The effects of different irrigation protocols on removing calcium hydroxide from the root canals

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Abstract

Objective: The aim of this study was to evaluate the efficiencies of different irrigation protocols and solutions in the removal of calcium hydroxide ($Ca[OH]_2$).

Materials and Methods: Sixty-eight maxillary incisors were used. Root canals were prepared and filled with $Ca(OH)_2$. Two control (n = 4) and six experimental groups (n = 10) were adjusted: Group 1:1% peracetic acid (PAA) + master apical file (MAF); Group 2: 17% ethylenediaminetetraacetic acid (EDTA) + MAF; Group 3: 9% 1-hydroxyethylidene-1,1-bisphosphonate (HEBP) + MAF; Group 4: 1% PAA + ultrasonic activation (UA); Group 5: 17% EDTA + UA; Group 6: 9% HEBP + UA. The cleanliness of root canal thirds were evaluated with scanning electron microscopy. Statistical analysis were performed (α = 0.05).

Results: At coronal thirds; PAA + UA was superior to EDTA + MAF, HEBP + MAF; and PAA + MAF was superior to EDTA + MAF, HEBP + MAF ($P < 0.05$). At middle thirds; PAA + MAF and PAA + UA were superior to EDTA + MAF and EDTA + UA; and, PAA + UA was superior to HEBP + MAF ($P < 0.05$). There were no significant differences among the rest of the experimental groups ($P > 0.05$).

Conclusion: Complete removal of $Ca(OH)_2$ could not be achieved by none of the irrigants at all root thirds.

Key words: Calcium hydroxide removal, irrigation solutions, ultrasonic activation

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Introduction

Calcium hydroxide ($Ca[OH]_2$) is the most popular intracanal medicament used to achieve disinfection of root canals and many other goals such as healing periapical inflammation, arresting inflammatory root resorption, and preventing the reinfection of the root canal system throughout interappointment periods.[1-3] Several studies have reported that the presence of $Ca(OH)_2$ on dentine walls can affect the penetration of sealers into the dentinal tubules.[4,5] Furthermore, there is a consensus that $Ca(OH)_2$ must be removed before the obturation of root canal; however, its complete removal from the root canal remains a problem today.[6-9]

Due to its chelating and antimicrobial properties, peracetic acid (PAA) is suitable for final irrigation of root canals.[10] PAA consists of peroxygen and acetic acid. Peroxygen is responsible for sporicidal, bactericidal, virucidal, and fungicidal activities at low concentrations of <0.5%, even in the presence

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of protein;[11,12] and acetic acid bonds to calcium to form complexes that are easily soluble in water. It has been reported that a 2.25% PAA solution removes the smear layer, which is comparable to that of 17% ethylenediaminetetraacetic acid (EDTA) after 1% sodium hypochlorite (NaOCl).[12] In addition, Sagsen *et al.*[9] reported that a 1% of PAA solution has better efficacy than a 17% EDTA solution in the removal of $Ca(OH)_2$. Nevertheless, recommendations are to use PAA solutions in concentrations lower than 2.25% because of its caustic effects on oral mucosa.[13]

Etidronic acid (also known as 1-hydroxyethylidene-1, 1-bisphosphonate [HEBP] or etidronate) is a weak, biocompatible chelator that can be used in combination with NaOCl without affecting its proteolytic or antimicrobial properties.[14-16] This combination keeps the hypochlorite-hypochlorous acid equilibrium towards hypochlorite, which has better tissue dissolution capability than hypochlorous acid and also has less cytotoxicity.[17,18] It is also reported that this combination improved adhesion of the root fillings performed with an epoxy resin-based sealer and gutta-percha.[19,20]

Several techniques have been used to remove $Ca(OH)_2$ from root canals.[8,21-24] The passive ultrasonic irrigation (PUI) technique was been shown to be more effective in dentine debris removal from the root canal than the delivery of the irrigant using a syringe.[25] PUI works on the principle of placing a small file at the center of a previously shaped root canal and activating it to produce acoustic streaming and cavitation.[26,27] It was reported that PUI was more effective in removing $Ca(OH)_2$ paste from root canal walls than syringe delivery of irrigants.[23,24,28] Furthermore, agitation with the master apical file (MAF) was found to be more effective than irrigant-only techniques the procedure for removing $Ca(OH)_2$ from the root canal.[23]

The aim of the present study was to compare the $Ca(OH)_2$ intracanal medicament removal capability of EDTA, HEBP + NaOCl, and PAA solutions using either PUI or manual agitation with an MAF. No previous study has evaluated the efficiency of HEBP + NaOCl as an irrigant to remove $Ca(OH)_2$ medicament from root canals. The null hypothesis was that there was no significant difference among the irrigation protocols in of the removal of $Ca(OH)_2$ from root canals.

Materials and Methods

Approval of the Ethical Board of the Research Foundation of Erciyes University of Medical Sciences in Kayseri, Turkey, was obtained to conduct this investigation (ethics approval number 341).

Sixty-eight extracted human maxillary central incisor teeth with straight root, single canal, and mature apices were used in this study. After extraction, the teeth were stored for 10 min in 5.25% (NaOCl) at room temperature to facilitate the removal of organic tissue residue. Subsequently, they were scaled with ultrasonic instruments, washed with distilled water, and immersed in a sodium azide solution until required. The crowns of the teeth were removed 12 mm from the apex to standardize the length of the roots. The working length (WL) was set 1 mm short of the apical foramen and root canals were prepared by the same operator using the Mtwo Ni-Ti Rotary System (VDW Dental, Munich, Germany) up to 40.06 instruments as the MAF according to manufacturer's instructions. Between each instrument, each canal was irrigated manually with 3 mL of 2% NaOCl using a 30 gauge needle tip syringe (NaviTip; Ultradent, South Jordan, UT). Following preparations, the root canals were irrigated with a final sequence of 5 mL of 2% NaOCl, 5 mL of 17% EDTA and 10 mL of physiological saline solution and dried with paper points. $Ca(OH)_2$ powder (Merck, Darmstadt, Germany) and distilled water at a powder-to-liquid ratio of 1:1.5 were mixed, and the canals were filled with $Ca(OH)_2$ paste using a lentulo spiral at low speed at the WL. Barium sulfate was added to the distilled water solution for radiopacity. The access cavities were sealed with a temporary filling material (Cavit; 3M ESPE, St Paul, MN, USA). Two radiographs were taken from the buccolingual and mesiodistal sides of each sample to confirm complete filling of the canals. The teeth were kept for 1-week at 37°C and 100% relative humidity to simulate the clinical situation. The specimens were randomly divided into negative control ($n = 4$), positive control ($n = 4$), and 6 experimental ($n = 10$) groups, used for removal of $Ca(OH)_2$ residue according to final irrigating protocols. In the negative control group ($n = 4$), the teeth were not filled with $Ca(OH)_2$. In the positive control group ($n = 4$), the teeth were filled with $Ca(OH)_2$, but no removal procedure was applied.

The treatment for each experimental group was as follows:

Group 1

The root canals were flushed using 3 mL of 1% PAA and agitated with an MAF. The rubber stop of the file was adjusted to 1 mm shorter than the WL and fixed with a flowable composite (Filtek Supreme XT Flowable; 3M ESPE, ST. Paul, MN, USA) and used in the root canal with approximately 2 mm amplitude (measured with a ruler and marked with a red acetate marker on the file) without rotating the instrument.

Group 2

The root canals were flushed using 3 mL of 17% EDTA and agitated with an MAF as described in Group 1.

Group 3

The root canals were flushed using 3 mL of a 1:1 mixture of 5% NaOCl and 18% HEBP (Sigma-Aldrich Co., St. Louis, USA). This resulted in a solution that was 2.5% NaOCl and 9% HEBP. It was agitated with an MAF as described for Group 1.

Group 4

The root canals were flushed using 3 mL of 1% PAA and agitated with ultrasonic activation (UA) for 30 s using a smooth noncutting ultrasonic instrument (ESI ‑ Endo soft instrument, EMS, Le Sentier, Switzerland) attached to an ultrasonic device (EMS, Le Sentier, Switzerland). The ultrasonic instrument was inserted into the root canal 1 mm short of WL.

Group 5

The root canals were flushed using 3 mL of 17% EDTA and agitated with UA for 30 s with a smooth noncutting ultrasonic instrument attached to an ultrasonic device. The ultrasonic instrument was inserted into the root canal 1 mm short of WL.

Group 6

The root canals were flushed using 3 mL of a 1:1 mixture of 5% NaOCl and 18% HEBP. This resulted in a solution of 2.5% NaOCl and 9% HEBP. It was agitated with UA for 30 s using a smooth noncutting ultrasonic instrument attached to an ultrasonic device. The ultrasonic instrument was inserted into the root canal 1 mm short of the WL.

All the root canals in each experimental group were then manually irrigated using a 30 gauge needle tip with 3 mL of distilled water.

After the removal of the temporary fillings, a number 15 K‑file was introduced to the WL to loosen the $Ca(OH)_2$ and to create space for an irrigation needle. The 30 gauge needle tip (NaviTip; Ultradent, South Jordan, UT) was placed 1 mm short of the WL without binding.[8,9,29]

UAs were performed without touching the walls with the instrument that was placed into the canal 1 mm short of the WL, enabling it to vibrate freely. The ultrasonic file was activated at the 5 power setting for 30 ss, and irrigations were performed passively at a 3 mL/min flow rate through the unit. The removal process of $Ca(OH)_2$ intracanal medication from the root canals is shown in Figure 1.

After the irrigation procedures, the canals were dried with paper points. Grooves were prepared with a water‑cooled cylindrical diamond bur in a high‑speed handpiece on the buccal and lingual surfaces to avoid damaging the inner shelf of dentine surrounding the canal. The teeth were split along their long axis in a buccolingual direction using a hammer and chisel. All endodontic procedures were performed by a single operator to avoid interoperator variability.

For scanning electron microscopy analysis, the samples were dehydrated and coated with gold‑palladium particles (20 nm) and magnification of ×1000 was used to evaluate the cleanliness of the canal. Walls at the apical, middle, and cervical root canal thirds (3, 6, and 9 mm from the apex, respectively) were observed.[30] A scoring system that was used in the study of Kuga *et al.*[31] was defined to assess the quantity of the residues on the canal walls. The scores used were as follows:

- Score 0: No residue
- Score 1: Small amount of residues (20% of the root canal surface was covered)
- Score 2: Moderate amount of residues (20–60% of the root canal surface was covered)
- Score 3: A large amount of residues (more than 60% of the root canal surface was covered).

A scanning electron microscopic image for each score is shown in Figure 2. Two experienced investigators who were blinded to the experiment scored the specimens until there was a consensus between them. Selected specimens were calibrated for energy dispersive X‑ray microanalyzer to verify the $Ca(OH)_2$ residue on the root canal walls.

Statistical interpretations were performed using Kruskal–Wallis and Tukey's *post‑hoc* tests, at the 95% level of significance. Statistical results are shown in Table 1.

Results

Comparisons were done according to the cleanliness of the root canal thirds. There were significant differences among the groups ($P < 0.05$).

Comparisons of the coronal thirds showed that; PAA + UA was superior to EDTA + MAF and HEBP + MAF.

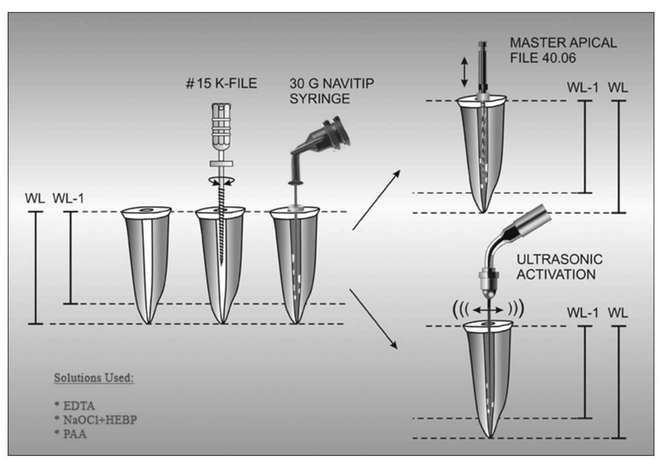

Figure 1: The schematic drawing of test set-up and the removal process of calcium hydroxide intracanal medication from the root canals

Table 1: The medians and the quartiles of the groups for each root thirds were shown

	Median	Coronal thirds		Median	Middle thirds		Median	Apical thirds	
		25%	75%		25%	75%		25%	75%
PAA + MAF	0^{a}	0	0	0^{x}	0	1	1z	0	1
EDTA + MAF	1^{b}	1	2	1^{y}	1	2	1z	1	2
HEBP + MAF	1^{b}	1	1	1^{xy}	1	1	1z	1	2
PAA + US	0^{a}	0	0	0^{x}	0	0	1z	0	1
EDTA + US	1^{a}	0	1	1^{y}	1	2	1z	1	1
HEBP + US	0.5^{a}	0	1	1^{xy}	1	1	1z	1	1
Control+	3	3	3	3	3	3	3	3	3
Control−	0	0	0	0	0	0	0	0	0

Each root third was evaluated independently. For each root third, different superscript letters show significant differences. MAF=Master apical file, US=Ultrasonic, PAA=Peracetic acid, EDTA=Ethylenediaminetetraacetic acid, HEBP=1-hydroxyethylidene-1,1-bisphosphonate

Table 2: The percentages of each score are shown

	Coronal root thirds (%)				Middle root thirds (%)				Apical root thirds (%)			
	S0	S1	S2	S3	S0	S1	S2	S3	S0	S1	S2	S3
PAA + PUI	60	40	-	-	60	40	-	-	10	90	-	-
HEBP + PUI	50	50	-	-	30	70	-	-	-	100	-	-
EDTA + PUI	30	70	-	-	-	60	40	-	-	100	-	-
PAA + MAF	80	20	-	-	50	50	-	-	30	70	-	-
HEBP + MAF	-	90	10	-	-	80	20	-	-	70	30	-
EDTA + MAF	10	80	10		10	70	20	-	-	70	20	10
Control+	-	-	-	100	-	-	-	100	-	-	-	100
Control−	100	-	-	-	100	-	-	-	100	-	-	-

PAA=Peracetic acid, PUI=Passive ultrasonic irrigation, EDTA=Ethylenediaminetetraacetic acid, HEBP=1-hydroxyethylidene-1,1-bisphosphonate, MAF=Master apical file

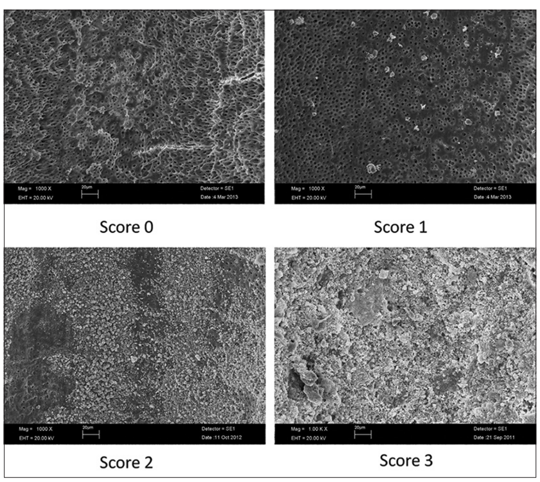

Figure 2: The scanning electron microscopy photographs of each score are shown

PAA + MAF was superior to EDTA + MAF and HEBP + MAF ($P < 0.05$).

Comparisons of the middle thirds showed that; both PAA + MAF and PAA + UA were superior to EDTA + MAF and EDTA + UA. PAA + UA was superior to HEBP + MAF ($P < 0.05$).

There were no significant differences among the rest of the experimental groups at the coronal, middle, and apical thirds ($P > 0.05$).

The frequency of "Score 0" of the PAA groups (Group 1 and Group 4) were higher than the other of groups at all root thirds. Scores of each experimental group with percentages are given in Table 2.

Discussion

The removal of $Ca(OH)_2$ intracanal dressing from the root canals have been evaluated by several authors. However; complete cleaning of the root canal from residual $Ca(OH)_2$ still could not be achieved [9,24,30-32] and it showed that the residual $Ca(OH)_2$ may affect the sealing ability and adhesion of endodontic sealers to the root canals.[33]

Different types of irrigation protocols have been used in the removal of $Ca(OH)_2$ medicament from the root canals. Kuga *et al.*[31] used NaOCl or EDTA solutions in combination with two types of rotary instruments and reported that final irrigation solutions were not very effective in the removal of $Ca(OH)_2$ residues. Wiseman *et al.*[26] evaluated the effect of sonic and ultrasonic agitation techniques in the removal of $Ca(OH)_2$ paste and reported the ultrasonic technique as more effective. Kenee *et al.*[23] reported that ultrasonic and rotary instrumentation in $Ca(OH)_2$ removal were more effective than the irrigant-only technique. In the present study, we also preferred agitation with the MAF and ultrasonic methods over the irrigant-only techniques for the effective removal of $Ca(OH)_2$.

PAA solutions have been evaluated in endodontics as an irrigant by several authors.[9,10,12,15] It showed that PAA solutions could dissolve the smear layer as well as 17% EDTA solution.[10,12] One study evaluated the removal of $Ca(OH)_2$ intracanal medicament from the root canal and used irrigant-only techniques with 1% PAA, 0.5% PAA, NaOCl + EDTA and only EDTA solutions and reported that 1% PAA was significantly superior to the other groups at the coronal and apical thirds.[9] Due to the successful results of 1% PAA, we also preferred 1%PAA as an irrigant in our study.

NaOCl-alone as an irrigant was reported to be inadequate in the removal of $Ca(OH)_2$ intracanal medicament.[5,23] The combination of NaOCl and EDTA was found to be more effective in the removal of $Ca(OH)_2$ from the root canal.[21] In the present study, to standardize the total irrigant volume and ultrasonic agitation time of the experimental groups we did not use the NaOCl + EDTA group.

HEBP was stated as a weak chelator which does not harm the mineral content of dentine, and it does not diminish the proteolytic or antimicrobial activity of NaOCl in combined usage.[14-16] Neelakantan *et al.*[20] stated that during root canal instrumentation the use of HEBP + NaOCl irrigation improved the push-out bond strength of an epoxy resin-based sealer in all root thirds. In another study, it was also reported that irrigation with HEBP reduced the hard tissue debris accumulation more than irrigation in the NaOCl group.[34]

In the present study, we used the instrument 1 mm short of the WL during agitation with the ultrasonic system and an MAF to avoid the preparation of root canal walls. A 30 gauge needle was also used to prevent the extrusion of irrigants and so that the procedure could be applicable to small and curved root canals.

Under the tested conditions, the complete removal of $Ca(OH)_2$ medicament from the root canal walls was not obtained as it was stated before,[23,24] and $Ca(OH)_2$ residue were found in all root thirds regardless of the solution and agitation technique used.

According to the results of the study, at the coronal root thirds, regardless of the agitation technique, PAA showed a significantly cleaner root canal wall compared to the EDTA-MAF and HEBP-MAF groups ($P < 0.05$). These results could be related to the study of Sagsen *et al.*[9] who stated that 1% PAA was more effective than 17% EDTA in the removal of $Ca(OH)_2$ from the root canals. On the other hand, according to comparisons in the coronal thirds, there were no significant differences between the ultrasonically activated PAA, EDTA and HEBP groups ($P > 0.05$). Furthermore, there were no significant differences among the remaining experimental groups ($P > 0.05$). In the coronal thirds, we could say that UA improved the $Ca(OH)_2$ removal capability of the EDTA and HEBP solutions and these results are in agreement with the results of Kenee *et al.*[23] who stated that UA of irrigants improved the removal efficiency of $Ca(OH)_2$.

The statistical results of the middle root thirds showed that the activity of the 1% PAA-ultrasonic and 1% PAA-MAF groups were significantly superior to the EDTA-ultrasonic and EDTA-MAF groups ($P < 0.05$). As we mentioned in the results of the coronal root thirds; in regardless of the agitation technique, the 1% PAA irrigant was reported to be more effective than the 17% EDTA irrigant. This result is in accordance with the results of Sagsen *et al.*[9] who stated that 1% PAA is more effective than 17% EDTA in the removal of $Ca(OH)_2$. This result could also be related to different decalcification properties of the 1% PAA solution and the 17% EDTA solution.[12] It was stated that the acidity of PAA is stronger than EDTA.[16] Due to the stronger acidity of PAA, more calcium ions remain in the solution and do not reprecipitate.[35] This reaction could be responsible for the cleaner root canal surfaces. In addition, the 1% PAA-ultrasonic group was significantly superior to the HEBP-MAF group ($P < 0.05$). However, there were no significant differences among PAA-ultrasonic group and the HEBP-ultrasonic group, and the PAA-MAF and the HEBP-MAF groups, and the HEBP-ultrasonic and the PAA-MAF groups ($P > 0.05$). In previous studies, it was stated that the NaOCl alone is not an effective irrigant in $Ca(OH)_2$ removal from the root canals.[5,23,30] Therefore, we preferred to use NaOCl in combination with HEBP. HEBP is known to be a weak chelator[16] and it is certain that a lower amount of Ca^{+2} ions remains than when using the 1% PAA solution. At this point, in the middle thirds; ultrasonic agitation seemed to improve the $Ca(OH)_2$ removal capability of HEBP and PAA irrigations more than the MAF agitation.

In the apical root thirds, there were no significant differences among the groups ($P > 0.05$). Sagsen *et al.*[9] reported that 1% PAA is more effective than 17% EDTA, 2.5% NaOCl + 17% EDTA and 0.5% PAA for removing $Ca(OH)_2$ from the apical third of the root canals. According to our results, at the apical thirds, the agitation of irrigation solutions improved the $Ca(OH)_2$ removal capability from the root canal. Apical root dentine has a sclerotic structure and less number of dentine tubules. It may be that $Ca(OH)_2$ particles could not find a reservoir opening such as an opened dentine tubule orifice. In the apical thirds, the circulation volume of the irrigants was lower because of the volume of the apical root thirds. The results of apical thirds could be related to irrigant volumes. Furthermore, the irrigant needle type could have been affected from this result. An open ended needle type was used in this study, and open-ended needles were reported to create a jet toward the apex with maximum irrigant replacement.[36] The agitation method used in the present study could improve the effect of this replacement.[23,24,28]

In the present study, at all root thirds, there were no significant differences between the HEBP + NaOCl group and the EDTA group regardless of agitation techniques ($P > 0.05$). As stated before, HEBP is a weaker chelator than EDTA[37] and as a result of this preserves less Ca^{+2} ions. However, it seems that agitation techniques can improve the capability of HEBP in the removal of $Ca(OH)_2$.

Conclusions

In the present study, the agitation techniques improved the intracanal $Ca(OH)_2$ medicament removing capability of the irrigants. However, further investigation with present irrigation protocols is needed on this topic.

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Nil.

Conflicts of interest
There are no conflicts of interest.

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