Efficacy of Three Irrigation Methods in Removing Calcium Hydroxide from Curved Root Canals: An *In Vitro* Micro-CT Study

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Department of Conservative Dentistry, Faculty of Dental Medicine, Medical University-Sofia, Bulgaria Aims and Objectives: This in vitro study aimed to compare the efficacy of three different irrigation methods in removing calcium hydroxide from curved root canals and to estimate the amount of residual medicament using microcomputed tomography. Materials and Methods: Thirty extracted molars with curved mesial root canals, ranging from 25° to 30°, were prepared with ProTaper Next using X2 to full working length and were filled with calcium hydroxide. Teeth also were randomly divided into four groups: group 1 (n = 8)— Erbium: Yttrium-Aluminum-Garnet (Er:YAG) laser; group 2 (n = 8)—EndoActivator; group 3 (n = 8)—EndoVac; and a control group (n = 6)—negative control with no calcium hydroxide. Microcomputed tomography was used to evaluate the residual amount of filling material. Statistical analysis was performed using IBM SPSS Statistics 26, with a significance level of 0.05. Descriptive statistics, student t test, and Chi-square test were applied for data analysis. **Results:** The greatest amount of residual calcium hydroxide was observed in the samples with EndoVac irrigation (0.515 mm³), followed by EndoActivator (0.381 mm³) and Er:YAG laser (0.121 mm³). However, no statistically significant difference was observed between the groups. The greatest residual amount was observed in the apical third (0.419 mm³), followed by the middle (0.050 mm³) and the coronal (0.015 mm³). Conclusions: No statistically significant differences were observed in the efficacy of the tested techniques for removing calcium hydroxide from curved root canals, whereas a significant difference was observed in the distribution of residual material depending on the technique used. The removal of the calcium hydroxide was more effective in the coronal and middle third of the canal. The greatest residual material was found in the apical portion of the root canal.

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INTRODUCTION

There are several instrumentation techniques and irrigation solutions that could be used to reduce and remove the microorganisms from the root canal system.^[1,2] Still, thorough and complete disinfection of the root canal system remains challenging, especially in cases with polymicrobial infection. Intracanal medicament placement could lead to better disinfection in such cases.^[3,4] Calcium hydroxide (Ca(OH)₂) is one

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of the most widely used intracanal medicaments for treating infected root canals. Its antibacterial activity is due to its high pH—12.5.^[5,6] Usually, it is applied for 7–14 days. Before the final obturation, it should

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be completely removed as remnants of Ca(OH)₂ left in the root canal could interfere with the properties of the sealer and its sealing ability.^[7,8] A series of studies reveal that removing Ca(OH)₂ from the dentinal walls, especially in the apical third of the canal, is very difficult.^[9] Cases with curved canals are even more challenging. The presence of lateral canals and isthmuses also interferes with its complete removal.^[10-12]

Various techniques have been recommended for the removal of $Ca(OH)_2$ from root canals.^[13] The most frequently applied technique is irrigation with sodium hypochlorite (NaOCl), using a syringe and endodontic needle, combined with filing motion and activation with the master apical file. The efficacy of the method depends on the proximity of the needle tip to the root canal end, the space available in the apical third, and the amount of the irrigation solution used. However, with this technique, the complete removal of the Ca(OH)₂ could not be achieved, especially in the apical third of the root canal.^[14-18]

Several studies have shown that a combination of sonic and ultrasonic irrigation with NaOCl and ethylenedaminetetraacetic acid (EDTA) significantly improves the removal of Ca(OH)₂. However, achieving complete removal still poses difficulties.^[10,19] Another device that could be helpful is the EndoVac (Discus Dental, Culver City, California), which creates negative pressure in the apical third of the root canal after a mechanical enlargement until 35 or more, allowing for better penetration of the irrigating solution.^[20,21] One of the main advantages of the technique is the safe delivery of the irrigant without the risk of extrusion. However, the limited maximal speed of the fluid current leads to a slow exchange of the irrigation solution, reducing its cleaning efficacy.^[22]

There are also studies on the ability of laser-assisted irrigation to remove Ca(OH)₂, which state that Erbium: Yttrium-Aluminum-Garnet (Er:YAG) laser improves the cleaning of Ca(OH)₂ from the root canal system.^[23-25]

Despite the various devices and methods available, complete removal of $Ca(OH)_2$ is difficult and cannot always be guaranteed, which is the reason why still studies are being conducted to find the most appropriate combination of techniques and irrigants.^[4,13,26,27]

The presented study aimed to compare the efficacy of three irrigation methods for removing $Ca(OH)_2$ from the root canals and to estimate the amount of residual material using micro-computed tomography (CT) analysis. The null hypothesis posits that all presented methods are equally effective in removing $Ca(OH)_2$ along the entire length of curved root canals.

MATERIALS AND METHODS

SELECTION OF EXPERIMENTAL TEETH

Thirty extracted human molars with fully developed apices, free from cracks (evaluated using a stereomicroscope "Leica S6"), denticles (evaluated by X-ray), and root caries lesions were selected. These 24 teeth constitute our sample, which was randomly selected from 97 teeth. The sample was subsequently reduced to a size that would yield statistically significant results. The teeth (n = 6) in the control group (without Ca(OH)₂) are randomly chosen from a total of 25 teeth. The curvatures of their mesial root canals were from 25° to 30°. Duerr Dental Imaging system was used for the radiographic determination of the curvature [Figure 1]. Ethical approval for the project (contract no. 146/2022) was obtained by the Commission on scientific research ethics at Medical University.

ENDODONTIC PREPARATION OF THE EXPERIMENTAL TEETH

To standardize the samples, the coronal part of the each tooth was removed using a high-speed handpiece and a diamond bur, leaving a standardized length of 16 mm until the end of the root.

Endodontic preparation of the samples involved using a K-file ISO 10 (Dentsply Sirona, Bellaigues, Switzerland) for determining the working length. The file was inserted until its tip was seen apically, and 1 mm was subtracted from that length. All root canals were enlarged using machine rotary Ni-Ti files—Pro Taper Next (Dentsply Sirona Bellaigues, Switzerland) to an master apical file—X2 (0.25/.06). The standard



Figure 1: Measuring the curvature of a sample tooth using Duerr Dental Imaging program and Shneiders' method.

endomotor program for this system was used with a speed of 300 rpm and torque of 2 N/cm).

Thorough irrigation with 2% sodium hypochlorite (NaOCl) was performed after each instrument. The final irrigation included 5mL 2% NaOCl, followed by a rinse with 5mL of distilled water and 3mL of 17% EDTA for 1min, using an endodontic needle 30G, inserted 2mm shorter than the working length. The canals were then dried using paper points size X2.

All extracted human teeth were prepared only by one operator to maintain consistency.

CALCIUM HYDROXIDE PLACEMENT

Water-based $Ca(OH)_2$ (Calcipast, Cerkamed, Stalowa Wola, Poland) was introduced into the canal using a Lentulo spiral size 25 until the canal was filled 1 mm shorter than the working length. Following this, a sterile cotton pellet and a temporary filling were placed coronally. Control X-ray for determination of the level of the intracanal medication was taken.

The samples were stored in 100% humidity at 37°C for 7 days.

CALCIUM HYDROXIDE REMOVAL

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The prepared samples were randomly assigned to three groups (n = 8) based on the technique used for the removal of Ca(OH)₂. These techniques included using the Er:YAG laser 2940 nm, sonic activation (EndoActivator, Dentsplay Sirona UK), negative pressure system (EndoVac, Sybron Endo, USA). In addition, there was a control group, in which no Ca(OH), was used.

In group 1 (n = 8), the removal of Ca(OH)₂ was achieved by using Er:YAG laser 2940 nm (Light Walker, Fotona, Ljubljana, Slovenia) with the following parameters—0.15 W power, 15-Hz frequency, 10 mJ, 50-µs impulse length, 300-µm size of the optical fiber, and 16-mm length. The fiber was placed 2 mm shorter than the working length of the root canal and is moved up and down along the canal. After placing 5mL of 2% NaOCl in the canal, it was activated with the laser for 30s. Then, an additional irrigation with 5mL NaOCl, 5mL distilled water, and 3mL 17% EDTA was performed. The effectiveness of medication removal was confirmed when the rinsing solution became clear.

In group 2 (n = 8), sonic activation was done using EndoActivator. The sonic tip was positioned 2mm shorter than the working length. The canals are filled with 2% NaOCl, and the solution is activated for 30 s, followed by irrigation with distilled water and then 3 mL of 17% EDTA for 1 min. Final irrigation with distilled water was done to neutralize the effect of EDTA. The canals were dried with X2-sized paper points.

In group 3 (n = 8), the removal of the Ca(OH)₂ was achieved using negative apical pressure with EndoVac. A macrocannula size of 0.32 was used to evacuate the solution, creating a vortex-like cleaning. The procedure involved using 5mL of NaOCl initially, followed by irrigation with 3mL distilled water and 3mL 17% EDTA for 1 min, and final irrigation with 5mL distilled water. The canals were dried with X2-sized paper points.

The control group (n = 6) received no medication and served as the negative control.

MICRO-CT SCANNING AND EVALUATION OF THE RESIDUAL AMOUNT OF CALCIUM HYDROXIDE

The samples were scanned after removing the Ca(OH)₂. The residual medication was segmented using the region growing method, represented in white and measured in mm³. NIKON XT H 225 (Nikon Metrology, Tring, UK) industrial micro-CT was used. The voltage and power of energy used were 105 kV and 106 μ A, respectively. The maximal geometric magnification allowed a resolution of 16 μ m.

The reconstruction of the images was performed using CT Pro 3D version XT 3.1.3, (Nikon Metrology, Hertfordshire, UK).

STATISTICAL ANALYSIS

Descriptive statistics were done, including the arithmetic mean (mean), total count (*N*), and relative percentage (%). Statistical hypothesis testing was done at a risk of error of 5% ($\alpha = 0.05$) and a confidence probability of 95%. Data were statistically analyzed using IBM SPSS Statistics 26 (New York, USA), level of significance of 5%. The graphics were made using Excel 2015. Student *t* test to compare two independent groups.

• Chi-square test to assess the association between two variables. If a relationship was identified in this test, Cramer's coefficient was then used to estimate the strength of that relationship. The coefficient ranged from 0 to 1 and fell within the following categories:

0 < | Cramer's V | < 0.3—weak relationship

0.3 < | Cramer's V | < 0.5—moderate association

< | Cramer's V | < 0.7—significant association

0.7 < | Cramer's V | < 0.9—high association

0.9< | Cramer's V | < 1.0—very high association

Table 1: Comparative analysis of the amount of residual Ca(OH), in mm ³ along the entire length of the root canal							
Technique used	n	Min	Max	Standard deviation	Mean	<i>P</i> value	
Er:YAG laser	8	0.050	0.191	0.099	0.121	0.188	
EndoVac	8	0.291	0.738	0.182	0.515	0.100	
EndoActivator	8	0.034	0.896	0.372	0.381		
Control group	6	0.00	0.00	0.00	0.00		

Er:YAG, Erbium: Yttrium-Aluminum-Garnet



Figure 2: Amount of residual calcium hydroxide in mm³ along the entire length of the root canal



Figure 3: Micro-CT scan of extracted teeth after removal of Ca(OH), with Erbium: Yttrium-Aluminum-Garnet laser

Results

Regarding the amount of residual material, the results revealed the greatest amount of residual $Ca(OH)_2$ in the group that used negative apical pressure with EndoVac measuring 0.515 mm³. This was followed by the group with sonic activation (EndoActivator) with 0.381 mm³ and Er:YAG laser group with 0.121 mm³ [Table 1 and Figure 2]. However, no statistically significant difference was observed between the studied groups (P = 0.188). The control group, which served as a negative control, was not included in the following comparisons and analysis due to the absence of Ca(OH)₂. Remnants of Ca(OH)₂ were found along the entire length of the root canal (coronal, middle, and apical portions) [Figures 3–5].



Figure 4: Micro-CT scan of extracted teeth after removal of Ca(OH), with EndoActivator



Figure 5: Micro-CT scan of extracted teeth after removal of Ca(OH), with EndoVac

Table 2: Distribution of the residual material along the length of the root canal				
Position of the	Average value of the	<i>P</i> value		
residual Ca(OH) ₂	Ca(OH) ₂ in mm ³			
Coronal third	0.015	0.035		
Middle third	0.050	0.055		
Apical third	0.419			

The mean amount of residual Ca(OH)₂ was 0.050 mm^3 in the middle portion of the canal, 0.419 mm^3 in the apical, and 0.015 mm^3 in the coronal. Most Ca(OH)₂ was found in the apical portion, accounting for 64% of the total. The *P* value of 0.035 (<0.05) indicates that there is a significant difference in the volume of residual material at various portions of the canal [Table 2].

Table 3 presents the distribution of the residual material in the root canal based on the removal technique.

Table 3: Correlation between the technique used and the position of the residual material						
Position of residual Ca(OH) ₂	Technique					
	Er:YAG laser	EndoVac	EndoActivator			
Coronal third	-	-	50.0%			
Middle third	50.0%	-	-			
Apical third	50.0%	100.0%	50.0%			
Significance level		P = 0.039				

Er:YAG, Erbium: Yttrium-Aluminum-Garnet

The Ca(OH), was randomly distributed in the apical and middle portion in the group with Er:YAG laser activation, whereas in the group with sonic activation, half of the amount of residual material was found in the coronal and apical portion. In the group where the EndoVac system was used, all residual Ca(OH), was found apically. The P value of 0.039 (<0.05) indicates a significant correlation between the method of removal of the Ca(OH), and the position of the residual material. This conclusion could be stated with a probability of 95%. We could also determine the connection and evaluate its strength using Cramer's V coefficient. Its value was in the range of 0.5 < | Cramer's V | < 0.7, which indicates the strength of the correlation between the position of the residual material and the cleaning technique as a significant association.

DISCUSSION

The highest mean scores for residual Ca(OH), were observed in the samples that used EndoVac irrigation. However, no statistically significant differences were observed in the efficacy of the tested techniques for removing Ca(OH), from curved root canals. The removal of the Ca(OH), was more effective in the coronal and middle third of the canal. Statistically significant differences were found in the amount of residual material at various parts of the root canal when using different irrigation methods. The residual Ca(OH), was evenly distributed along the apical and middle third of the root canal when Er:YAG laser was used, whereas in the EndoActivator group, half of the material was found in the coronal and apical portions. All residual material was in the apical third of the canal in the EndoVac group. The null hypothesis is partially accepted as there is no significant difference in the efficacy for removing Ca(OH), but a significant difference in the amount of the residual material at various parts of the root canal when different techniques were used.

Based on the existing literature, the removal of $Ca(OH)_2$ from the root canal system poses challenges, especially in cases with curved root canals.^[27,28] The complex anatomy of the root canal system, with its curvatures, lateral canals, and isthmuses, not only makes the penetration of irrigants in the mechanically untreated space more difficult but also interferes with the complete removal of the different intracanal medications.^[29]

In the present study, NaOCl (2%) and 17% EDTA were used for irrigation during the root canal mechanical preparation and the subsequent removal of the intracanal medication. However, certain studies in the literature suggest that 10% of citric acid with EDTA is more effective in the removal of Ca(OH)₂.^[30,31] The application of EDTA for more than 60s leads to penetration in more than 50-µm depth in the dentinal tubules and subsequent demineralization of the dentin.^[32] To mitigate this issue, controlling the time for exposure to the decalcifying solution, using the proper concentration and sequence of irrigants, and using lessaggressive instruments are essential measures.^[11]

In this study, remnants of $Ca(OH)_2$ were observed in all samples, regardless of the removal technique. These results are consistent with the findings of other auth ors.^[10,11,13,21,30]

The most effective removal of intracanal medicament was observed in the samples that used laser-assisted irrigation, which is in alignment with results reported by other researchers. Arslan *et al.*^[15] use Ca(OH)₂ on teeth with artificial grooves prepared apically. They try to remove it with photon-induced photoacoustic streaming (PIPS), ultrasonic activation, EndoActivator, and conventional techniques. The group with PIPS showed the best results.^[15]

EndoActivator, a sonic device with a polymer tip, does not cause damage to the root canal walls compared with ultrasonic devices that use metal files. When comparing the effectiveness of the sonic and ultrasonic activation, some studies report that the application of the ultrasonic technique during the removal of $Ca(OH)_2$ has the potential to change the morphology of the root canal system. Another study states that there could be created significant forces during the mechanical preparation of the root canal system, leading to defects and fractures in dentin.^[33] Some studies found that passive ultrasonic irrigation removes $Ca(OH)_2$ better than sonic activation and leaves less residual material in the root canal.^[10,18] In the presented study, the results achieved with sonic activation were better than other groups. Song *et al.*,^[34] in their study, reported that EndoActivator and precurved ultrasonic files were more effective in removing Ca(OH)₂ than precurved K-files and needle irrigation alone. In another study, Moon *et al.*^[35] studied removing Ca(OH)₂ with EndoActivator, EQ-S, and Vibringe in straight, moderate, and severe curved canals. They found that in the canals with severe curvature, the volumes of residual Ca(OH)₂ were significantly higher than in straight canals.^[35] Ulusoy *et al.*^[36] concluded that sonically activated systems with flexible tips could be beneficial for removing intracanal dressing in the curved apical canals.

Alturaiki et al.[27] conducted a study to compare the effectiveness of various irrigation techniques (conventional needle and syringe, EndoActivator, ProUltra Piezo flow Ultrasonic, and EndoVac) in root canal treatment. They found that in the group with EndoActivator, the removal of Ca(OH), from all parts of the root canal was better. In the EndoVac group, there were no statistically significant differences in the coronal and middle portions of the root canal compared with the conventional technique, but the differences were significant for the apical part.^[27] The results in our study are close to these-we found statistically significant differences in the amount of the residual material at various parts of the root canal when different techniques were used. The residual Ca(OH), was evenly distributed along the apical and middle third of the root canal when Er:YAG laser was used, whereas in the EndoActivator group, half of the material was found in the coronal and apical portions. All residual material was in the apical third of the canal in the EndoVac group. The best cleaning of the medicament was achieved in the coronal third of the canal (0.015mm³). This could be attributed to the better preparation in this region, allowing for irrigation solutions and improved cleaning.^[37] Other authors also have reported common results.[15,17,18] There are also studies that report the presence of Ca(OH), mainly in the apical third of the root in cases with curved canals the greater amount is found within 0-1 mm from the apex.^[25,38] Different micro-CT study shows that after removing the Ca(OH), the residual material is usually left in the apical part and in the irregularities in the root canal due to the complex anatomy in this region.^[36,39] These results are in correlation with the results, which we found in our study.

Micro-CT, a nondestructive method, was employed to study the anatomy of root canals. This technique can measure the volume of debris after instrumentation or residual $Ca(OH)_2$ after using different removing methods. The findings of this study will help dentists to choose a better method for removing $Ca(OH)_2$ from curved root canals. However, the study was unable to show the statistical difference because of anatomical variation of molars, as these variations might have influenced the results.

CONCLUSIONS

Within the limitation of the present study, there were no statistically significant variations observed among the techniques Er:YAG laser, EndoActivator, and EndoVac when employed to eliminate Ca(OH), from curved root canals. However, statistically significant differences were identified in the residual material at various parts of the root canal depending on the technique used. When the Er:YAG laser was used, the residual Ca(OH), was uniformly spread along the apical and middle third of the root canal. On the other hand, in the EndoActivator group, approximately half of the material was concentrated in the coronal and apical regions. As for the EndoVac group, all the residual material was confined to the apical third of the canal. The removal of the Ca(OH), was more effective in the coronal and middle third of the canal, whereas the highest amount of residual material was found in the apical portion of the root canal.

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CONFLICTS OF INTEREST

There are no conflicts of interest.

AUTHORS CONTRIBUTIONS

ER, JM, MM-T conceptualized the study and collected the data. ER wrote the paper. All authors read and approved the final manuscript.

ETHICAL POLICY AND INSTITUTIONAL REVIEW BOARD STATEMENT

Ethical approval was obtained by the Comission on Scientific Research Ethics at Medical University, Sofia, Bulgaria.

PATIENT DECLARATION OF CONSENT

Not applicable.

DATA AVAILABILITY STATEMENT

Not applicable.

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