Identification of Bacteria in Dental Plaque

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Abstract

Oral health has an important role in overall body health. This study aims to evaluate the antibacterial efficacy of toothpaste with cinnamon ethanol extract against Streptococcus bacteria in vivo, analyze changes in the number of bacterial colonies before and after using toothpaste, and identify the type of Streptococcus bacteria in dental plaque. This pure experimental research was conducted at the Pharmacy Laboratory of the Poltekkes Kemenkes Medan and the Microbiology Laboratory of the Faculty of Medicine, University of North Sumatra. Samples included cinnamon extract, toothpaste formulation with 12.5% extract concentration, and dental plaque from 40 respondents. Data analysis used descriptive univariate test with SPSS 22.0 application. Results showed Gram-positive bacterial infections, especially Rothia dentocariosa (24%), dominated dental plaque, followed by Streptococcus mitis (16.4%). The use of cinnamon-active toothpaste was able to significantly reduce the number of bacterial colonies, especially at high concentrations. In addition, active compounds in cinnamon extracts such as cinnamaldehyde and eugenol showed the ability to damage bacterial cell walls and inhibit biofilm formation. Gargling was also shown to be effective in reducing bacterial colonies by clearing plaque and food debris. The conclusion of this study suggests that Gram-positive bacterial infections in dental plaque require special attention in management with appropriate antibiotics, while gargling and the use of cinnamon-based antimicrobial agents can be an effective preventive strategy. These findings support the development of natural ingredient-based oral health products to improve infection prevention and the quality of oral health care.

Keywords: Oral Health, Streptococcus, Cinnamon, Antibacterial, Dental Plaque, Gargling

INTRODUCTION

Oral health plays an important role in the general health and well-being of the body (Hescot, 2017). There are many diseases that start from the teeth and mouth because the mouth is the entrance of all kinds of foreign objects into the body (Akanchi et al., 2022). Oral health greatly affects a person's quality of life because the teeth and mouth are important for speech and masticatory functions (Waty et al., 2023).

The age of children with dental caries is currently influenced by community behavior factors. Food residues attached to children's teeth that are not cleaned can cause tooth decay which results in porous teeth, cavities, etc. The impact caused by dental caries that occurs in children will hamper the process of growth and development in children. The impact caused by dental caries that occurs in children will hamper the process of growth and development in children. The 2018 Basic Health Research recorded the proportion of oral problems at 57.6%

and those who received services from dental medical personnel at 10.2%. The proportion of correct tooth brushing behavior was 2.8% (Kemenkes RI, 2018). This figure is still far from the target of the World Health Organization (WHO) which wants 50% of children aged 5-6 years to be free from dental caries (Santamaria et al., 2019). The occurrence of caries and dental supporting tissue abnormalities begins with the formation of dental plaque (Loban et al., 2021). Plaque in the form of a thin layer attached to the surface of the teeth and sometimes also found on the gums and tongue is caused by food debris which is broken down by bacteria in the oral cavity (Jakubovics et al., 2021).

Prospective studies have also reported high incidence rates in a Danish study, where all dental injuries occurring from birth to 14 years of age were carefully registered, showing that 30% of children had injuries to primary teeth and 22% to permanent teeth (Petti et al., 2018). Overall, every child had experienced TDI by the age of 14 (Born et al., 2019). In Australia, TDIs were more common in children under 5 years of age (56.1%) with the majority of injuries sustained by males (63.8%). Injury characteristics included weekend occurrences (35.6%), the most common etiology was falls (64.4%) and many incidents occurred at home (48.5%). Overall, 654 teeth were injured with the majority affecting primary teeth (58.4%) and maxillary central incisors (69.9%). The most common injury was lateral luxation (27.5%) (Ng et al., 2020).

The proliferation of Streptococcus bacteria is affected by multiple interconnected factors. Temperature is a primary factor (Mishra et al., 2018). Streptococcus typically proliferates optimally at human body temperature, approximately 37°C (Jans & Boleij, 2018). Extreme environmental temperatures may impede the growth of these microorganisms. Nevertheless, several species of these bacteria can endure varying temperature circumstances, albeit their development rate is not as rapid as at the optimal temperature (Cebrián et al., 2017).

The pH level of the environment also influences the proliferation of Streptococcus (Do et al., 2019). These bacteria favor an environment with a neutral to slightly acidic pH, approximately between 6.5 and 7.5 (Wang et al., 2020). Excessively low or high pH levels may compromise bacterial enzyme activity, hence hindering growth and reproduction (Machado et al., 2017).

Bacteria in the mouth multiply from food debris that is not cleaned properly, especially those containing sugar and starch. If you do not brush your teeth regularly and clean between your teeth with dental floss, plaque will form. This plaque is a sticky layer containing bacteria, and over time it will trigger the formation of acids that can damage the tooth enamel layer (Septiani et al., 2021). Diet also plays an important role in bacterial growth. Foods and drinks

rich in sugar, such as candies, cakes and soft drinks, can increase acid production by bacteria in the mouth (S S et al., 2022). Irregular dental care can worsen tooth decay. Regular check-ups with the dentist are essential to detect plaque build-up, cavities, or other oral health issues before they become more severe (Freiberg et al., 2020).

Nuriyah et al., (2022) research found several factors that can cause dental caries, including consuming cariogenic foods and a lack of knowledge in elementary school children to consume foods that can help improve oral health. According to Saputri et al., (2022), knowledge about brushing teeth includes how to brush teeth correctly, namely brushing not only aims to clean dirty parts that are easily visible or only aims to clean teeth, but attention is also aimed at cleaning plaque or gums. This study seeks to evaluate the antibacterial efficacy of toothpaste infused with ethanol extract of cinnamon bark against Streptococcus bacteria in vivo, quantify alterations in bacterial colony counts pre- and post-application of the toothpaste formulation, and classify the specific types of Streptococcus bacteria present in the dental plaque of participants.

METHODS

This type of research is a pure experiment, which is to make ethanol extract of cinnamon bark and formulate it into toothpaste then test its antibacterial activity. the independent variable in this study is antibacterial activity and the dependent variable in this study is the cinnamon bark ethanol extract toothpaste formula. This research was conducted at the Pharmacy Laboratory of the Poltekkes Kemenkes Medan and the Microbiology Laboratory of the Faculty of Medicine, University of North Sumatra. The samples in this study were cinnamon bark extract and dental plaque taken from 40 respondents of Pharmacy students of Poltekkes Kemenkes Medan.

The research materials at this time were cinnamon bark extract (Cinnamomum burmanni), CaCO3 (Calcium carbonate), glycerin, sodium cyclamate, menthol, sodium lauryl sulfate, NA CMC, distilled water, ethanol, TYCSB (Tryptone Yeast Cystein) Agar Base medium, MHA (Muller Hilton Agar) medium, physiological NaCl 0.9%, cinnamon bark extract toothpaste formula with 12.5% extract concentration, toothpaste without cinnamon bark extract, toothpaste on the market, dental plaque samples taken from students. The data obtained in this study will be presented in the form of figures and distribution tables. Statistical data analysis using SPSS version 22.0 statistical application through univariate descriptive analysis test. The ethical clearance codes for this research are 01.258/KEPK/POLTEKKES.

RESULTS

Table 1. Type and Percentage Frequency of Bacteria from Dental Plaque

Type of Bacteria	Frequency	Percentage
Gram-negative Rod Bacteria		
Aeromonas salmonicida	5	6.3
Oligella ureolytica	4	5.1
Spingomonas paucimobilis	2	2.5
Gram-positive Rod Bacteria		
Rothia dentocariosa	19	24
Kocuria rosea	5	6.3
Gram positive coccus		
Granulicatella elegans	8	10,1
Staphylococcus aureus	3	3.7
Staphylococcus lentus	3	3.7
Streptococcus mitis	13	16.4
Streptococcus oralis	3	3.7
Streptococcus pluranimalium	3	3.7
Streptococcus pneumoniae	1	1.2
Streptococcus pseudoporcinus	1	1.2
Streptococcus salivarius	2	2.5
Streptococcus sanguinis	3	3.7
Streptococcus thermophilus	1	1.2
Yeast		
Candida famata	1	1.2
Candida albicans	2	2.5
Total	**79**	**100**

Based on the bacteria and yeast distribution Table 1, it can be seen that Gram-positive rod bacterial infections dominate, especially by Rothia dentocariosa which accounts for 24% of the total isolates. This indicates that these bacterial infections are more common than other bacteria, so there needs to be specific interventions aimed at controlling these infections. Effective antibiotic use and clinical guidelines for the management of infections caused by Gram-positive bacteria should be strengthened, with an emphasis on cross-infection prevention strategies in healthcare facilities.

In addition, infections caused by Gram-positive coccus bacteria are also quite significant, especially by Streptococcus mitis with a frequency of 16.4% as well as other species of the genus Streptococcus. Intervention measures need to focus on routine culture and sensitivity checks to ensure that antibiotics are used appropriately. Emphasis should be placed on antibiotic stewardship to avoid resistance that could complicate treatment in the future. In addition, improved hygiene and education about nosocomial infections for medical personnel is needed to reduce the incidence of infection by Gram-positive coccus bacteria.

Additional interventions required are related to the control of yeast infections such as Candida albicans and Candida famata, although the frequency of occurrence is relatively small

(2.5% and 1.2% respectively). High-risk patients such as those who are immunosuppressed should receive more attention in the prevention of these fungal infections. Appropriate use of antifungal agents, coupled with monitoring of the patient's immune condition, can help prevent more serious infections due to Candida.

DISCUSSION

The results showed that Gram-positive bacterial infections, particularly Rothia dentocariosa, predominate in dental plaque. These infections require more attention in management, especially through the use of appropriate antibiotics and strengthening clinical guidelines to prevent the spread of infection. In addition, coccus-type Gram-positive bacterial infections, such as Streptococcus mitis, also have a significant frequency, which demands strict management against antibiotic resistance. Although infections by fungi such as Candida albicans are relatively rare, special attention should be paid to high-risk patients such as those with compromised immunity, focusing on the appropriate use of antifungal agents and monitoring of the patient's condition.

The study also highlighted the effectiveness of gargling in reducing bacterial colonies, where the method significantly decreased colony counts in all samples tested. These results support the use of gargling as a preventive intervention in maintaining oral hygiene. In addition, increasing the concentration of antimicrobial agents was shown to expand the zone of bacterial inhibition, indicating that higher concentrations are more effective in inhibiting bacterial growth. These findings provide an important basis for the development of more potent antibacterial products, especially in oral health care.

Gram-positives are known to be more resistant to environmental changes, such as pH and humidity, which often occur in the oral cavity (Baquero et al., 2021). In addition, inappropriate or irregular use of antibiotics can be one of the contributing factors to the increased prevalence of these bacterial infections, as bacteria can develop resistance (Chokshi et al., 2019). Lack of awareness of the importance of routine dental care also contributes to the high infection by these bacteria (Haque et al., 2019). This confirms that interventions such as education on oral hygiene and rational management of antibiotic use need to be strengthened (Bessa et al., 2022).

In research on the effectiveness of gargling, the significant reduction in the number of bacterial colonies after gargling suggests that this simple practice can directly disrupt the survival of bacteria in the oral cavity (Inchingolo et al., 2022). Most likely, gargling is able to clean plaque and food debris that serve as a source of nutrition for bacteria (Juwita Husaini et

al., 2024). This suggests that gargling plays an important role in reducing the bacterial load on teeth and oral tissues, which if neglected can lead to the development of periodontal disease (Lee, 2022). In addition, the increase in bacterial inhibition zones at higher antimicrobial concentrations suggests that the right dose is a key factor in stopping bacterial growth, so optimal formulations are needed to improve the effectiveness of antimicrobial therapy in the prevention of oral infections (Li et al., 2017).

Firmansyah & Lingga (2024) research concluded that cinnamon bark extract (Cinnamomun burmannii) with a concentration of 10% and 20% has the ability to inhibit the growth of the fungus Trichophyton mentagrophytes. Cinnamon bark extract (Cinnamomun burmannii) with 20% concentration is more effective in inhibiting the growth of Trichophyton mentagrophytes fungus. (the highest clear zone) compared to cinnamon bark extract (Cinnamomun burmannii) with a concentration of 10%. After conducting research on the inhibition test of cinnamon bark extract (Cinnamomum burmannii) against the growth of dermatophyte fungi in vitro.

According to Intan et al., (2021) research, the most effective concentration of cinnamon to inhibit the growth of Staphylococccus aureus is a concentration of 75% with an average clear zone diameter of 12.7 mm. Consecutively, the best potential is the extract with a concentration of 65%, 70%, and the best is 75%. Based on the results of Putri et al., (2023) research on the inhibition test of cinnamon bark extract (Cinnamomum Burmannii Blume) with 96% ethanol solvent, it can be concluded that the antibacterial inhibition of cinnamon bark extract (Cinnamomum Burmannii Blume) against Staphylococcus aureus bacteria with concentrations of 40%, 50%, and 60% produces an inhibition zone. The most effective concentration of cinnamon bark (Cinnamomum Burmannii Blume) in inhibiting the growth of Staphylococcus aureus bacteria is at a concentration of 60% with an effectiveness level of 11.2 mm.

Siregar et al., (2023) research found that ethanol extract of cinnamon leaves (Cinnamomum burmannii) has an antibiotic effect on the growth of Staphylococcus aureus bacteria. The most effective concentration to stop the growth of Staphylococcus aureus bacteria is with 100% concentration of cinnamon leaf ethanol extract which has an intermediate inhibition response. Further research on the antimicrobial effect of cinnamon leaf extract (Cinnamomum burmannii) can be done with different methods, types of microorganisms and extract concentrations. Testing lower extract concentrations can be done to determine the minimum inhibitory level of cinnamon leaf extract (Cinnamomum burmannii) against the growth of Staphylococcus aureus.

Komala et al., (2018) research findings, Cinnamon in mask preparations has activity against staphylococus aureus bacteria, the higher the concentration of cinnamon extract in the mask, the greater the activity in inhibiting bacterial growth. Parhusip & Cynthia, (2019) research findings found that the ethyl acetate extract of cinnamon bark contains alkaloid, saponin, tannin, phenolic, flavonoid, triterpenoid, and glycoside phytochemical compounds. The selected concentration of cinnamon bark ethyl acetate extract is 5%, because it is able to form an inhibition diameter of more than 10 mm. E. coli bacteria had the largest MIC and MBC values with values of 0.55 and 2.18%.

Research on the efficacy of ethanol derived from cinnamon bark (*Cinnamomum spp.*) shows that ethanol extracts have significant antibacterial properties. The active chemicals, including cinnamaldehyde and eugenol, are thought to significantly suppress the growth of many bacteria, which include Gram-positive and Gram-negative strains. Cinnamaldehyde can damage bacterial cell walls and impair membrane function, ultimately resulting in bacterial cell death.

The efficacy of cinnamon bark ethanol in inhibiting bacterial proliferation is further corroborated by its capacity to disrupt biofilm formation, a protective coating established by bacteria to shield themselves from external environmental factors, including antimicrobials (Didehdar et al., 2022). Cinnamon bark ethanol can enhance bacterial exposure to antibacterial drugs and promote bacterial eradication by reducing biofilms (Elcocks et al., 2020). Cinnamon bark may be utilized in the development of oral health products or natural disinfectants to mitigate bacterial illnesses (Waty et al., 2018).

CONCLUSIONS

This study concludes that Gram-positive bacterial infections, notably Rothia dentocariosa and Streptococcus mitis, predominate in dental plaque and necessitate careful management, particularly with the judicious use of antibiotics. Furthermore, gargling was demonstrated to substantially diminish the quantity of bacterial colonies, illustrating its efficacy as a preventive measure. Elevated concentrations of antimicrobials were demonstrated to enhance the zone of bacterial inhibition, indicating that optimum dosing is crucial for efficiently preventing bacterial proliferation. These findings endorse the advancement of oral health care products that are more efficacious in infection prevention.

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