



Efficacy of different irrigation solutions and techniques for removing intracanal medicaments used in regenerative endodontics: An in vitro study

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Purpose: This study assessed the efficacy of various irrigation solutions (EDTA, EDTA with benzalkonium chloride [BAC], ethanol, and glycolic acid) and techniques (conventional needle irrigation, sonic, and ultrasonic systems) for removing calcium hydroxide Ca(OH)₂ and double antibiotic paste (DAP) medicaments.

Methods: Ca(OH)₂ and double DAP were placed in the canals of extracted single rooted human mandibular premolars. The specimens were divided into groups based on the irrigation solutions and techniques (conventional needle irrigation, sonic and ultrasonic activation) used. After removal of the medicaments, residues were evaluated using stereomicroscope. The Mann-Whitney U, the Kruskal-Wallis and by Dunn-Bonferroni post hoc tests were used to analyze the data. The study results at a 95% confidence level.

Results: The lowest residue scores were observed with 17% EDTA and 17% EDTA with BAC, regardless of irrigation techniques for Ca(OH)₂, and with 17% EDTA, 17% EDTA with BAC, and ethanol for DAP (p<0.05). The highest scores were obtained with 10% glycolic acid. The methods of irrigation showed similar effectiveness in each solution group.

Conclusion: EDTA and EDTA + BAC solutions provided the lowest residue scores, confirming EDTA's effectiveness regardless of the irrigation method. While BAC or ethanol may enhance intracanal medication removal, glycolic acid likely caused precipitation, leading to higher scores.

Keywords: Calcium hydroxide; double antibiotic paste; benzalkonium chloride; glycolic acid; ethanol.

Introduction

The process of root development of permanent teeth can be interrupted by decay or trauma resulting in incomplete root tips, thin dentin walls, and shorter root canal length (1). Apexification therapy with calcium hydroxide (Ca(OH)₂) or calcium silicate-based materials has been used for years for the treatment of these teeth. Alternative-

ly, regenerative endodontic treatments (RETs) are emerging as an alternative treatment due to their potential to support the development of root thickness and length (2). With RETs, Ca(OH)₂ or combinations of antibiotics are used as intracanal medicaments for the disinfection of root canal. In recent years, double antibiotic paste (DAP) is now preferred over triple antibiotic paste (TAP) for RETs

Cite this article as: İlgen S, Aslantaş EE. Efficacy of different irrigation solutions and techniques for removing intracanal medicaments used in regenerative endodontics: An in vitro study. Turk Endod J 2026;11:45-53.

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Submitted: March 19, 2025 Revised: September 01, 2025 Accepted: September 11, 2025 Published: April 27, 2026

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because of its reduced cytotoxicity and lower potential for tooth discoloration (3). Studies highlight that DAP is equally effective in eradicating bacterial biofilms while minimizing adverse effects (4). However, a common issue with applied intracanal medicaments is that they cannot be entirely removed from the root canal walls. Ethylenediaminetetraacetic acid (EDTA) is the most commonly used final irrigation solution in endodontics to dissolve inorganic tissues and remove intracanal medicaments. Also, EDTA significantly improves the efficacy of RETs by inducing the release of growth factors from the dentin matrix (5). For improved penetration depth of irrigation solutions, increased dentin permeability, and enhanced cleaning efficiency of canal walls, surfactants are added to irrigation solutions (6,7). One such surfactant, benzalkonium chloride (BAC), is a common cationic detergent used in ophthalmology to prevent contamination of eye solutions (8). In dentistry, BAC provides antimicrobial properties in dentin bonding agents and orthodontic composite resins (9,10). As such, the addition of a surfactant to EDTA is expected to benefit cell attachment.

In addition, studies investigating the use of alcohol solutions have shown that final irrigation with ethanol increases the wettability of root dentin by reducing surface tension and enhances $\text{Ca}(\text{OH})_2$ removal from the apical third (11,12).

Glycolic acid (GA), also known as hydroxyethanoic acid or hydroxyacetic acid, is a colorless, odorless, and water-soluble substance that can be used for acid etching of enamel and dentin (13). GA has been reported to eliminate Gram positive and Gram negative bacteria and is used in the production of some antibiotics (14). Acid can eliminate free radicals and stimulate fibroblast production by inducing the formation of collagen fibers (15). Additionally, the fact that GA is more compatible with the environment and biodegradable could make it a viable solution for the final irrigation of root canals.

Improving the effects of irrigation solutions with physical methods and enhancing the level of disinfection of root canals has been a focus of interest. Irrigation with conventional needles (CNI) is a common and simple clinical application. However, this practice can also lead to problems given its lower degree of disinfection due to the vapor lock effect or if the needle tip is not inserted deep enough in the root canals.

The EndoActivator (Dentsply Tulsa, OK, USA) device generates sonic energy for irrigation of root canal systems. The sonic energy generated can cause a swirling movement of the solution in the pulp chamber and root canals, resulting in strong solution movement. During passive ultrasonic irrigation (PUI), acoustic flow and cavitation occur allowing the solution to flow from the apical to coronal

direction within the root canal. These activation methods are applied to help improve the penetration and flow of irrigation into hard-to-reach areas of the root canal (16).

In a comprehensive literature review, few studies were found that compare the removal of DAP and $\text{Ca}(\text{OH})_2$ paste that are used in RETs with various final irrigation solutions (eg, 17% EDTA, 17% EDTA with 0.008% BAC, 10% GA, and 70% ethanol) and irrigation methods (eg, CNI, sonic activation [SA], and PUI). Therefore, the aim of this study was to compare the effectiveness of various irrigation solutions and methods for DAP and $\text{Ca}(\text{OH})_2$ paste removal in RETs. The null hypothesis of the study was that there would be no difference between the removal of DAP and $\text{Ca}(\text{OH})_2$ paste between irrigation solutions and methods.

Materials and Methods

This study was approved by Hacettepe University's ethics committee (No: GO 22/247/2022/05-05, Date: 05/05/2022) and conducted according to Declaration of Helsinki. The study used a total of 252 single-rooted maxillary central and lateral incisor teeth from individuals aged 18 to 65 years that were extracted due to periodontal disease. Radiographs were taken from various angles for all teeth to confirm the presence of single canals. The teeth were examined under magnification using an operating microscope (Zeiss S7, Carl Zeiss, Germany) and those exhibiting caries, resorption, cracks, root fractures, or previous root canal treatment were excluded. Following radiographic and microscopic evaluations, teeth with a similar root morphology were selected. Soft tissue debris on the teeth was removed with a brush and calculus was removed mechanically with a periodontal scaler.

All teeth were decoronized at the cemento-enamel junction. Root canals were shaped with a ProTaper Universal rotary system (Dentsply-Maillefer, Ballaigues, Switzerland) using up to a F5 file at the specified working length. Between each file, 2 mL of 5.25% sodium hypochlorite (NaOCl) was used for irrigation of the root canals. For the final irrigation, 2 mL of 5.25% NaOCl followed by 2 mL of 17% EDTA was used. The root canals were then dried with paper points. To create an immature tooth form and to remove the apical deltas, the apical 3 mm of the specimens were removed with diamond discs and standardized to an average root length of 12 ± 1 mm. To evaluate the canals after medicament removal, two longitudinal grooves were prepared on the buccal and lingual surfaces of each root using diamond discs ensuring not to damage the inner dentin layer of the root canal (17). The specimens were stored in saline to prevent dehydration before applying intracanal medicaments.

First, specimens were randomly assigned into two main groups: DAP or $\text{Ca}(\text{OH})_2$ paste treated groups and then subsequently split into four subgroups for which the final irrigation solutions were to be applied. To evaluate the efficacy of irrigation methods, the subgroups were further divided into three groups, resulting in a total of 24 study groups ($n=10$ teeth per group). In addition, three positive and three negative control samples were prepared for DAP and $\text{Ca}(\text{OH})_2$ groups.

The $\text{Ca}(\text{OH})_2$ paste was made by mixing powder and glycerin-containing liquid (Kalsin, Aktu Tic., İzmir, Türkiye) according to the manufacturer's recommendation. For the preparation of the DAP, equal amounts of metronidazole (Flagyl 500 mg, Sanofi, Lüleburgaz-Kırklareli, Türkiye) and ciprofloxacin (Cipro 500 mg, Biofarma, İstanbul, Türkiye) were ground in a mortar and then mixed 1:1 with glycerin-containing liquid (Kalsin). $\text{Ca}(\text{OH})_2$ and DAP was applied to the root canals with #40 Lentulo spirals until the extrusion of paste was observed apically. Six untreated teeth were used as the negative control for DAP and $\text{Ca}(\text{OH})_2$ groups. The apical root of the teeth was sealed with dental wax and the coronal part temporarily sealed with cotton pellets and Cavit (3M ESPE, St. Paul, MN, USA). The specimens were placed in a silicone impression material (President Dental, Allerhausen, Germany) and incubated in Eppendorf tubes for 2 weeks at 37°C and 100% humidity.

Commercially purchased 17% EDTA (Werax, İzmir, Türkiye) was used. 17% EDTA (Werax, İzmir, Türkiye) with 0.008% BAC (Dermosept Zefiran, İstanbul, Türkiye), 10% GA (Smart Kimya, İzmir, Türkiye), and 70% ethanol were freshly prepared immediately before the removal procedures.

For CNI, 27-gauge (ISO 0.41 mm) needles (Genject, Ankara, Türkiye) were used. With irrigation needles placed in

the teeth 2 mm shorter than the working length, 2.5 mL of solution was applied to the root canal for 30 seconds with up-and-down movements. This procedure was repeated eight times for each specimen, using a total 20 mL of solution per specimen.

The large (Blue, 35/.04) tip of the EndoActivator (Dentsply Tulsa, OK, USA) was used for SA. After each 5 mL of solution was applied to the root canals, the device was moved up and down in the root canal for 30 seconds and repeated four times. For PUI, a single 20/.02 ultrasonic tip of EndoUltra (Vista Dental Products, Wisconsin, USA) was used. The series in the SA group was repeated ensuring to avoid contact of the ultrasonic tip with the walls. In the SA and PUI groups, 20 mL of solution was used for each sample, similar to the CNI group. The total solution activation time for each sample in the activation groups was 2 minutes. The placement and removal of medicaments was performed by a single researcher (S.I.). The evaluation of the samples was performed independently by two researchers (S.I. and E.E.A.). In cases of disagreement, the same sample was re-evaluated by both researchers and a consensus was agreed.

After removal of the medicament, the root canals were dried with paper cones. The roots were split in two with a hammer applied to thin cement spatula positions in the notches. Two distinct specimens were obtained from each tooth. Both samples were scrutinized using a stereomicroscope and the fragment with a higher residue was scored. The specimens were examined under 25x magnification using an Olympus SZ61 stereomicroscope (Olympus, Tokyo, Japan) and imaged using an Olympus DP12 camera (Olympus, Tokyo, Japan) attached to the microscope. Two observers independently evaluated and scored the stereomicroscope images of the teeth three times at 1-week intervals. Scoring was performed according to the criteria defined by Aksel et al. (18) (Fig. 1). Interobserver agreement

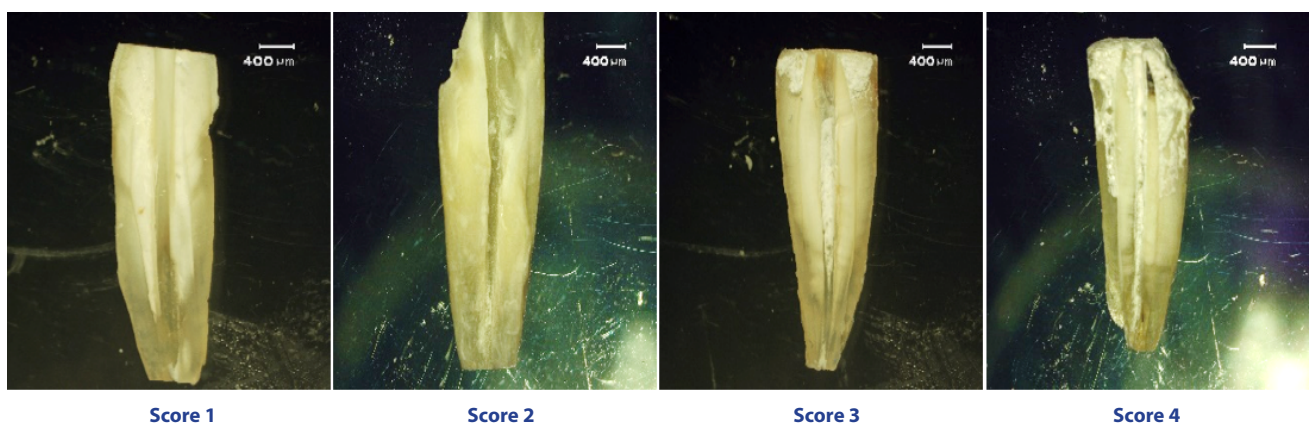


Fig. 1. Representative examples of medicament removal scoring and scoring criteria of samples.

was determined by Cohen's kappa test.

Data analyses were conducted with IBM SPSS Statistics v26 software. Comparisons between two independent groups were assessed by the Mann–Whitney U Test, while comparisons involving more than two independent groups were performed using the Kruskal–Wallis test. In cases where a significant difference was found among the groups following the Kruskal–Wallis test, the Dunn–Bonferroni multiple comparison test was used to identify the specific groups contributing to the difference. A 95% confidence interval ($p=0.05$) was used for all tests.

Results

Ca(OH)₂ removal

All irrigation methods in the EDTA group and SA and PUI in the 17% EDTA + 0.008% BAC group, were significantly more effective in removing Ca(OH)₂ compared with the positive control group ($p<0.05$). However, no significant differences were found between irrigation methods within the same groups ($p>0.05$; Table 1). The GA group showed the least Ca(OH)₂ removal compared with the negative control group ($p<0.05$). None of the

Table 1. Scoring results of calcium hydroxide paste groups

Solution	Irrigation Method	Median	Percentile 25 (Q1)	Percentile 75 (Q3)	Mean
17% EDTA	CNI ^{h,y}	1.50	1.00	2.00	1.50
	SA ^{h,x}	1.00	1.00	2.00	1.40
	PUI ^z	2.00	1.00	2.00	1.70
17% EDTA + 0.008% BAC	CNI	2.00	2.00	2.00	2.20
	SA ^{d,f}	1.00	1.00	1.00	1.00
	PUI ^{e,g}	1.00	1.00	1.00	1.10
70% Ethanol	CNI	2.00	2.00	2.00	2.00
	SA	2.00	2.00	3.00	2.30
	PUI	2.00	2.00	2.00	2.10
10% Glycolic Acid	CNI ^{a,i}	4.00	4.00	4.00	3.80
	SA ^{b,f,h}	4.00	3.00	4.00	3.60
	PUI ^{c,g,i}	4.00	4.00	4.00	3.80
Negative Control ^{a,b,c,j}		1.00	1.00	1.00	1.00
Positive Control ^{d,e,i,x,y,z}		4.00	4.00	4.00	4.00

BAC: Benzalkonium Chloride; CNI: Conventional Needle Irrigation; PUI: Passive Ultrasonic Irrigation; SA: Sonic Activation. *Same letters indicate a significant difference between the groups ($p<0.05$).

Table 2. Scoring results of the double antibiotic paste experimental groups

Solution	Irrigation Method	Median	Percentile 25 (Q1)	Percentile 75 (Q3)	Mean
17% EDTA	CNI ^{h,y}	1.50	1.00	2.00	1.50
	CNI	2.00	2.00	2.00	2.10
	SA ^{o,u}	1.00	1.00	2.00	1.40
17% EDTA + 0.008% BAC	PUI ^o	1.50	1.00	2.00	1.50
	CNI ^{m,u}	2.00	1.00	2.00	1.60
	SA ^r	2.00	1.00	2.00	1.80
	PUI ^t	1.00	1.00	2.00	1.30
70% Ethanol	CNI	2.00	2.00	3.00	2.30
	SA ^s	2.00	2.00	2.00	1.90
	PUI ^{n,s}	1.00	1.00	2.00	1.30
10% Glycolic Acid	CNI ^{k,u}	4.00	4.00	4.00	3.80
	SA ^{l,u}	3.00	3.00	4.00	3.30
	PUI ^{m,s,t}	3.00	3.00	3.00	3.20
Negative Control ^{k,l,m}		1.00	1.00	1.00	1.00
Positive Control ^{n,o,u,p,r,s}		4.00	4.00	4.00	4.00

BAC: Benzalkonium Chloride; CNI: Conventional Needle Irrigation; PUI: Passive Ultrasonic Irrigation; SA: Sonic Activation. *Same letters indicate a significant difference between the groups ($p<0.05$).

Table 3. Comparative evaluation of the CNI group for both intracanal medicaments

	Median	Percentile 25 (Q1)	Percentile 75 (Q3)	Mean	P
CNI + EDTA					
Ca(OH) ₂	1.50	1.00	2.00	1.50	0.035*
DAP	2.00	2.00	2.00	2.10	
CNI + EDTA^BAC					
Ca(OH) ₂	2.00	2.00	2.00	2.20	0.52
DAP	2.00	1.00	2.00	1.60	
CNI + Ethanol					
Ca(OH) ₂	2.00	2.00	2.00	2.00	0.28
DAP	2.00	2.00	3.00	2.30	
CNI + GA					
Ca(OH) ₂	4.00	4.00	4.00	3.80	1.00
DAP	4.00	4.00	4.00	3.80	

BAC: Benzalkonium Chloride; GA: Glycolic Acid; CNI: Conventional Needle Irrigation. *The marked value indicates a statistically significant difference ($p < 0.05$).

irrigation methods in the GA group could effectively remove Ca(OH)₂ ($p > 0.05$; Table 1).

DAP removal

Similar to the Ca(OH)₂ group, no significant differences were found between activation methods for the same irrigation solutions ($p > 0.05$). SA and PUI used with 17% EDTA significantly improved canal cleanliness compared with the positive control group ($p < 0.05$). SA and PUI with 17% EDTA + 0.008% BAC were also highly effective in removing antibiotic paste ($p < 0.05$; Table 2). Additionally, SA and PUI with 70% ethanol showed significantly better removal of DAP compared with the positive control group ($p < 0.05$; Table 2).

Comparative data analysis

The Mann–Whitney U test was used for pairwise compari-

sons of Ca(OH)₂ and DAP removal scores (Tables 4–6). With the CNI method, EDTA was significantly more effective in removing Ca(OH)₂ than DAP ($p < 0.05$; Table 3). Using the SA method, Ca(OH)₂ was better removed when combined with EDTA + BAC ($p < 0.05$; Table 4). PUI combined with ethanol resulted in significantly more DAP removal compared to the control ($p < 0.05$; Table 5).

Discussion

In this study, maxillary central and lateral incisors from individuals aged 18–65 years were included. Due to the required sample size, full homogeneity could not be achieved; however, teeth with similar root canal morphology were selected based on radiographic and microscopic evaluation. As the study focused solely on the mechanical removal of medicaments—not their dentinal tubule pen-

Table 4. Comparative evaluation of the SA group for both intracanal medicaments

	Median	Percentile 25 (Q1)	Percentile 75 (Q3)	Mean	P
SA + EDTA					
Ca(OH) ₂	1.00	1.00	2.00	1.40	1.00
DAP	1.00	1.00	2.00	1.40	
SA + EDTA^BAC					
Ca(OH) ₂	1.00	1.00	1.00	1.00	0.007*
DAP	2.00	1.00	2.00	1.80	
SA + Ethanol					
Ca(OH) ₂	2.00	2.00	3.00	2.30	0.165
DAP	2.00	2.00	2.00	1.90	
SA + GA					
Ca(OH) ₂	4.00	3.00	4.00	3.60	0.28
DAP	3.00	3.00	4.00	3.30	

BAC: Benzalkonium Chloride; GA: Glycolic Acid; SA: Sonic Activation. *The marked value indicates a statistically significant difference ($p < 0.05$).

Table 5. Comparative evaluation of the PUI group for both intracanal medicaments

	Median	Percentile 25 (Q1)	Percentile 75 (Q3)	Mean	P
PUI + EDTA					
Ca(OH) ₂	2.00	1.00	2.00	1.70	0.481
DAP	1.50	1.00	2.00	1.50	
PUI + EDTA^BAC					
Ca(OH) ₂	1.00	1.00	1.00	1.10	0.481
DAP	1.00	1.00	2.00	1.30	
PUI + Ethanol					
Ca(OH) ₂	2.00	2.00	2.00	2.10	0.004*
DAP	1.00	1.00	2.00	1.30	
PUI + GA					
Ca(OH) ₂	4.00	4.00	4.00	3.80	0.023*
DAP	3.00	3.00	3.00	3.20	

BAC: Benzalkonium Chloride; GA, Glycolic Acid; PUI: Passive Ultrasonic Activation. *The marked value indicates a statistically significant difference (p<0.05).

etration—this limitation can be considered as negligible. None of the intracanal medicaments could be completely removed from the root canals with the tested irrigation solutions and techniques. Remnants of intracanal medicaments might also affect the properties of root canal sealer, such as hardening, adaptation, and penetration. However, with RETs, antibiotic paste residues have been shown to have a detrimental effect on stem cells in the apical papilla (19). Conversely, the positive or negative effect of Ca(OH)₂ paste residues on the bonding of tricalcium silicate-based materials to root canal dentin, which remains unclear in the literature. Some studies have shown that the detachment resistance of calcium silicate-based materials bonded to Ca(OH)₂-treated dentin is affected and even increased (20). However, these results have not yet been confirmed in the oral environment; therefore, the necessity for effective removal of intracanal medicaments from root canals remains until further studies are conducted.

In endodontics, there is increasing interest in the use of surfactants and changing the wetting properties by adding surfactants to solutions. As such, there are various studies in which BAC is used in combination with NaOCl (21-23). Bukiet et al. (23) reported that 0.008% BAC added to 2.4% NaOCl solution did not change the amount of free chlorine in NaOCl and also did not affect its cytotoxicity. Eren et al. (24) evaluated the effects of BAC when added to 17% EDTA solution at varying concentrations (0.008% and 0.1%) on the release of TGF-β1 and the level of dentin binding and proliferation of dental pulp stem cells (DPSCs). Both EDTA solutions with and without BAC increased DPSC binding and proliferation (p>0.05), while TGF-β1 release did not show a significant difference compared with the control group. There are few studies on the efficacy of BAC when added to 17% EDTA solu-

tion to remove intracanal medicaments; therefore, in our study, 0.008% BAC was added to 17% EDTA.

Given its low surface tension, the use of ethanol solution and its effects has been investigated in endodontics. Recent studies show that 70% ethanol does not change the inorganic content of dentin after removal of Ca(OH)₂ and can additionally increase the wetting properties of solutions by increasing surface energy (11). Dias-Junior et al. (12) showed that irrigation with 70% ethanol was significantly more effective than 2.5% NaOCl and 17% EDTA-T (17% EDTA + 0.2% lauryl sodium sulfate) in removing Ca(OH)₂ from root canals. On the other hand, Arslan et al. (25) reported that ethanol caused more TAP residues in root canals compared with 1% NaOCl combined with PUI. However, no study on the efficacy of ethanol in removing DAP exists. Therefore, 70% ethanol was included in our study to evaluate its effectiveness in removing intracanal medicaments.

GA is a colorless, odorless, and water-soluble substance. Due to its acidic nature, its effects on the removal of debris and intracanal medicaments have been investigated. The effectiveness of GA in removing the smear layer from the canal walls after root canal preparation was reportedly similar to EDTA and citric acid. Since GA, which was shown to be less cytotoxic than EDTA by Bello et al. (26), is a biodegradable acid, its use as an irrigation solution could be promising. In previous studies, GA has been tested at many different concentrations (27,28). However, since its negative effects on the apatite to collagen ratio and dentin microhardness have been shown at increasing concentrations (29), it was used at a concentration of 10% in our study.

When the mean results of both intracanal medicaments groups were analyzed, EDTA and EDTA with BAC solu-

tion groups had lower scores for removal. Studies comparing 17% EDTA solution with different final irrigation solutions for the removal of $\text{Ca}(\text{OH})_2$ and antibiotic paste from root canals confirm the strong efficacy of the solution. Salgado et al. (30), reported that the combined use of NaOCl and EDTA-T had higher efficacy for the removal of $\text{Ca}(\text{OH})_2$ paste from root canals compared with citric acid and NaOCl alone. Üstün et al. (31), compared the efficacy of 17% EDTA solution for the removal of TAP from root canals with peracetic acid and etidronic acid and reported the superior efficacy of 17% EDTA over the other solutions. In the literature, there is no study in which BAC was added to EDTA for the removal of intracanal medicaments. In our study, the SA and PUI subgroups of the EDTA with BAC irrigation group showed a similar efficacy to the EDTA only solution group, but produced lower residual scores, especially for the removal of $\text{Ca}(\text{OH})_2$ paste ($p>0.05$). These results could be explained by the fact that BAC might have improved the wetting properties of the solution.

In our study, 70% ethanol showed an effective removal efficiency in the SA and PUI subgroups of the DAP group compared with the positive control group ($p<0.05$), which was not observed for $\text{Ca}(\text{OH})_2$ ($p>0.05$). Arslan et al. (25) compared 17% EDTA with 100% ethanol for the removal of TAP and showed that the removal efficiency of both solutions was similar ($p>0.05$). Dias-Junior et al. (12) compared the efficacy of 70% ethanol and 17% EDTA-T for the removal of $\text{Ca}(\text{OH})_2$ and showed that 70% ethanol produced significantly cleaner root canal walls ($p<0.05$). These results could be explained by the low surface tension of ethanol, which allows it to clean deeper areas of the dentinal tubules compared with other solutions.

Conversely, the ethanol + PUI subgroup in our study showed an effective removal efficiency compared with the GA + PUI subgroup ($p<0.05$). 10% GA showed the lowest removal efficiency, causing the highest scores in both medicament groups. Altıntaş et al. (32) compared the effectiveness of 17% EDTA, 10% GA, and 70% ethanol for the removal of $\text{Ca}(\text{OH})_2$ that was applied to root canals. Similar to our study results, 10% GA had the highest residual scores. On the other hand, Keskin et al. (33) removed $\text{Ca}(\text{OH})_2$ paste placed in simulated resorption cavities in extracted teeth with 5%, 10% GA, 17% EDTA, and 10% citric acid solutions. Similar to our study, CNI and PUI were used as irrigation techniques. In the group activated with PUI, 10% GA removed more $\text{Ca}(\text{OH})_2$ compared with other irrigation solutions ($p<0.05$). In the CNI group, the level of $\text{Ca}(\text{OH})_2$ removal was not affected by the different solutions used ($p>0.05$). The positive results obtained with GA solution in the mentioned study are

not consistent with our study, which might be due to the small size of the simulated cavities and final rinsing with distilled water. Observationally, after irrigation with GA, a salt-like precipitate formed within and around the canals. This precipitate dissolved completely upon rinsing with distilled water after the experiment. It is possible that this precipitate was a salt formed as a reaction between GA and $\text{Ca}(\text{OH})_2$ and DAP. Therefore, irrigation with distilled water following the use of GA is recommended.

Each of the solution groups in our study were evaluated individually and the efficacies of CNI and activation methods (SA and PUI) were similar ($p>0.05$). In the literature, studies comparing needle irrigation or other activation methods show varied results. Some studies highlight the superiority of PUI over the EndoActivator (34,35), while others suggest that these methods have similar efficacy for the removal of intracanal medicaments (36,37). However, a number of studies have reported the superiority of EndoActivator and PUI over CNI (38,39). Sarıçam et al. (40) compared different irrigation needle tips and the effectiveness of EndoActivator in their study where 17% EDTA solution was used as irrigation solution for the removal of antibiotic paste. According to micro-CT data, there was no significant difference between the effectiveness of needle irrigation and EndoActivator ($p>0.05$). However, Lloyd et al. (41) compared the effectiveness of the EndoUltra PUI device with CNI for the removal of $\text{Ca}(\text{OH})_2$ paste that was applied to the mesial canals of mandibular molars. However, in our study, tips with different diameters were used in sonic and ultrasonic activation. A 35/04 tip was preferred for the EndoActivator due to its adaptation to the wide canals created in the samples. It is reported that during PUI, an ultrasonic tip freely oscillating within the root canal has a greater effect than a tip that contacts the canal wall (42). Additionally, EndoUltra has a single type of tip, which is sized 20/02. Although its smaller tip diameter compared with the EndoActivator was expected to negatively affect medicament removal, our results did not indicate inferior performance in terms of removal efficiency. The tip diameter alone might not determine the success of irrigation; other factors, such as activation, also appear to play a considerable role. Nevertheless, this issue remains one of the limitations of the study.

Conclusion

In our study, EDTA and EDTA + BAC solutions provided the lowest residue scores, highlighting EDTA's effectiveness for the removal of $\text{Ca}(\text{OH})_2$ paste from root canals regardless of the irrigation method. While BAC or ethanol can enhance intracanal medication removal, GA likely caused precipitation, leading to higher scores.

Acknowledgements: This research was presented as an poster in FDI World Dental Federation World Dental Congress 12-15 September 2024 İstanbul, Türkiye.

Authorship Contributions: Concept: S.I., E.E.A.; Design: S.I., E.E.A.; Supervision: E.E.A.; Fundings: S.I., E.E.A.; Materials: S.I.; Data: S.I., E.E.A.; Analysis: S.I.; Literature Search: S.I., E.E.A.; Writing: S.I.; Critical Revision: E.E.A.

Use of AI for Writing Assistance: Not declared

Source of Funding: None declared.

Conflict of Interest: None declared.

Ethical Approval: This study was approved by Hacettepe University's ethics committee (No: GO 22/247/2022/05-05, Date: 05.05.2022)

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