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EVALUATION OF ANTIBACTERIAL EFFICACY OF CINNAMOMUM BURMANII LEAF ESSENTIAL OIL IN TOOTHPASTE FORMULATION AGAINST STREPTOCOCCUS MUTANS: AN EXPERIMENTAL STUDY

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ABSTRACT

Background: Traditional cinnamon bark harvesting practices often discard valuable leaves, neglecting their potential benefits such as essential oil with antibacterial properties. Previous studies have highlighted the antibacterial effects of Cinnamomum burmannii leaves from Indonesia, including their potential as antibacterial agents. The study aimed to evaluate the antibacterial effect of Cinnamomum burmanii leaf essential oil in toothpaste formulation against Streptococcus mutans
Method: This quasi-experimental study compared the antibacterial effectiveness of toothpaste containing essential oil with Streptococcus mutans cultures in Brain-heart Infusion Broth (BHIB) overnight. Six wells were created on agar plates using sterile 6 mm punches. Each well was filled with the following: Positive control (Chlorhexidine), pure essential oil, 100% essential oil toothpaste, 75% diluted essential oil toothpaste, 50% diluted essential oil toothpaste, and negative control (aquadest). The diameter of inhibition zones was measured after incubating all plates at 36°C for 24 hours.
Result: The inhibition zone diameter was measured to evaluate the effectiveness of essential oil toothpaste against dental plaque bacteria. Non-parametric Kruskal-Wallis statistical analysis revealed no significant difference in the diameter produced between Cinnamomum zeylanicum leaf essential oil and the positive control minosep (p=0.10). However, after formulation into toothpaste, a significant decrease in effectiveness was observed (p=0.015).
Conclusion: It can be concluded that cinnamon leaf essential oil is effective in inhibiting the growth of Streptococcus mutans bacteria. However, when formulated into toothpaste, its effectiveness decreases with lower concentrations of the toothpaste dilution..

Keywords: *Cinnamomum burmannii*; *Streptococcus mutans;* essential oil; toothpaste; antibacterial efficacy

INTRODUCTION

Cinnamon (genus *Cinnamomum*, family *Lauraceae*) is a spice primarily derived from the bark of the cinnamon tree, an evergreen belonging to the Lauraceae family. Prominent species include Cinnamomum cassia (L.) J. Presl, Cinnamomum camphora, Cinnamomum burmannii and Cinnamomum zeylanicum. This plant has been discovered for centuries predominantly for its culinary uses. Renowned for its distinctive aroma, cinnamon is also used for perfume compositions (Błaszczyk et al., 2021).

The extensive use of cinnamon bark in human life undoubtedly results in a high demand for its harvesting. When harvesting cinnamon bark, the leaves are typically discarded and become waste. Conventional methods of harvesting cinnamon bark typically involve unintentionally disregarding its associated leaves, leading to the forfeiture of a valuable resource. These leaves, constituting an essential component of the Cinnamomum tree, possess untapped potential, particularly in the form of essential oil endowed with significant antibacterial properties. Regrettably, the oversight of these leaves has endured, hindering the acknowledgment of their intrinsic therapeutic benefits over the years.

Cinnamomum burmannii, also known as Cassiavera or Korintje cinnamon, is an endemic species in Indonesia and native to Sumatra, grows along with the Bukit Barisan mountain range, from Aceh, in North Sumatra, down through west Sumatra, Jambi, Bengkulu, and Lampung. In Jambi itself, Cinnamon is highly produced in Kerinci (Menggala et al., 2019).

The antimicrobial active compounds cinnamaldehyde and eugenol are the main reasons for the antibacterial activity of cinnamon leaves. Previous studies showed that Cinnamon extract has antibacterial effectivity against five common food-borne pathogenic bacteria such as Bacillus cereus, Listeria monocytogenes, Staphylococcus aureus, Escherichia coli, and Salmonella anatum (Al-Dhubiab, 2012; Parisa et al., 2019).

The high trans-cinnamaldehyde levels in cinnamon are also has anti- inflammatory effects. A decrease in the volume of edema, a decrease in the number of inflammatory cells and TNF-α were found in previous study using the essential oil of cinnamon on paw of rat (Budiastuti et al., 2021).

In prior studies, it was established that the extract from cinnamon leaves has demonstrated the capability to inhibit the growth of the Microsporum sp. fungus. This observation underscores another potential of antifungal properties in cinnamon leaf extracts. The findings from earlier research contribute valuable insights into the inhibitory effects of cinnamon leaf extract on Microsporum sp. Fungi (Siagian & Lubis, 2021)

Further beneficial of the cinnamon for human health was analgesic, antidiabetic activity antioxidant activity, anti tumor, antithrombotic and immune response activity. Inhibition of dental plaque formation and periodontal disease also found in previous studies(Al-Dhubiab, 2012).

Strategies for preventing plaque formation in oral health encompass both mechanical methods, such as brushing with toothpaste, and chemical interventions, like the application of mouthwash. These preventive measures played important role in the progression of oral diseases such as caries and periodontal diseases. Although brushing is effective in plaque removal, it does not guarantee that the eradicated plaque won't swiftly reappear post-brushing. In response to this issue, diverse antibacterial elements are integrated into the formulations of toothpaste and mouthwash to impede the resurgence of plaque. Essential oils, extensively employed in both mouthwash and toothpaste, are recognized as agents with anti-plaque properties among these various components(Fitria et al., 2023).

Previous study has shown the effectiveness of cinnamon leaf on other bacteria such as Staphylococus aureus and Estherichia coli (Jafri et al., 2021; Nabila et al., 2021; Parisa et al., 2019), but lack evidence of the effectiveness on bacteria that has major role in dental caries such as Streptococcus mutans. Therefore, this present study aimed to evaluate the antibacterial effect of Cinnamomum burmanii leaf essential oil in toothpaste formulation against Streptococcus mutans.

METHOD

A quasi-experimental method was employed on this research. Toothpaste was formulated using essential oils extracted from cinnamon leaves from Kerinci, Jambi as the active ingredient. The formula comprises of 3 ml of essential oil, Karboppol 934 0,5 gr, Tween 80 0,25 ml, Gliserin 0,25gr, Sodium Benzoate 0,25 gr, Trietanolamin 0,31 gr, Aquadest 20,4 ml.

Streptococcus mutans (ATCC25175) was grown on Brain-heart Infusion Broth

(BHIB). Six wells were created on agar plates using sterile 6 mm punches. Each well was filled with the following: Positive control (Chlorhexidine), pure essential oil of the cinnamon, 100% essential oil toothpaste, 75% diluted essential oil toothpaste, 50% diluted essential oil toothpaste, and negative control (aquadest). The diameter of inhibition zones was then measured after all plates were incubated at 36°C for 24 hours.

Data of the inhibition zone on Streptococcus mutans were collected and analyzed with the Kruskal Wallis to evaluate the effectiveness of the toothpaste containing cinnamon leaf essential oil on Streptococcus mutans.

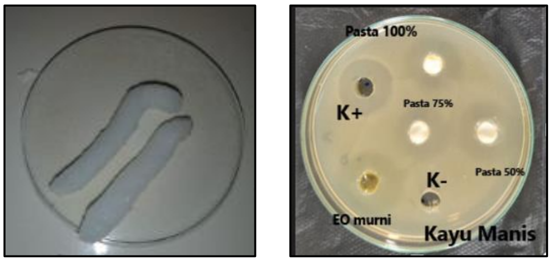
RESULT AND DISCUSSION

The inhibition zone diameter was measured to evaluate the effectiveness of essential oil toothpaste against dental plaque bacteria. Non-parametric Kruskal-Wallis statistical analysis revealed no significant difference in the diameter produced between Cinnamomum burmannii leaf essential oil and the positive control Chlorhexidine (p=0.10). However, after formulation into toothpaste and dilution, a significant decrease in effectiveness was observed (p=0.015).

Table 1. Mean Inhibition of Streptococcus mutans

Variable	Mean Inhibition (cm)(±SD)	p value
Control +	2,51 (±0,36)	0,10 ƚ
Pure Essential Oil	2,14 (±0,29)	
100% EO toothpaste	1,77 (±0,29)	0,015 ƚƚ
75% EO toothpaste	1,69 (±0,28)	
50% EO toothpaste	1,67 (±0,29)	
Control -	0	

ƚ Kruskal Waliis Test between pure EO with positive control
ƚƚKruskal Wallis Test between toothpaste with dilution



Picture 1. (Left) paste of Cinamon leaf essential oil; (right) inhibition zone

The cinnamon plant contains cinnamaldehyde, cinnamate, cinnamic acid, and several types of essential oil compounds as its main components. The secondary metabolite content in a plant is influenced by various factors, both internal and external. Internal factors such as genetics, and external factors include light, temperature, humidity, pH, nutrient content in the soil, and altitude. Different altitudes result in different temperatures. Altitude is one of the factors that affect the growth of a plant. A series of metabolic processes in plants will be disrupted, leading to differences in the compounds produced at different altitudes. These factors can affect the amount of antibacterial substances present in a sample. This can lead to the absence of an inhibition zone due to the inability to damage the cell membrane and disrupt the physiological processes of the cell.(Nabila et al., 2021)

Streptococcus mutans is a gram-positive bacteria which its cell wall was composed mainly of peptidoglycan with fewer LPS. Therefore, gram-positive bacteria are more polar, making it easier for the essential oil to penetrate their cell walls. Previous study showed that when compared to gram negative bacteria such as Escherichia coli, the Cinnamomum burmanii extract has higher sensitivity to gram positive bacteria such as Staphylococcus aureus (Angelica, 2014).

The saponin compounds in cinnamon leaves can kill bacteria by damaging the bacterial cell membrane, leading to cell death. Flavonoids halt bacterial growth by altering protein structure and causing damage to the bacterial cell membrane by breaking down fats in the cell wall. When the cell membrane is damaged, the activity and biosynthesis of specific enzymes necessary for metabolic reactions cannot occur, resulting in bacterial death. Tannins cause the bacterial cell membrane to shrink, making the cell more permeable. Consequently, bacterial metabolism becomes disrupted, leading to lysis and death. Alkaloids work by inhibiting the peptidoglycan portion of the bacterial cell wall. Peptidoglycan is a chemical that stiffens the cell wall, maintaining the cell's shape. If the component responsible for peptidoglycan is damaged, the bacterial cell wall layer does not form properly, causing the cell to die(Siregar et al., 2023).

By combining with vanillin, cinnamon leaves essential oil were also noteworthy in giving antimicrobial effect when used in food packaging. It will decrease the use of chemical additives for preservatives of packaged food that will have negative health consequences, such as carcinogenicity, teratogenicity, and toxicity, as well as environmental problems related to their long periods of degradation (Cava-Roda et al., 2021).

While pure Cinnamomum burmannii leaf essential oil demonstrated antibacterial properties against Streptococcus mutans, its effectiveness was significantly reduced when incorporated into toothpaste. These findings shed light on the challenges of translating essential oil's antibacterial potential from its raw form to a formulated dental product.

The promise of C. burmanii leaf essential oil as a natural alternative to synthetic antimicrobial agents. This is particularly significant given the growing concerns about antimicrobial resistance and the desire for safe and sustainable oral care products. Decline in antibacterial efficacy warrants further investigation and may be attributed to several factors such as formulation challenges related to stability, compatibility, and concentration.

Interactions between essential oil compounds and other toothpaste ingredients may impact the antibacterial activity of the oil. Future research should explore methods to enhance the stability and efficacy of essential oils in toothpaste formulations, potentially through encapsulation techniques or modified delivery systems. These observations shed light on the practical implications of utilizing cinnamon leaf essential oil as a potential natural antimicrobial agent in oral care products.

CONCLUSION

It can be concluded that cinnamon leaf essential oil is effective in inhibiting the growth of Streptococcus mutans bacteria. However, when formulated into toothpaste, its effectiveness decreases with lower concentrations of the toothpaste dilution.

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