

The Effectiveness of Clove Leaf Extract (*Syzygium Aromaticum*) in Cleaning Smear Layer in Tooth Root Canals

Nur Asmah,¹ Syamsiah Syam¹, Sarahfin Aslan¹, Widya Lestari², Resky Masyandah³

¹Department of Conservative Dentistry, Faculty of Dentistry, Universitas Muslim Indonesia, Makassar, Indonesia

²Department of Fundamental Dental and Medical Sciences, Kulliyah of Dentistry, International Islamic University Malaysia, Kuantan

³Student, Faculty of Dentistry, Universitas Muslim Indonesia, Makassar, Indonesia

Corresponding Author: nur.asmah@umi.ac.id

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ABSTRACT

The target of root canal treatment is to eliminate infected pulp tissue and preserve tooth function. Instrumentation produces a smear layer composed of organic and inorganic debris, which may promote microbial colonization and reduce sealer adhesion. Ethylene diamine tetra acetic acid (EDTA) 17% is widely used for smear layer removal but may cause periapical tissue toxicity, suggesting the need for safer natural alternatives. Clove leaf extract (*Syzygium aromaticum*) contains bioactive compounds such as flavonoids, phenolics, saponins, steroids, and alkaloids that may aid in smear layer removal. Therefore, this study aimed to evaluate the effectiveness of clove leaf extract in removing the smear layer from root canals. The extract was prepared using the maceration method and tested at concentrations of 10%, 15%, and 20%, with 17% EDTA used as the control group. The smear layer cleanliness was evaluated with a Scanning Electron Microscope (SEM), and data were analyzed using the Mann-Whitney test. Statistical analysis showed no significant difference between the 20% clove leaf extract group and the 17% EDTA group ($p > 0.05$). The 20% concentration showed comparable effectiveness to 17% EDTA in removing the smear layer from root canals.

Keyword: *Syzygium Aromaticum*; Smear Layer; Root Canal Irrigation

ABSTRAK

Perawatan saluran akar gigi bertujuan membersihkan jaringan pulpa terinfeksi melalui tahap preparasi, sterilisasi, dan obturasi untuk mempertahankan fungsi gigi. Proses instrumentasi menghasilkan *smear layer* yang terdiri dari komponen organik dan anorganik, yang dapat membentuk mikroorganisme dan mengganggu adhesi sealer pada saluran akar gigi. *Ethylene Diamine Tetra Acetic Acid* (EDTA 17%) digunakan untuk membersihkan *smear layer*, namun bersifat toksik terhadap jaringan periapikal. Diperlukan bahan alami yang biokompatibel. Ekstrak daun cengkeh (*Syzygium aromaticum*) mengandung flavonoid, fenolik, saponin, steroid dan alkaloid yang efektif membersihkan *smear layer* pada saluran akar gigi. Penelitian ini bertujuan untuk mengetahui efektivitas ekstrak daun cengkeh (*Syzygium aromaticum*) dalam membersihkan *smear layer* pada saluran akar gigi. Ekstrak dibuat menggunakan metode maserasi dan diuji pada konsentrasi 10%, 15%, dan 20%, dengan EDTA 17% sebagai kelompok kontrol positif. dan dievaluasi menggunakan SEM. Analisis statistik dilakukan dengan uji *Mann-Whitney*. Nilai *p-value* sebesar 0,575 ($p-value > 0,05$) menunjukkan tidak terdapat perbedaan signifikan antara ekstrak daun cengkeh 20% dan EDTA 17% dalam kebersihan *smear layer* pada saluran akar gigi. Ekstrak daun cengkeh konsentrasi 20% menunjukkan efektivitas yang sebanding dengan EDTA 17% dalam membersihkan *smear layer* pada saluran akar gigi.

Kata kunci: Ekstrak Daun Cengkeh (*Syzygium Aromaticum*), *Smear Layer*, Kebersihan Saluran Akar Gigi



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1. Introduction

Root canal treatment is a therapeutic procedure performed on teeth affected by pulp infection or damage to relieve pain, eliminate microorganisms, restore function, and maintain dental esthetics. The primary objective of root canal treatment is to prevent the spread of pathological processes from the pulp tissue to the periapical tissues, and restore the biological condition of the tooth for optimal acceptance by the surrounding periodontal tissues. The treatment has three main stages, namely (1) biomechanical preparation, which includes cleaning and shaping of the root canal, (2) sterilization through irrigation and the use of intracanal medicaments, and (3) obturation consisting of hermetic filling of the root canal to prevent reinfection [1,2].

During the root canal preparation stage, the instrumentation process leads to the formation of a smear layer on the surface of the root canal walls. The smear is an amorphous layer with a thickness of approximately 1–5 μm , composed of organic and inorganic components, including dentin debris, remnants of pulp tissue, and microorganisms. The presence of this material reduces the success of root canal treatment through the support provided to the proliferation of residual microorganisms within the area and the inhibition of sealer adhesion to the root canal wall, increasing the risk of microleakage following obturation. Therefore, smear layer elimination is considered a critical factor in achieving successful root canal treatment [3,4].

Smear layer removal can be achieved using root canal irrigants that possess demineralizing and chelating properties [5]. Root canal irrigation is a crucial component of the sterilization phase, aimed at removing necrotic tissue, microorganisms, and inorganic debris from the root canal system [6,7]. The appropriate use of irrigating solutions enhances the effectiveness of mechanical cleaning and assist in the decontamination of areas within the root canal that are difficult to reach using instruments [8].

Sodium hypochlorite (NaOCl) at concentrations of 0.5%–2.5% is the most commonly used irrigating solution due to the bactericidal potential and the ability to dissolve necrotic pulp tissue [9]. As an adjunctive agent, ethylene diamine tetra acetic acid (EDTA) 17% is used for chelation to remove the inorganic component of the smear layer from the root canal walls. Although the combination of 2.5% NaOCl and 17% EDTA has been proven effective, both agents are known to show toxic and irritative effects on periapical tissues when applied excessively. Therefore, alternative irrigating agents with better biocompatibility are required [10,11].

Indonesia is rich in natural resources, such as the clove plant (*Syzygium aromaticum*) [12]. The extract of this plant offers various benefits due to being an easily accessible herbal remedy that is relatively safe and more affordable than some conventional treatments for dental and oral problems [13]. The leaf is available year-round and contains various beneficial bioactive compounds [14]. According to a study by Dewi et al. (2021), ethanol extract of clove leaf contains secondary metabolites such as saponins, phenols, terpenoids, glycosides, flavonoids, and tannins. The chemical structure of saponins consists of glycosides composed of a glycone and an aglycone. The glycone portion has sugar groups such as glucose, fructose, and other types of sugars, while the aglycone is a sapogenin. This amphiphilic nature allows saponins to function as natural surfactants (Nurzaman, 2018). Saponins can reduce the surface tension of aqueous solutions and produce foam, thereby providing soap-like properties.

The ability of saponins enhances wetting and the penetration of the solution into the root canal system, potentially aiding the removal of organic debris and supporting the detachment of the smear layer—which contains both organic and inorganic components—from the dentin surface. In addition, phenolic compounds damage bacterial cell walls through protein denaturation, leading to a decrease in cell membrane permeability, inhibiting bacterial growth and initiating cell death. Eugenol is among the main phenolic compounds in clove leaves. This is a hydrophobic compound capable of penetrating and damaging cell membranes, particularly in Gram-negative bacteria, which have a high lipid content. Eugenol decreases cell viability by affecting the release of intracellular substances and inhibiting the production of β -lactamase enzymes and urease activity, thereby reducing the ability of the bacteria to survive [12,15]. Based on the bioactive content and surfactant properties, ethanol extract of clove leaf has the potential to be used as an alternative irrigation agent to aid in smear layer removal from root canals [16,17].

2. Methods

Approximately 5 kg of clove leaf (*Syzygium aromaticum*) were collected, washed under running water, cut into small pieces, and dried in a dry cabinet at 40°C for 3 days. The dried leaf was ground into powder using a blender. A total of 500 g of clove leaf powder was extracted using the maceration method with 2 L of 96% ethanol for 3 × 24 hours, which was stirred every 24 hours. The extract was filtered with Whatman filter paper and concentrated using a rotary evaporator at 40°C to obtain a thick extract.

Phytochemical screening was performed to identify the presence of flavonoids, phenolics, saponins, steroids, terpenoids, and alkaloids. The concentrations of 10%, 15%, and 20% were adopted from a previous study on the antibacterial activity of clove leaf extract, which used the same concentrations [16]. These concentrations were selected to evaluate the effectiveness in removing the smear layer from root canals. Approximately 1.25 g, 1.50 g, and 1.75 g of the thick extract was diluted in 5 mL dimethyl sulfoxide (DMSO) to obtain concentrations of 10%, 15%, and 20%, respectively. A total of 24 extracted maxillary incisors were collected and stored in saline solution at room temperature before treatment. The samples were randomly divided into four groups (n = 6), including Group I (10% clove leaf extract), Group II (15% clove leaf extract), Group III (20% clove leaf extract), and Group IV (17% EDTA, positive control). The crowns were sectioned at the cemento-enamel junction (CEJ) using a diamond disc. Access cavities were prepared, and the working length was determined using a size #10 K-file until the file tip was visible at the apical foramen and then reduced by 1 mm. Root canal preparation was performed using K-files up to size #80.

During instrumentation, irrigation with 2.5% NaOCl was performed at each file change. Following canal preparation, each canal was irrigated with 2 mL of the assigned solution for 10 seconds, then dried using paper points. Smear layer cleanliness in the apical third of the root canal was evaluated using a Scanning Electron Microscope (SEM) at 4000× magnification. The SEM images were assessed using the *Hülsmann* scoring system. Score (1) implies that all dentinal tubule orifices are open, and the surface is free of smear layer. Score (2) shows that some dentinal tubule orifices are open with a small amount of smear layer present. Score (3) implies that only a few dentinal tubule orifices are open, and the smear layer covers part of the surface. Score (4) shows that all dentinal tubule orifices are closed, and the entire surface is covered by smear layer. Score (5) represents heavy smear layer, with a thick layer covering the entire surface and dentinal tubule orifices. Lower scores indicate cleaner root canal walls and a greater number of open dentinal tubes. Statistical analysis was performed to compare smear layer cleanliness among the experimental groups and the positive control group. This study received ethical approval from the health Research Ethics committee of the Muslim University of Indonesia with the Approval No: 688/A.1/KEP-UMI/IX/2025.

3. Results

This study evaluated the effectiveness of clove leaf extract at concentrations of 10%, 15%, and 20% in removing the smear layer from root canals. The extract testing was conducted with six repetitions for each solution to determine the effectiveness. A total of 24 samples were used, and phytochemical analysis was performed. The results of the color-based phytochemical screening of clove leaf extract are presented in Table 1.

Table 1. Phytochemical Screening Results of Clove Leaf Extract (*Syzygium aromaticum*)

Test	Reagent	Result	Observation
Flavonoids	Magnesium powder and HCl	+	Formation of a reddish precipitate
Phenolics	FeCl ₃	+	Formation of a blue precipitate
Saponins	Distilled water and 2N HCl	+	Formation of foam
Steroids	Liebermann–Burchard	+	Formation of a bluish-green ring
Terpenoids	Liebermann–Burchard	-	No formation of brownish or violet ring
Alkaloids	Mayer, Wagner, Dragendorff	+	Formation of white, brown, and orange precipitates

Note: (+) = Positive for the presence of compounds. (–) Negative for the presence of compounds.

The effectiveness of smear layer removal was assessed based on the absence of smear layer covering the dentinal tubules in the root canals, as observed using an SEM. The cleanliness of root canals irrigated with clove leaf extract at concentrations of 10%, 15%, and 20%, as well as with 17% EDTA, was examined using SEM at a magnification of 4000× for each group. Additionally, the level of smear layer cleanliness was assessed with the scoring system based on the *Hülsmann* criteria.

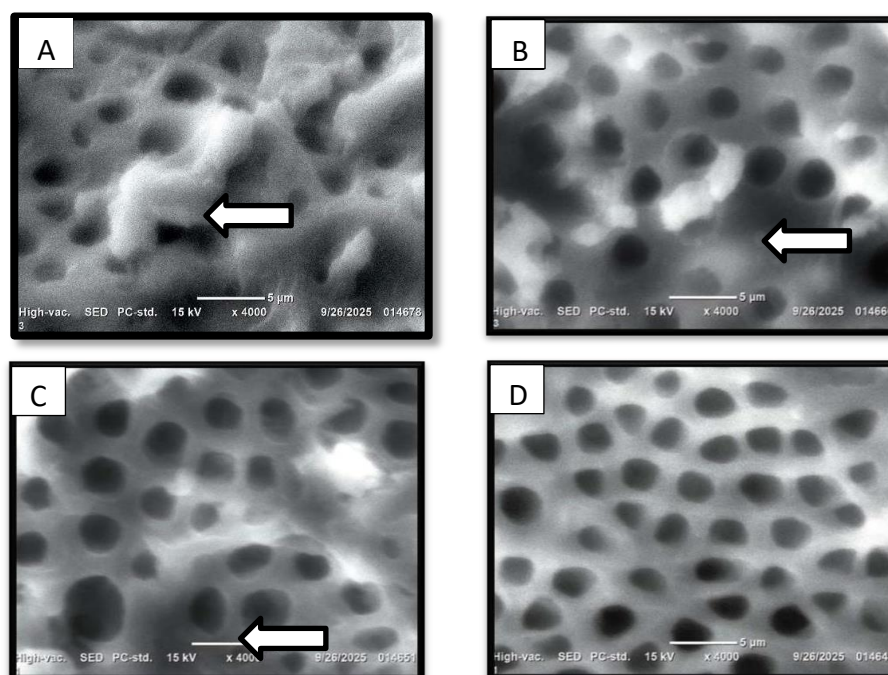


Figure 1. Representative SEM images (4000× magnification) showing smear layer cleanliness in the apical third of root canals irrigated with (A) 10% clove leaf extract, (B) 15% clove leaf extract, (C) 20% clove leaf extract, and (D) 17% EDTA.

Table 2. Smear layer cleanliness scores of root canals irrigated with clove leaf extract (*Syzygium aromaticum*) at concentrations of 10%, 15%, and 20%, and with 17% EDTA

Sa mple	Extract 10%	Extract 15%	Extract 20%	Extract 17%	EDTA
1	3	2	1	1	
2	3	2	2	1	
3	3	3	2	1	
4	4	3	1	2	
5	5	3	1	1	
6	4	3	2	2	

Note: Smear layer cleanliness was assessed using the *Hülsmann* scoring system.

Score (1) implies that all dentinal tubule orifices are open, and the surface is free of smear layer. Score (2) shows that some dentinal tubule orifices are open with a small amount of smear layer present. Score (3) indicates that only a few dentinal tubule orifices are open, and the smear layer covers part of the surface. Score (4) implies that all dentinal tubule orifices are closed, and the entire surface is covered by smear layer. Score (5) represents heavy smear layer, with a thick *layer* covering the entire surface and dentinal tubule orifices. Meanwhile, lower scores indicate cleaner root canal walls and a greater number of open dentinal tubes.

Based on Table 2, an improvement in smear layer cleanliness was observed with increasing concentrations of clove leaf extract. The 10% concentration had relatively high cleanliness scores (scores 3–5), indicating that the smear layer still covered most of the dentin surface. At 15%, cleanliness scores improved (scores 2–3), with a greater number of open dentinal tubule orifices. The highest effectiveness was observed with the 20% extract, which showed scores of 1–2, indicating consistent and effective smear layer removal. Similarly, 17% EDTA

showed scores of 1–2 as the gold standard for smear layer removal, indicating that 20% clove leaf extract had a performance nearly comparable to 17% EDTA.

Table 3. Descriptive analysis of smear layer cleanliness based on clove leaf extract (*Syzygium aromaticum*) and 17% EDTA

Clove Leaf Extract	Mean	Std. Deviation	<i>p</i> -value
Extract 10%	3.667	0.816	0.000
Extract 15%	2.667	0.516	
Extract 20%	1.500	0.548	
EDTA 17%	1.333	0.516	

Notes: Statistical test: *Kruskal–Wallis* test; significant ($p < 0.05$).

Table 3 shows that the smear layer cleanliness data of the root canals were not entirely normally distributed. However, the data were homogeneous ($p > 0.05$). The *Kruskal–Wallis* test found a statistically significant difference among the treatment groups ($p = 0.000 < 0.05$). The highest mean score was observed in the 10% clove leaf extract group, while the lowest was found in the 17% EDTA group. The 20% extract had a mean score closest to the value obtained for 17% EDTA, indicating better effectiveness compared with the other extract concentrations.

Table 4. Comparative analysis of smear layer cleanliness based on clove leaf extract (*Syzygium aromaticum*) and 17% EDTA

Clove Leaf Extract	Mean	Post hoc Comparison			
		Extract 10%	Extract 15%	Extract 20%	EDTA 17%
Extract 10%	3.667		0.031*	0.003*	0.003*
Extract 15%	2.667			0.011*	0.007*
Extract 20%	1.500				0.575
EDTA 17%	1.333				

*Notes: Post hoc comparison using the *Mann–Whitney* test; significant ($p < 0.05$).

Table 4 shows significant differences in *smear layer* cleanliness scores among the treatment groups ($p < 0.05$). Clove leaf extract at concentrations of 15% and 20% produced better results compared with the 10% concentration. The 20% extract achieved greater effectiveness in root canal cleaning than the 10% and 15% concentrations and did not show a significant difference when compared with 17% EDTA ($p > 0.05$).

4. Discussion

This study investigated the use of clove leaf extract, an herbal material that contains various bioactive compounds, including flavonoids, phenolics, saponins, steroids, and alkaloids. The compounds possess biological activities that may support root canal cleaning and decontamination, thereby suggesting clove leaf extract as an alternative irrigating agent in root canal treatment. Chemically, saponins are glycoside compounds composed of aglycone and glycone components. The aglycone, known as sapogenin, is hydrophobic and capable of binding inorganic components. Meanwhile, the glycone consists of sugar groups such as glucose, fructose, and other sugars that are hydrophilic and readily soluble in water, facilitating the smear layer removal through the irrigating solution

flow. This amphiphilic property enables natural materials containing saponins to function as surfactants and emulsifiers (detergents), dissolving both organic and inorganic components of the smear layer [16,18].

Plants containing saponins, which act as natural surfactants, are capable of reducing water surface tension and forming foam when agitated. The presence of saponins in various herbal extracts contributes to the ability to remove the smear layer. Additionally, clove leaf contains eugenol, which has antibacterial activity. Eugenol is hydrophobic in nature and can disrupt cell membranes, particularly in Gram-negative bacteria that have a high lipid content [16,19].

Previous studies have reported that herbal plants containing saponins, such as lime (*Citrus aurantifolia*), cocoa pulp extract, and bilimbi (*Averrhoa bilimbi*), show the ability to remove the *smear layer*. However, removal effectiveness varies among different plants, which may be influenced by differences in saponin concentration [14,15,20].

In this study, 17% EDTA was used as a comparison because of the wide recognition as an effective chelating agent for smear layer removal. EDTA functions by binding calcium ions from inorganic dentin, leading to the dissolution of hydroxyapatite minerals and the opening of dentinal tubules. Despite the effectiveness, EDTA has limitations, including low antibacterial activity and the potential to cause irritation to periapical tissues when used excessively [10,11].

The greater smear layer removal effectiveness observed in the 20% clove leaf extract group may be attributed to the higher concentration of saponins and other bioactive compounds present in the extract. Saponins act as natural surfactants by reducing surface tension and enhancing the penetration of the irrigating solution into dentinal tubules. At higher concentrations, the surfactant activity becomes more pronounced, facilitating more effective removal of debris and smear layer from the dentinal surface. Consequently, the 20% extract had superior cleaning efficacy compared with the 10% and 15% concentrations.

Based on the results, increasing concentrations of clove leaf extract were associated with greater smear layer removal effectiveness in root canals. The 20% concentration showed the best performance among the clove leaf extract groups, with removal efficacy that was most comparable to 17% EDTA. This suggests that the higher concentration of saponins enhanced the surfactant activity, improving irrigant penetration into dentinal tubules and facilitating more effective removal. Although EDTA removes the inorganic component of the smear layer through calcium chelation, the 20% clove leaf extract may achieve comparable cleaning efficacy through the combined surfactant activity of saponins and the antimicrobial properties of eugenol.

5. Conclusion

In conclusion, the test results showed that clove leaf extract had varying abilities in removing the smear layer from root canals depending on the concentration used. Clove leaf extract at concentrations of 10% and 15% was considered less effective, as residual smear layer remained on the dentin surface of the root canals. However, the 20% concentration had the effectiveness most comparable to 17% EDTA in removing the smear layer from root canals.

6. Acknowledgement

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7. Conflict of Interest

The authors declare that there is no conflict of interest regarding this study.

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