

## Effectiveness of Triton Irrigation Solution in Smear Layer Removal: An *in-vitro* Study

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### ABSTRACT

**Objective:** This *in vitro* study aimed to compare and evaluate the effectiveness of different irrigation solutions, including Triton, 0.5% Chitosan nanoparticles (CNP), and 17% ethylenediaminetetraacetic acid (EDTA), on the smear layer removal of the root canal walls.

**Methods:** Forty extracted sound mandibular premolars were examined; the samples were decoronated to obtain a root length of 14 mm. Each sample was instrumented using ProTaper Next rotary file X4 (40/0.06). The samples were longitudinally sectioned and examined under a scanning electron microscope at 3000x magnification in the coronal, middle, and apical thirds using a four-level scoring system.

**Results:** Triton demonstrated the lowest mean smear layer removal ( $p>0.05$ ) compared to the other irrigation solutions at all the levels of the root canal. No significant differences were observed ( $p>0.05$ ) at the coronal and middle levels of the root canal between the CNP and EDTA groups. CNP demonstrated significantly more smear layer removal at the apical level compared to EDTA.

**Conclusion:** Smear layer removal was least effective with Triton at all the levels of the root canal compared to the other irrigation solutions tested in this study. CNP demonstrated superior smear layer removal at the apical level compared to the other irrigation solutions.

**Keywords:** Chitosan nanoparticles, ethylenediaminetetraacetic acid, irrigation solution, smear layer removal, triton

### HIGHLIGHTS

- Triton irrigation is less effective in the removal of the smear layer than 0.5% CNP and 17% EDTA at three levels.
- 0.5% CNP removes the smear layer better than other irrigation at apical level.
- 0.5% CNP and 17% EDTA remove the smear layer in the same manner at the coronal and middle levels.

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### INTRODUCTION

Root canal preparation involves the combined action of endodontic instruments and irrigation solutions to ensure thorough disinfection. Mechanical instrumentation of the canal creates a granular amorphous smear layer containing both organic and inorganic substances that cover the canal wall and occlude the openings of the dentinal tubules (1). Inadequately cleaned areas may harbor bacteria and debris that can cause lingering infections and ultimately result

in failure of the root canal therapy. Root canal cleansing and cleaning are clinically challenging owing to the complex anatomy of the root canal (2). In a systematic review, Shahravan et al. (3) investigated if removing the smear layer avoids leakage after root canal filling. Of the comparisons, 54% found no significant difference in the leakage of root canal filling if the smear layer removed or not removed and 41% recommended removing the smear layer in order to accomplish better outcome of root canal filling and stop

leaks, and 5% said they would keep it. This systematic review concluded that while other factors like the type of sealer or the filling technique cannot have a significant impact, smear layer removal can effectively promote a fluid-tight seal. The root canal geometrics and structure morphology, the type and quantity of irrigation solution used during instrumentation, the instrument configuration, and other factors all affect the thickness of the smear layer (4). Therefore, irrigation is considered an important factor in the cleaning and shaping of root canals; it aids in eliminating necrotic tissue and dentine debris from manually or mechanically prepared areas. The ideal requirements of irrigation solutions are as follows: low cost, good cleaning action, friction reduction, improved dentine cutting by instruments, temperature control, good root canal system penetration, nontoxic to periapical tissue, and non-allergenic nature (5).

Sodium hypochlorite (NaOCl) (2.25–5.25%) is the most frequently used irrigation solution for root canal treatment; however, its action is limited to the removal of the organic component of the smear layer when used alone. Thus, chelating agents, such as ethylenediaminetetraacetic acid (EDTA), are used in combination with NaOCl as a final irrigant for the removal of the inorganic component of the smear layer (6, 7). EDTA is the most widely used irrigant for smear layer removal. In addition to cleaning, it also decalcifies dentine within 5 minutes at depths of 20–30  $\mu\text{m}$  by interacting with the calcium ions in dentine, thereby causing calcium chelation (8). EDTA, which has a powerful demineralizing effect, causes widening of the dentinal tubules, softening of dentine, and denaturation of collagen fibers (9). This could in turn interfere with the adaptation of the obturating material to the root canal walls. Another drawback of EDTA is that it is regarded as a contaminant because it is not originally found in nature (10). Researchers investigated a number of alternatives to EDTA to reduce the adverse effects of irrigants on the periapical tissues. Chitosan is another potential chelating agent, which is obtained by deacetylating chitin, a natural substance that is abundant in nature, inexpensive, and is a component of crab and shrimp shells (11). Chitosan, a natural glucosamine, possesses many favorable properties such as biocompatibility, biodegradability, bio-adhesion, and antimicrobial activity (12, 13) and is used in various fields, such as food, cosmetics, biomedical, and pharmaceutical applications (14). Additionally, the high biocompatibility and superior chelating capacity for different metallic ions in acidic environments (1) make Chitosan a promising potential irrigant in the field of dental research. Because Chitosan nanoparticles (CNP) have greater absorption and penetration into the dentinal tubules, they have been used to maximize the efficiency of Chitosan for root canal irrigation (15). Triton (Brasseler, Savannah, USA) is a newly developed endodontic irrigation solution that combines the advantages of NaOCl, EDTA, and chlorhexidine (CHX) in a single-step (all-in-one irrigation solution) using a lower concentration of NaOCl solution and a patent-pending unique combination of surfactants and mild chelating agents. Triton functions differently from traditional irrigants or other advanced solutions, as it is used for as a single irrigation along the procedure rather than as a final irrigant. Triton comprises two parts with distinct components, with Part A containing chelators (CA), surfactants, pH modifiers, and stabilizers, while

Part B containing 8% NaOCl and a pH modifier. The automix technique precisely mixes the two parts to deliver the final solution (4% NaOCl), allowing for simultaneous organic and inorganic tissue dissolution. Triton's non-NaOCl ingredients actively dissolve dentinal debris, thereby reducing the amount of buffering action required when the organic debris are exposed to a lower concentration of NaOCl. Triton shortens the chairside time by precluding the need for several irrigation solutions and sterile water rinses. To the author's knowledge, there were limited studies on the efficiency of Triton on smear layer removal.

Thus, this study aimed to compare and evaluate the effectiveness of different irrigation solutions, including Triton, 0.5% Chitosan nanoparticles (CNP), and 17% ethylenediaminetetraacetic acid (EDTA), on the smear layer removal of the root canal walls. The null hypothesis that there is no significant difference in the smear layer removing efficacy of Triton in comparing with other irrigants.

## MATERIALS AND METHODS

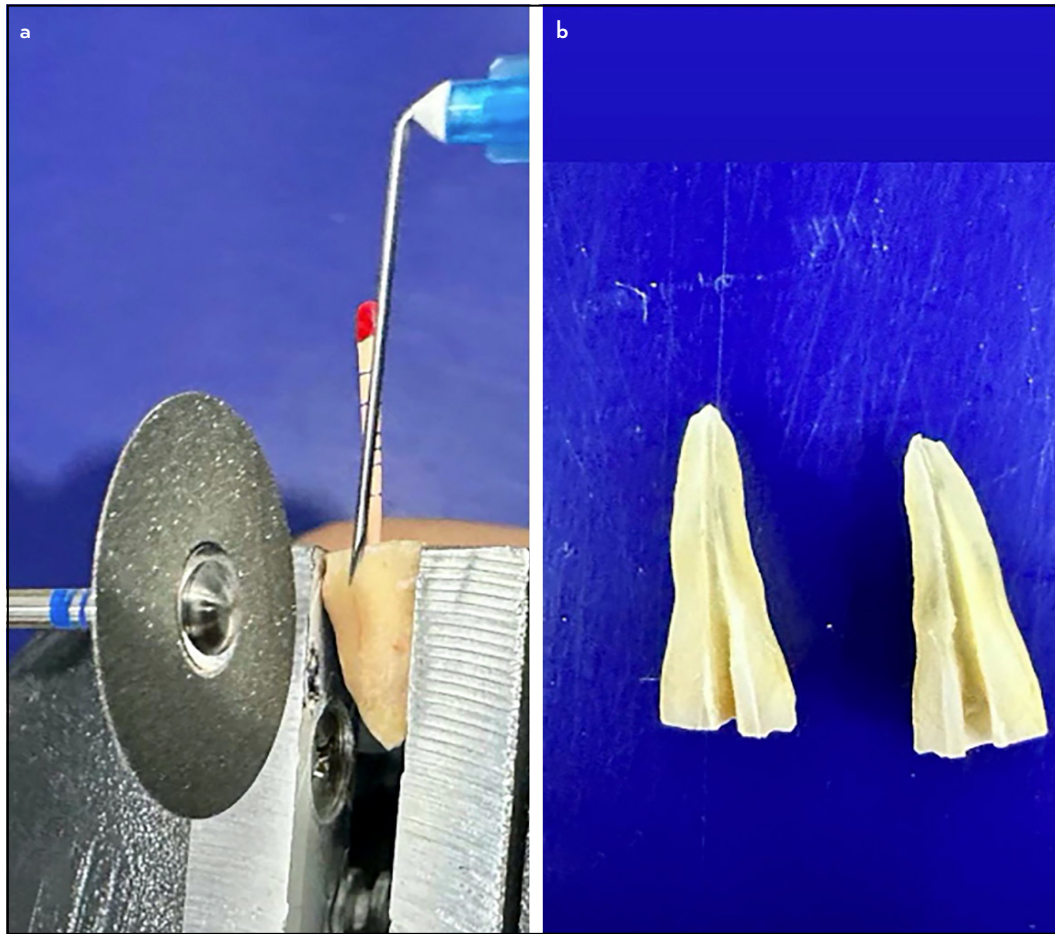
### Sample Selection and Preparation

This study received ethical approval (No. The MUOPR23) by the Ethical-Scientific Committee of the local institution (college of Dentistry, Mustansiriyah University), the study was conducted in accordance with the Declaration of Helsinki.

Forty mandibular premolars from patients aged 25–35 years were collected. The criteria for tooth selection were as follows: all roots should be free from caries, cracks by inspection, while using the radiographic view to confirm if the teeth had previous endodontic treatment, internal or external resorption, calcified canal. The teeth were single straight canals and mature apices. Teeth were collected, cleaned to eliminate soft tissue debris and/or hard attached tissues, and placed in 0.1% thymol solution at 37°C for 24 hours and then stored in normal saline to maintain hydration (16). The samples were fixed using a bench vice and the crowns were cut transversally with the double-faced diamond disc at high speed, along with water coolant, to get 14 mm root length. The working length (WL) was verified using the #10 K file. The file was then inserted into the canal until the tip passed through the foramen. The WL was established 1 mm short of the anatomical apex (13 mm). Roots that permitted instrumentation with size #20 initially was selected. The samples were divided into the following four groups (n=10 each) based on the type of irrigation solution used: Group I, Triton all-in-one irrigant; Group II, 5.25% NaOCl+ 0.5% CNP; Group III, 5.25% NaOCl+ 17% EDTA; and Group IV, Distilled water (control group).

### CNP Preparation and Evaluation

After dissolving 0.5 g CNP powder (EPRUI, China) in 100 ml of 1% (v/v) acetic acid, the mixture was stirred continuously for 8 h. The samples were sonicated for 40 min. Separately, sodium tripolyphosphate (STPP) (0.1 g) was dispersed in 10 ml of distilled water and stirred continuously for 8 h, after sonication for 40 min. Then, using a 50 ml syringe and a drip rate of 15 drops per minute, the STPP solution was added to the Chitosan solution (CS) dropwise until the ratio of CS: STPP reached 2:1. This mixture was mixed for an additional 8 h and sonicated for



**Figure 1.** Splitting of the tooth, (a) Parallel longitudinal grooves on the buccal and lingual aspects of each root were made by using a high-speed diamond bur under water-cooling. (b) A chisel separates the root into two parts longitudinally along the grooves

40 min (17). The size of CS-TPP was identified using dynamic light scattering and a NanoBrook 90Plus Particle Size Analyzer (Brookhaven Instruments, USA). The effective diameters of the CS suspensions were 84.4 nm.

### Root Canal Instrumentation and Irrigation

#### *The irrigation protocol was as follows*

Group I (Triton Brasseler, Savannah, USA): Canals were irrigated as required throughout instrumentation with approximately 5 ml according to the manufacturer's instructions (3–6 ml per root canal treatment). After final irrigation with 1 ml of Triton for 1.5 min, 5 ml distilled water was used for washing upon completion.

Group II (0.5% CNP, EPRUI, China): Canals were irrigated with 1 ml of 5.25% NaOCl for a total volume of approximately 5 ml after every three strokes of the instrument, and then rinsed with 5 ml distilled water, followed by drying with an absorbent paper point (Diadent, Korea) prior to final irrigation. Final irrigation with 5 ml of 0.5% CNP for 3 min was performed followed by irrigation with 5 ml distilled water and drying of the canals with paper point #40 (18).

Group III (17% EDTA, CerKamed, Poland): Canals were irrigated with 1 ml of 5.25% NaOCl at every three strokes of the instrument for a total volume of approximately 5 ml and then rinsed with

5 ml distilled water and dried with absorbent paper point (Diadent, Korea) size #40 prior to final irrigation. Final irrigation was performed using 5 ml of 17% EDTA for 3 min followed by irrigation with 5 ml distilled water and drying by paper point #40 (18).

Group IV (distilled water): Canals were irrigated with 1 ml distilled water at every three strokes of the instrument for a total volume of approximately 5 ml, followed by final irrigation with 5 ml distilled water for 3 min.

The irrigation needle was inserted within 2 mm of the WL of the canal for irrigation (9).

### Root Canal Instrumentation

All the root canals were instrumented using ProTaper Next rotary file (Dentsply Maillefer) X4, (#40/0.06). A 300-rpm electric motor (Dentsply Maillefer, Ballaigues, Switzerland) with 4.0 Ncm torque was used to drive the files to their complete WL according to the manufacturer's recommendations.

### Preparation of Sections for Viewing Under Scanning Electron Microscopy

A high-speed diamond bur was used to create two parallel longitudinal grooves on the buccal and lingual portions of each root

after the samples were removed from the mold (19). The opening was sealed with a tiny cotton plug; gutta-percha was inserted into the canal, which served as a gauge for the groove depth to prevent bur incursion into the canals, resulting in contamination from sectioning-related debris (20). The root was then split half longitudinally along the grooves using a chisel as shown in Figure 1. A scanning electron microscope (FESEM, TESCAN, Mira 3) was used to analyze the selected samples (half section of 180° or less) at three different levels (2.5, 6, and 10 mm from the root apex). Two calibrated and blinded examiners reviewed all the photographs taken at 3000× magnification. The Kappa agreement test was used for the evaluation of the correlation between the two examiners (kappa = 0.75). A scoring system (ranging from 1 to 4) based on the Hülsmann et al. (21) scores was used to determine the amount of the smear layer removal:

Score 1: Completely open dentinal tubules

Score 2: The dentinal tubules are open more than 50%.

Score 3: The dentinal tubules are open to a maximum of 50%.

Score 4: The smear layer almost entirely covers the dentinal tubules.

### Statistical Analysis

SPSS 22.0 (IBM, Armonk, United States) was used for the statistical analysis. To verify that the distribution was normal, the Shapiro-Wilk test was employed. To analyze the data, the Mann-Whitney U test and the Kruskal-Wallis test were used. ( $p \leq 0.05$ ) was chosen as the significance threshold.

### RESULTS

High agreement between the two examiners was found using kappa, with values of 0.8 or greater for each of the different categories as shown in (Table 1).

**TABLE 1.** Kappa test for reliability between two observers at all tested group

Groups	Kappa agreement	Significance level
Group I	0.929	>0.001
Group II	0.938	>0.001
Group III	0.946	>0.001
Group IV	0.902	>0.001
(0.81–0.99) Almost perfect agreement		

The scores of smear layer removal for four groups at three different levels are shown in Table 2.

Mann-Whitney U test was used for pair-wise comparisons between groups. The Triton irrigation solution demonstrated significantly ( $p < 0.05$ ) lower mean smear layer removal than the other experimental groups at the coronal, middle, and apical levels. CNP demonstrated similar efficiency to EDTA as a chelating agent at the coronal and middle levels of the root canal, but was significantly ( $p \leq 0.05$ ) more efficient than EDTA apically (Figs. 2, 3).

### DISCUSSION

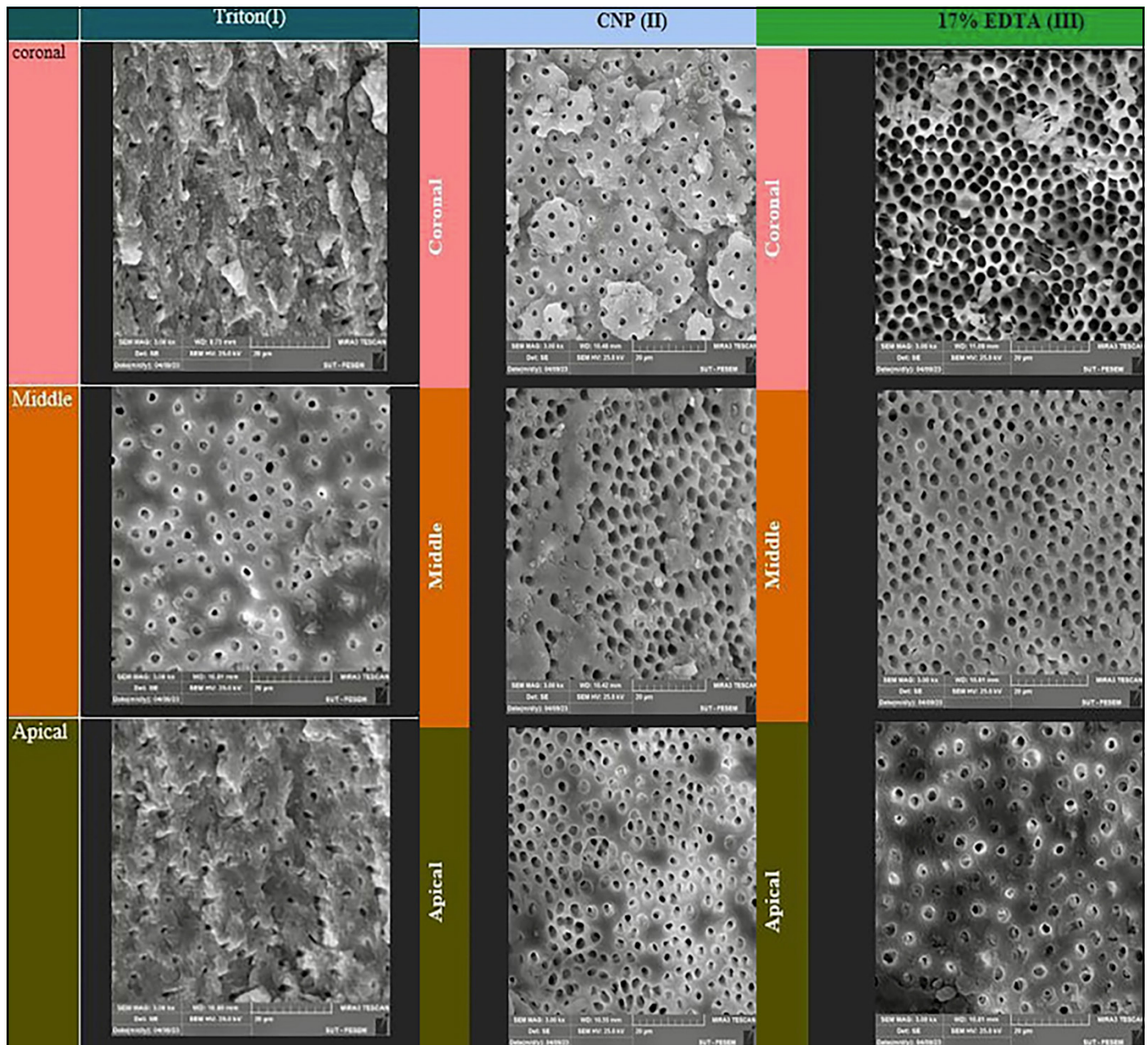
A successful root canal treatment depends critically on the efficient chemico-mechanical preparation of the root canal space (22). However, residual smear layers is usually formed on inner dentinal walls of the root after shaping procedures (23), which may lead to persistent contamination and failure (4). According to the findings of this study, no irrigation solution was able to completely eliminate the smear layer in any of the three root regions. This could be attributed to the complex canal shapes, inability of irrigation solutions to penetrate every surface of the root canal, and limited effect of each irrigation solution (24).

Similar to other solutions, the ability of Triton to remove the smear is attributed to the presence of citric acid (CA) in its composition. When used as a chelator, CA can dissolve inorganic ma-

**TABLE 2.** Kruskal-Wallis Test for the scores of smear layer removal for four groups at three different levels

Site	Groups	MIN	MAX	Median	MR	Kruskal-Wallis	
						Chi-square	p
Coronal	Triton	2	4	3	22.90	17.510	>0.001
	CNP	2	4	2	16.15		
	EDTA	1	3	2	9.95		
	Distilled water	3	4	4	33.00		
	Total	1	4	3			
Middle	Triton	3	4	4	24.40	17.298	>0.001
	CNP	2	3	3	15.40		
	EDTA	1	3	2	9.80		
	Distilled water	3	4	3	32.40		
	Total	1	4	3			
Apical	Triton	3	4	4	26.00	11.541	>0.001
	CNP	1	3	3	8.95		
	EDTA	2	3	3	15.05		
	Distilled water	3	4	4	32.00		
	Total	1	4	3			

MIN: Minimum, MAX: Maximum; MR: Mean rank, CNP: Chitosan nanoparticles, EDTA: Ethylenediaminetetraacetic acid



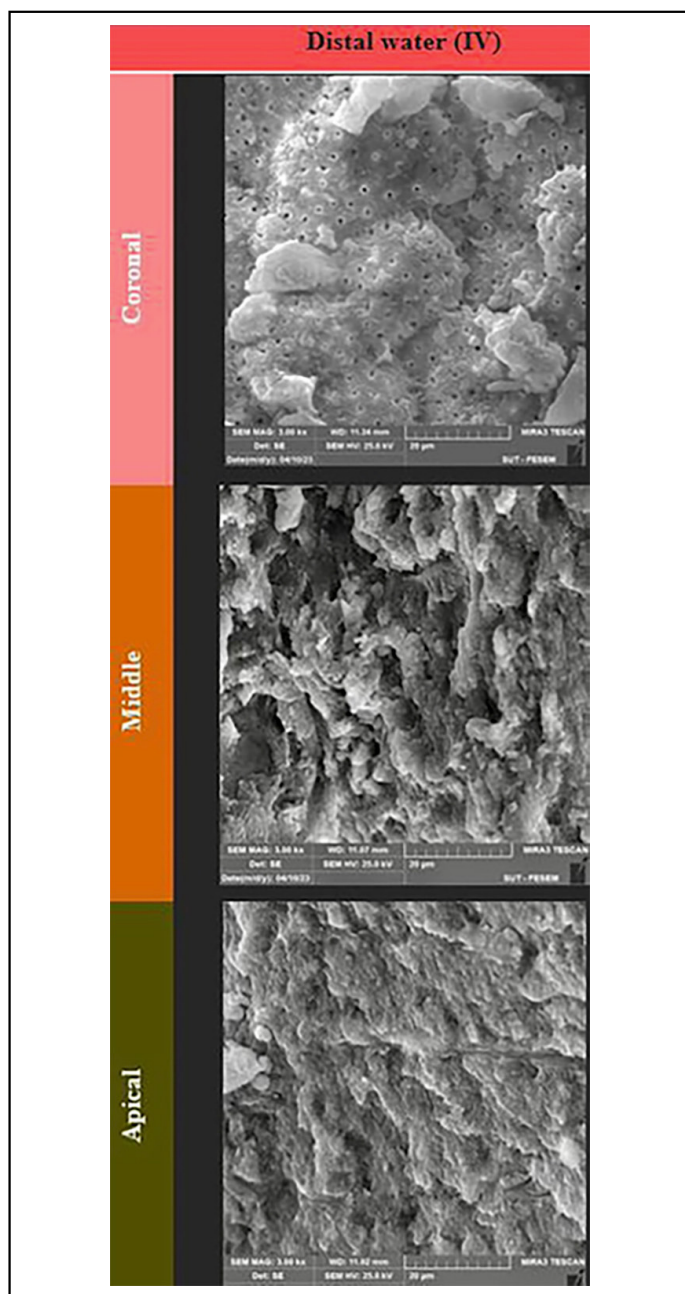
**Figure 2.** Scanning electron microscope photographs at 3000× magnification showing root samples treated with Group I (Triton), Group II (0.5% CNP) and Group III (17% EDTA) at different levels (coronal, middle, and apical) of the root canal

CNP: Chitosan nanoparticles, EDTA: Ethylenediaminetetraacetic acid

terials such as apatite crystals and remnants of dentine (25). CA can provide a synergistic action of mode interacting with other chemicals, according to recent literature (26, 27). Furthermore, stronger proteolytic effects are largely dependent on the stable, high pH condition of NaOCl solutions produced by pH modifiers. Previous research has shown that a significantly greater amount of necrotic, inflammatory tissue, dentine debris, and inorganic smear layer components are likely to dissolve in NaOCl (28, 29). The all-in-one design in Triton showed benefits for the combination of surfactants, pH modifiers, CA, and NaOCl. Triton irrigation showed a significant difference with both groups at the three levels and was less successful in removing smear layers than CNP and EDTA. Therefore, the null hypothesis was rejected because there was a significant difference in smear layer removal

between Triton and other solutions. The results of this study are not consistent with Sheng et al., (30); this may be explained by the time of irrigation used in that study which was 3 min for one or two rounds, while in the current study, the time was limited to 1.5 min, according to the manufacture instruction.

No obvious difference was noted between CNP and 17% EDTA at the coronal and middle levels, which is consistent with previous studies (1, 13, 18). A significant difference was observed in the apical level between the CNP and 17% EDTA groups. The decrease in the root canal diameter at the apical third makes smear layer removal challenging (31). Nano-sized Chitosan particles can accelerate the flow of irrigation solution into the dentinal tubules, thereby increasing smear layer removal



**Figure 3.** Scanning electron microscope photographs at 3000 $\times$  magnification showing root samples treated with Group IV (distilled water) at different levels (coronal, middle, and apical levels) of the root canal

(15). Owing to the hydrophilic nature of the Chitosan polymer, which favours close contact with root canal dentine, it is easily adsorbed to the root canal walls and transported to deep into the dentinal tubules (32). This result is consistent with other studies which found that CNP shows a stronger ability of smear layer removal from the apical area than 17% EDTA and citric acid (18, 33). The lower capacity of EDTA for smear layer removal in the apical third may be explained by the fact that the chelating activity of the neutral EDTA solution is based on the elimination of calcium from both the organic and inorganic components of dentine, such as water-soluble non-collagen proteins, which are present at lower concentrations in the apical area; thus, the amount of EDTA decalcification is reduced

(34). Tubular sclerosis of dentine in the apical third of the root canal may also reduce the effectiveness of EDTA (35, 36).

Within the limitation of this study, the irrigation time of 0.5% CNP and 17% EDTA was 3 min following the irrigation protocol of a previous study (16), while for Triton, it was only 1.5 min. In addition, the combination of NaOCl and EDTA may cause dentine erosion (37) rather than true smear layer removal, and since SEM was used in this study, it was difficult to differentiate between dentinal tubules opened by smear layer removal or by dentine erosion.

## CONCLUSION

Within the limitations of this study, it can be concluded that Triton irrigation solution removes the smear layer less efficiently than 0.5% CNP and 17% EDTA at all the levels of the root canal, while 0.5% CNP removes the smear layer more efficiently at the apical root level than the other tested solutions. However, 17% EDTA was as efficient as 0.5% CNP in smear layer removal at the coronal and middle levels of the root canal.

## Disclosures

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**Conflict of interest:** The authors deny any conflict of interest.

**Ethics Committee Approval:** This study was approved by The College of Dentistry, Mustansiriyah University Scientific Ethics Committee (Date: 10/03/2023, Number: MUOPR23).

**Peer-review:** Externally peer-reviewed.

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**Authorship contributions:** Concept – H.W.O.; Design – H.W.O.; Supervision – IM..A.Z.; Writing – H.W.O.

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