearing fixed orthodontic appliances can lead to more biofilm accumulation, which shifts the oral microbiota toward more harmful bacteria. Periodontal pathogens like *Porphyromonas gingivalis* and *Aggregatibacter actinomycetemcomitans* may increase,

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raising the risk of gingivitis and even periodontitis if not controlled. Therefore, managing bacterial biofilm is crucial to maintain good periodontal health during orthodontic treatment.[5,6]

Toothpaste is one of the most accessible and commonly used oral hygiene tools. Various formulations, including those containing probiotics, enzymes, and cetylpyridinium chloride have been developed to enhance antimicrobial activity and improve periodontal outcomes.[7–9] Despite their widespread use, evidence on their ability to reduce periodontal pathogens in orthodontic patients remains limited, as existing studies are scattered, heterogeneous, and often restricted to in vitro experiments or clinical trials. This gap underscores the need for a comprehensive scoping review to evaluate and map existing research on this topic. This review explores the range of toothpaste formulations studied for their antimicrobial effects on periodontal pathogens in patients with fixed orthodontic appliances.

Methods

2 A comprehensive search of electronic databases such as PubMed, Scopus, Google Scholar was conducted to identify relevant studies published from 2015 to 2025 by using the Boolean Search such as ["Porphyromonas gingivalis" OR "P. gingivalis" OR "Aggregatibacter actinomycetemcomitans" OR "A. actinomycetemcomitans" OR "periodontal pathogens" OR "periodontopathogenic bacteria"] AND ["Probiotics toothpaste" OR “probiotic toothpaste” OR "Lactobacillus paracasei toothpaste" OR "L. paracasei toothpaste"] AND ["Cetylpyridinium chloride" OR “cetylpyridinium” OR “CPC”] AND ["Enzyme-based toothpaste" OR "enzymatic toothpaste" OR "amyloglucosidase" OR "glucose oxidase" OR "enzymes-based formulations"] AND ["fixed orthodontic appliances" OR "fixed orthodontic" OR "fixed appliance"] AND ["bacterial reduction" OR "biofilm reduction" OR "antimicrobial effect" OR "antibacterial effect" OR "dental plaque"].

This review included studies involving patients with fixed orthodontic appliances or laboratory models that mimic orthodontic biofilm conditions. Eligible studies examined probiotics such as *Lactobacillus paracasei*, cetylpyridinium chloride (CPC), or enzyme-based toothpastes containing amyloglucosidase and glucose oxidase, and reported outcomes related to *Porphyromonas gingivalis* or *Aggregatibacter actinomycetemcomitans*. Randomized trials, clinical studies, observational studies, and in vitro or ex vivo experiments were all considered if the full text was available in English. Studies were excluded if they involved removable appliances or clear aligners, evaluated unrelated oral hygiene products, or did not measure microbiological outcomes for the targeted pathogens. Reviews, case reports, conference abstracts, animal studies, and articles lacking adequate data were also excluded. Data were extracted and synthesized narratively, focusing on types of formulations, study outcomes, and microbial targets. Two reviewers performed the screening process.

Results

The role of probiotics was first proposed by Metchnikoff in the 20th century. According to the World Health Organization (WHO), probiotics are live microorganisms that, when consumed, can provide benefits to the host.[10] Probiotics can be given in various forms such as tablets, mouthwash, toothpaste, and drinks. Several studies have proven the beneficial effects of probiotics for humans, such as preventing caries, gingivitis, halitosis, and improving the host's immune response.[10,11] *Lactobacillus sp.* and *Bifidobacterium sp.* are colonies of good probiotic bacteria that have an effect on the oral cavity and are often used in orthodontic treatment. Probiotics interact with other bacteria in the human body. Their mechanism of action includes protecting epithelial tissue, increasing mucosal adhesion, inhibiting adhesion of pathogenic bacteria, and modulating the immune system through IgA secretion. Probiotics produce organic acids such as acetic and lactic acids, which can inhibit the effects of Gram-negative bacteria.[12,13] As a type of Lactic Acid Bacteria (LAB), *L. paracasei* has the ability to secrete extracellular polysaccharides (EPS) which can provide protection and increase the adhesion of probiotic bacteria in the oral cavity.[14] *L. paracasei* functions also can competitive exclusion of pathogens, modulation of the host immune response, and secretion of bacteriocins that inhibit periodontal pathogens.[15]

Cethylpyridinium chloride (CPC) is one of the active ingredients added to toothpaste. This ingredient has a broad-spectrum antimicrobial effect that works by destroying the cell integrity of bacteria. The Food and Drug Administration (FDA) has deemed this ingredient safe and effective for the treatment of plaque. Using CPC toothpaste can also reduce the amount of plaque in the oral cavity.[9,16] The active CPC ingredient in toothpaste is a quaternary ammonium compound. The CPC content dispersed in the oral cavity after use is maintained through surfactant chains and cationic charges absorbed in the oral cavity.[17,18] The CPC molecule has two molecular groups including hydrophilic and hydrophobic. The positively charged, or hydrophilic, group increases the binding affinity for negatively charged pathogenic bacteria. The hydrophobic group interacts with the bacterial cell surface and causes the bacteria to

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integrate into the cytoplasmic membrane. These two interactions can disrupt cell membrane integration, disrupt cellular metabolism, increase cytoplasmic leakage, and inhibit growth, which can lead to bacterial cell death. The presence of these two groups also reduces microbial adhesion to oral surfaces.[18]

Toothpaste containing enzymes such as amyloglucosidase and glucose oxidase has an antimicrobial effect by reducing plaque and bacterial accumulation, gingival inflammation, and can prevent the formation of white spot lesions.[8] The amyloglucosidase enzyme can inhibit bacterial growth by converting D-glucose to D-glucono-1,5-lactone. Presence of D-glucono-1,5-lactone reduces the nutritional needs of bacteria in the oral cavity. The mechanism of the glucose oxidase enzyme is to activate the salivary immune system, the lactoperoxidase (LPO) system, by producing hydrogen peroxide (H2O2), which can react with the catalase enzyme to produce oxygen, thus reducing anaerobic bacteria. Furthermore, the LPO system contains hypothiocyanate, which is activated by H2O2, which has antibacterial properties. Both enzymes produce antibacterial activity.[19,20]

Different comparative insights and mechanistic approach of toothpaste formulations containing probiotics, cethylpyridinium chloride, and enzymes can be seen in Table 1.

Table 1. Comparative Insights and Mechanistic Approach of Toothpaste Formulations Containing *Lactobacillus paracasei* Probiotics, Cetylpyridinium Chloride, and Amyloglucosidase–Glucose Oxidase Enzymes

Toothpaste Formulation	Mechanism	Primary Target	Advantages	Limitation
Lactobacillus paracasei probiotics	Disrupts bacterial membranes	Broad spectrum periodontal. pathogens	Strong antimicrobial effect and safe for daily usage	Short retention time of probiotics in the oral cavity limits their ability to adhere to mucosal surfaces and transient taste alteration
Cetylpyridinium Chloride (CPC)	Modulates microbiota	Restores balance in oral cavity and suppresses the periodontal pathogens	Improves microbial diversity and safe for long term usage	Requires viable strain storage stability, tooth staining, taste alteration, mild oral mucosal irritation
Amyloglucosidase–Glucose Oxidase Enzymes	Generates H2O2 and enhances lactoperoxidase	Biofilm inhibition	Natural ingredients and non irritating	The activity affected by storage conditions, temperature, and pH, leading to a

	reduction in efficacy over time, also caused mild irritation or altered taste perception

Discussions

Toothpaste is a product used with a toothbrush to mechanically remove debris and dental plaque.[9,21] It is available in various forms, including paste, gel, powder, and liquid. In general, toothpaste consists of two main types of ingredients: non-active and active. Non-active ingredients do not provide therapeutic effects but contribute to the physicochemical properties of the toothpaste, such as consistency, taste, fragrance, pH, and texture. Non-active ingredients in toothpaste can include water, abrasives, moisturizing agents, binders, flavors, surfactants or detergents, preservatives, and coloring agents.[21,22] Therapeutic or active ingredients in toothpaste are those that provide therapeutic effects such as preventing caries, plaque formation, tooth sensitivity, bad breath, and antimicrobial effects. Active ingredients can include fluoride, sodium bicarbonate, natural ingredients, probiotics, cethylpyridinium chloride, and enzymes.[21]

Lactobacillus strains specifically play a significant role in improving oral health. Several studies have shown that *Lactobacillus sp.* can reduce gingival inflammation.[7] *Lactobacillus paracasei* is a probiotic bacteria that has an effect on the oral cavity and can be found in toothpaste. Using toothpaste containing *L. paracasei* twice daily can reduce the number of oral pathogens in saliva.[23] Toothpaste containing *L. paracasei* has a higher antimicrobial effect than toothpaste containing *L. acidophilus* in inhibiting *S. mutans* and *C. albicans*.[7] Twetman and Keller reported that daily use of a *L. paracasei*-enriched toothpaste for 12 weeks decreased bleeding on probing and increased microbial diversity compared to placebo in orthodontic adolescents.[15] Gruner et al. observed reductions in *P. gingivalis* and *Prevotella intermedia* following probiotic use, suggesting immunomodulatory benefits. In vitro, *L. paracasei* inhibits *A. actinomycetemcomitans* via bacteriocin like compounds.[24]

Furthermore, a clinical study by Lee et al. revealed that toothpaste containing *Lactobacillus paracasei* GMNL-143 has the ability to aggregate with oral pathogens and prevent their attachment to gingival tissues. The antibacterial activity of *L. paracasei* probiotics was found to be more pronounced in acidic environments than in neutral

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conditions.[25] This is due to the attraction between peptides and the phosphate groups of lipopolysaccharide (LPS) molecules under acidic pH, which triggers pore formation in bacterial membranes. The resulting increase in membrane permeability leads to structural alterations and cell membrane disruption, ultimately causing bacterial lysis.[26] *L. paracasei*-containing toothpastes help stabilize microbial balance and may attenuate inflammatory responses in orthodontic patients.

Although *Lactobacillus paracasei*–based toothpaste offers promising benefits for maintaining oral microbial balance, several limitations have been reported. The viability and stability of *L. paracasei* may decline during storage or when exposed to heat and moisture, reducing its probiotic effectiveness over time. Additionally, the short retention time of probiotics in the oral cavity limits their ability to adhere to mucosal surfaces and provide prolonged antimicrobial effects. Individual variation in salivary composition, microbiota, and oral hygiene practices can also influence probiotic efficacy. Some users have reported mild taste alteration or an unpleasant aftertaste, possibly due to probiotic metabolites or interactions with toothpaste flavoring agents, which may affect long-term compliance. Furthermore, most clinical studies have short follow-up periods, leaving long-term effects on gingival health and microbial colonization uncertain.[15,27,28]

Recent randomized trials demonstrate that CPC-containing toothpastes significantly reduce gingival inflammation and microbial counts in orthodontic and general populations. Vasconcelos et al. reported that toothpaste formulated with CPC was well tolerated and exhibited no adverse effects after six weeks of use. Moreover, the CPC-containing toothpaste demonstrated antimicrobial efficacy, as indicated by a reduction in plaque accumulation among patients.[9] Giertsen et al. showed that daily use of a 0.075% CPC toothpaste in fixed-appliance patients decreased *P. gingivalis* and *F. nucleatum* levels after 6 weeks compared with fluoride-only control.[29] Otherwise, Heitz-Mayfield et al found CPC mouthrinses and dentifrices achieved plaque reductions comparable to chlorhexidine but with fewer side effects.[30] It concluded that CPC dentifrices offer modest but significant reductions in gingival index and total bacterial counts in orthodontic cohorts. CPC dentifrices offer a practical adjunct for orthodontic patients, balancing efficacy and tolerability for long-term use.

Despite its proven antimicrobial efficacy, cetylpyridinium chloride (CPC) toothpaste has several limitations. Prolonged or frequent use may cause temporary tooth staining and taste alteration, which can affect patient compliance. Some individuals may experience mild oral mucosal irritation or a transient burning sensation, particularly at higher CPC concentrations. In addition, CPC exhibits limited substantivity compared to chlorhexidine, meaning its antimicrobial effects are shorter-lived and may require regular use for sustained plaque control. The interaction of CPC with anionic compounds in toothpaste formulations, such as sodium lauryl sulfate (SLS), can reduce its bioavailability and antimicrobial activity.[31] Moreover, there is limited evidence regarding the long-term impact of CPC use on the oral microbiome balance, particularly in orthodontic patients with altered plaque ecology.[29]

Amyloglucosidase and glucose oxidase form a dual-enzyme system that converts dietary starch residues into glucose, which is then used to produce low, sustained levels of hydrogen peroxide (H_2O_2) via glucose oxidase. The generated H_2O_2 enhances the antimicrobial activity of the lactoperoxidase system naturally present in saliva, helping to reduce bacterial load without disturbing commensal species. Pistorius et al. reported that enzyme-enriched dentifrices significantly reduced plaque scores and gingival inflammation in patients with fixed orthodontic appliances after 8 weeks of use, compared with a fluoride-only control.[32] Bosch et al. found that amyloglucosidase–glucose oxidase formulations enhanced salivary lactoperoxidase activity and suppressed *A. actinomycetemcomitans* and *P. gingivalis* in vitro.[33] A pilot orthodontic study reported improved plaque control and reduced gingival bleeding without disturbing commensal *Streptococcus* species.[34] Enzyme-based toothpastes enhance host-derived antimicrobial defense and may offer biocompatible plaque control in orthodontic therapy.

Although enzyme-based toothpastes containing amyloglucosidase and glucose oxidase provide a biomimetic approach to enhancing the salivary lactoperoxidase system, several limitations should be considered. The enzymatic activity of these formulations can be affected by storage conditions, temperature, and pH, leading to a reduction in efficacy over time. Furthermore, since the system relies on salivary components such as thiocyanate and peroxidase, its antimicrobial effect may vary among individuals depending on saliva flow and composition.[35] Some users have reported mild irritation or altered taste perception, which may result from hydrogen peroxide generated during enzyme activation. The antimicrobial action is also milder and slower compared to chemical agents like chlorhexidine or CPC, requiring consistent and prolonged use for noticeable benefits. Additionally, the clinical evidence supporting the long-term effects of enzyme-based toothpaste on periodontal pathogens in orthodontic patients remains limited.[32]

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Conclusion

L. paracasei probiotics, CPC, and amyloglucosidase and glucose oxidase enzymes based toothpastes represent a new generation of targeted formulations that offer effective adjuncts for controlling periodontal pathogens in patients with fixed orthodontic appliances. Each approach works through a different mechanism, allowing targeted control of various aspects of biofilm accumulation and pathogenicity. Combining these strategies could optimize microbial balance while minimizing adverse effects. While current evidence is promising, further orthodontic specific trials are needed to establish standardized protocols and confirm long term benefits in practical application.

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BUKTI KORESPONDENSI
ARTIKEL JURNAL NASIONAL

Judul Artikel : Efficacy of Probiotics, Cetylpyridinium Chloride, and Enzymes-Based Toothpaste Formulations Against Periodontal Pathogens in Fixed Orthodontic Patients: A Review

Jurnal : Journal of Orthodontic Science
URL Homepage:
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Penulis : Joko Kusnoto

No	Perihal	Tanggal
1	Bukti konfirmasi submit artikel dan artikel yang disubmit	24 Oktober 2025
2	Bukti konfirmasi review	28 Oktober 2025
3	Bukti konfirmasi submit revisi	31 Oktober 2025
4	Bukti konfirmasi artikel published	25 Desember 2025

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Joko Kusnoto <joko.k@trisakti.ac.id>

Submit manuskrip Sriwijaya Journal of Dentistry

16 messages

Joko Kusnoto <joko.k@trisakti.ac.id> Fri, Oct 24, 2025 at 10:56 AM
To: sitirusdiana@fk.unsri.ac.id
Cc: marthamozartha@fk.unsri.ac.id

Yth Drg, Siti Rusdiana Puspa Dewi
Editor in Chief Sriwijaya Journal of Dentistry

Dengan hormat, bersama ini kami ingin menanyakan apakah manuskrip kami berjudul: Probiotics, Cetylpyridinium Chloride, and Enzymes Toothpaste Formulations Against Periodontal Pathogens in Fixed Orthodontic Patients: A Review yang telah kami submit secara online melalui portal Sriwijaya Journal of Dentistry pada tanggal 14 Oktober 2025 telah diterima oleh redaksi? Mohon maaf kami menanyakan hal ini karena sampai sekarang kami belum mendapatkan notifikasi apapun via e-mail.

Mohon informasinya dan besar harapan kami agar manuskrip kami dapat dipublikasikan pada Sriwijaya Journal of Dentistry edisi mendatang Desember 2025. Atas perhatian dan kerjasamanya kami ucapkan terima kasih dan kami tunggu kabar baiknya.

Hormat kami,
Joko Kusnoto

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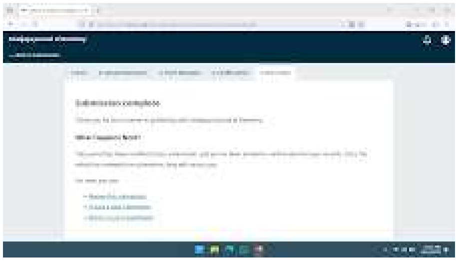
2 attachments



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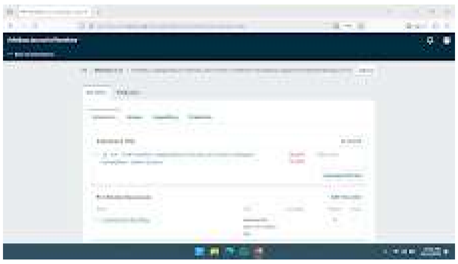

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martha mozartha <marthamozartha@fk.unsri.ac.id> Tue, Oct 28, 2025 at 10:50 AM
To: Joko Kusnoto <joko.k@trisakti.ac.id>

Yth Penulis,
Terima kasih telah memasukkan manuskrip Anda di jurnal kami, Sriwijaya Journal of Dentistry. Editor kami telah memeriksa manuskrip Anda dan perlu perbaikan, silakan cek dashboard dengan login ke website kami dan mohon segera dikirim kembali manuskrip yang telah diperbaiki. Terima kasih.

Chief in editor

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> [Quoted text hidden]

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Joko Kusnoto <joko.k@trisakti.ac.id> Fri, Oct 31, 2025 at 12:49 AM
To: martha mozartha <marthamozartha@fk.unsri.ac.id>

Yth Drg, Siti Rusdiana Puspa Dewi, M.Kes
Editor in Chief Sriwijaya Journal of Dentistry

Dengan hormat, bersama ini kami ingin menyampaikan bahwa kami telah melakukan revisi manuskrip kami berjudul: "Probiotics, Cetylpyridinium Chloride, and Enzymes Toothpaste Formulations Against Periodontal Pathogens in Fixed Orthodontic Patients: A Review" (terlampir) sesuai arahan editor dan telah kami submit secara online melalui portal Sriwijaya Journal of Dentistry.

Kami harap manuskrip kami telah memenuhi standar Sriwijaya Journal of Dentistry sehingga dapat dipublikasikan pada Sriwijaya Journal of Dentistry edisi mendatang Desember 2025. Atas perhatian dan kerjasamanya kami ucapkan terima kasih dan kami tunggu kabar baiknya.

Hormat kami,
Joko Kusnoto
[Quoted text hidden]

Draft Probiotics, Cetylpyridinium Chloride, and Enzymes Toothpaste Formulations - Submit SJD 02.docx
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Joko Kusnoto <joko.k@trisakti.ac.id> Tue, Nov 18, 2025 at 7:16 AM
To: martha mozartha <marthamozartha@fk.unsri.ac.id>
Cc: sitirusdiana@fk.unsri.ac.id

Yth Drg, Siti Rusdiana Puspa Dewi, M.Kes
Editor in Chief Sriwijaya Journal of Dentistry

Dengan hormat,

Bersama ini kami ingin menanyakan apakah revisi manuskrip kami berjudul: Probiotics, Cetylpyridinium Chloride, and Enzymes Toothpaste Formulations Against Periodontal Pathogens in Fixed Orthodontic Patients: A Review yang telah kami submit secara online melalui portal Sriwijaya Journal of Dentistry dan juga melalui e-mail pada tanggal 31 Oktober 2025 telah diterima oleh redaksi dan telah sesuai dengan arahan editor Sriwijaya Journal of Dentistry?

Mohon informasinya dan besar harapan kami agar manuskrip kami dapat dipublikasikan pada Sriwijaya Journal of Dentistry edisi Desember 2025 mendatang. Atas perhatian dan kerjasamanya kami ucapkan terima kasih dan kami tunggu kabar baiknya.

Hormat kami,
Joko Kusnoto

On Tue, Oct 28, 2025 at 10:46 AM martha mozartha <marthamozartha@fk.unsri.ac.id> wrote:
[Quoted text hidden]

Draft Probiotics, Cetylpyridinium Chloride, and Enzymes Toothpaste Formulations - Submit SJD 02.docx
124K

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martha mozartha <marthamozartha@fk.unsri.ac.id> Tue, Nov 18, 2025 at 11:49 PM
To: Joko Kusnoto <joko.k@trisakti.ac.id>

Yth Penulis,
Saat ini artikel berjudul "Probiotics, Cetylpyridinium Chloride, and Enzymes Toothpaste Formulations Against Periodontal Pathogens in Fixed Orthodontic Patients: A Review" sedang dalam proses review. Mohon menunggu hingga proses review selesai.
Demikian informasi yang dapat kami sampaikan, terima kasih atas kerjasamanya.

Salam hormat,
Editor
[Quoted text hidden]

---

martha mozartha <marthamozartha@fk.unsri.ac.id> Tue, Dec 9, 2025 at 3:35 PM
To: Joko Kusnoto <joko.k@trisakti.ac.id>

Yth Penulis,
Berikut kami lampirkan hasil telaah reviewer 1 dan 2 (*terlampir)* .
Rekomendasi dari reviewer: Revision required.
Mohon perbaikan artikel saudara dikirim kembali paling lambat tgl 16 Desember 2025. Terima kasih atas kerjasamanya.

On Fri, 24 Oct 2025 at 10:56, Joko Kusnoto <joko.k@trisakti.ac.id> wrote:
> [Quoted text hidden]
> [Quoted text hidden]

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Joko Kusnoto <joko.k@trisakti.ac.id> Sun, Dec 14, 2025 at 10:02 AM
To: martha mozartha <marthamozartha@fk.unsri.ac.id>
Cc: sitirusdiana@fk.unsri.ac.id

Yth. Editor Sriwijaya Journal of Dentistry,

Sesuai arahan Editor Sriwijaya Journal of Dentistry maka kami telah melakukan revisi manuskrip kami sesuai hasil telaah reviewer 1 dan 2. Kami harap revisi yang telah kami lakukan membuat manuskrip kami sesuai dengan standar Sriwijaya Journal of Dentistry dan layak diterbitkan pada edisi berikut.

Atas perhatian dan kerjasamanya kami ucapkan terima kasih dan kami tunggu kabar baiknya.

Hormat kami,
Michelle, Joko Kusnoto, dan Harryanto Wijaya

[Quoted text hidden]

Draft Probiotics, Cetylpyridinium Chloride, and Enzymes Toothpaste Formulations - Submit SJD 03.docx
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martha mozartha <marthamozartha@fk.unsri.ac.id> Wed, Dec 17, 2025 at 1:24 PM
To: Joko Kusnoto <joko.k@trisakti.ac.id>

Yth Penulis,
Mohon file naskah yang telah direvisi dapat di-upload di website SJD.
Terima kasih atas kerja samanya.

Regards
[Quoted text hidden]

Joko Kusnoto <joko.k@trisakti.ac.id> Wed, Dec 17, 2025 at 9:33 PM
To: martha mozartha <marthamozartha@fk.unsri.ac.id>
Cc: sitirusdiana@fk.unsri.ac.id

Yth. Editor Sriwijaya Journal of Dentistry,

Sesuai arahan Editor Sriwijaya Journal of Dentistry maka kami bermaksud mengupload file manuskrip kami yang telah direvisi ke website Sriwijaya Journal of Dentistry tapi mengalami kendala dengan pesan: Internal Server Error (screenshot terlampir).

Mohon arahannya lebih lanjut dan atas perhatian serta kerjasamanya kami ucapkan terima kasih.

Hormat kami,
Joko Kusnoto

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SJD Internal Server Error.jpg
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Joko Kusnoto <joko.k@trisakti.ac.id> Thu, Dec 18, 2025 at 9:34 AM
To: martha mozartha <marthamozartha@fk.unsri.ac.id>

Yth. Editor Sriwijaya Journal of Dentistry,

Sesuai arahan Editor Sriwijaya Journal of Dentistry maka pagi ini setelah website SJD pulih kami telah mengupload file revisi manuskrip kami.

Atas perhatian serta kerjasamanya kami ucapkan terima kasih, besar harapan kami agar manuskrip kami dapat dipublikasikan pada Sriwijaya Journal of Dentistry edisi Desember 2025 mendatang.

Hormat kami,
Joko Kusnoto

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martha mozartha <marthamozartha@fk.unsri.ac.id> Thu, Dec 18, 2025 at 10:56 AM
To: Joko Kusnoto <joko.k@trisakti.ac.id>

Yth Penulis,
Mohon maaf atas kendala yang dihadapi. Website SJD beberapa hari ke belakang ini sempat error, namun pagi ini sudah dapat diakses. Mohon untuk dapat dicoba kembali, mohon maaf atas ketidaknyamanan ini.
Terima kasih atas kerjasamanya.

Regards,
[Quoted text hidden]

Joko Kusnoto <joko.k@trisakti.ac.id> Thu, Dec 18, 2025 at 11:29 AM
To: martha mozartha <marthamozartha@fk.unsri.ac.id>

Yth. Editor Sriwijaya Journal of Dentistry,

Sesuai arahan Editor Sriwijaya Journal of Dentistry maka pagi ini setelah website SJD pulih pada Kamis 18 Desember pk 09.29 WIB kami telah mengupload file revisi manuskrip kami.

Atas perhatian serta kerjasamanya kami ucapkan terima kasih, besar harapan kami agar manuskrip kami dapat dipublikasikan pada Sriwijaya Journal of Dentistry edisi Desember 2025 mendatang.

Hormat kami,
Joko Kusnoto

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martha mozartha <marthamozartha@fk.unsri.ac.id> Sun, Dec 21, 2025 at 3:11 PM
To: Joko Kusnoto <joko.k@trisakti.ac.id>

Yth Penulis,
Mohon dapat mengunggah file naskah revisi di kolom Revisions seperti gambar di bawah ini:
Terima kasih atas bantuan dan kerja samanya

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Joko Kusnoto <joko.k@trisakti.ac.id> Sun, Dec 21, 2025 at 4:19 PM
To: martha mozartha <marthamozartha@fk.unsri.ac.id>
Cc: sitirusdiana@fk.unsri.ac.id

Yth. Editor Sriwijaya Journal of Dentistry,

Sesuai arahan Editor Sriwijaya Journal of Dentistry maka kami bermaksud mengupload file manuskrip kami yang telah direvisi ke kolom Revisi pada website Sriwijaya Journal of Dentistry tapi mengalami kendala dengan pesan: You are not allowed to add and edit these files (screenshot terlampir).

Mohon arahannya lebih lanjut dan atas perhatian serta kerjasamanya kami ucapkan terima kasih.

Hormat kami,
Joko Kusnoto

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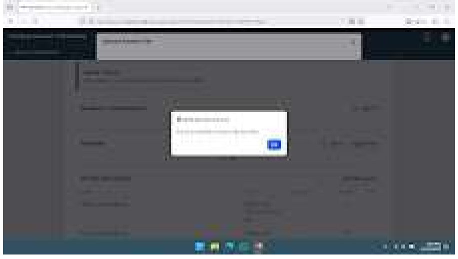

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Joko Kusnoto <joko.k@trisakti.ac.id> Mon, Dec 22, 2025 at 12:28 AM
To: martha mozartha <marthamozartha@fk.unsri.ac.id>
Cc: sitirusdiana@fk.unsri.ac.id

Yth. Editor Sriwijaya Journal of Dentistry,

Sesuai arahan Editor Sriwijaya Journal of Dentistry maka pada pagi ini Senin 22 Desember pk 00.16 WIB kami telah berhasil mengupload file revisi manuskrip kami pada kolom Revisions.

Atas perhatian serta kerjasamanya kami ucapkan terima kasih, besar harapan kami agar manuskrip kami dapat dipublikasikan pada Sriwijaya Journal of Dentistry edisi Desember 2025 mendatang.

Hormat kami,
Joko Kusnoto

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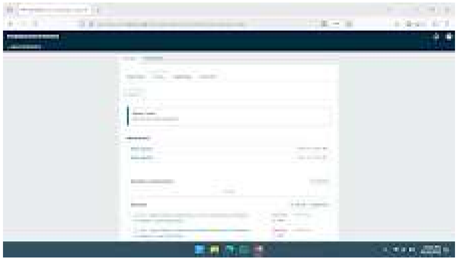

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martha mozartha <marthamozartha@fk.unsri.ac.id> Thu, Dec 25, 2025 at 12:58 AM
To: Joko Kusnoto <joko.k@trisakti.ac.id>

Dear author,
Selamat atas diterbitkannya naskah Anda di Sriwijaya Journal of Dentistry Vol. 6 No.2 2025. Silakan kunjungi web kami di https://sjd-fk.ejournal.unsri.ac.id/index.php/sjd/article/view/86.
Terima kasih atas kontribusi Anda, silahkan kirimkan kembali naskah ilmiah Anda untuk terbitan-terbitan selanjutnya.

Best Regards,
Editor
[Quoted text hidden]