Original Research
Journal of Pierre Fauchard Academy (India Section), *Vol 37(3-4), DOI: 10.18311/jpfa/2023/34938, July-December 2023 p. 49-55*
ISSN (Print) : 0970-2199
ISSN (Online) : 2405-772X

Removal of Calcium Hydroxide Medicament from the Apical Root Canal using Different Irrigation Methodologies: A Scanning Electron Microscopy Study

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Abstract

Introduction: To evaluate the efficiency of different irrigants and agitation techniques in the removal of calcium hydroxide $Ca(OH)_2$ medicament from the root canal walls. **Material and Methods:** N=84 mandibular premolars were selected for this study. The root canal of these 84 premolars was cleaned up using the ProTaper rotary system up to the file size F3 and then filled with $Ca(OH)_2$ medicament. For seven days, the root samples were maintained at 37° C and 100% relative humidity. The sample was divided into four groups (n=20 each) using random allocation employing different agitation methods: Group 1- MAF - Master Apical File, Group 2 - PUI - Passive Ultrasonic Irrigation, Group 3 - CB - Canal Brush, and Group 4 - FF - F File. Each group was further subdivided into two subgroups based on the specific chelating agent employed: (a) 10% Citric Acid and (b) 17% EDTA. Longitudinal bisection was performed in the apical (lower) third of the root structure. Further, an assessment was done using SEM (scanning electron microscopy) to study the elimination of $Ca(OH)_2$. Kruskal-Wallis and Mann-Whitney tests were used for statistical analysis. **Results:** Groups 1 (MAF) and Group 3 (CB) showed a significant statistical difference ($p<0.05$) when compared to Group 2 (PUI) and Group 4 (FF). An insignificant difference ($p>0.05$) was seen between the performance of chelating agents (10% Citric Acid and 17% EDTA). **Conclusion:** Irrespective of the specific chelating agents employed, both the PUI (Passive Ultrasonic Irrigation) and FF (F File) agitation techniques exhibited superior efficacy in the removal of $Ca(OH)_2$ medicament from the apical (lower) third when compared to the CB (Canal Brush) and MAF (Master Apical File) methods.

Keywords: Agitation Techniques, Calcium Hydroxide Removal, Chelating Agents

1. Introduction

The role of Calcium hydroxide as an intracanal medication has been associated with peri-radicular healing due to its antibacterial efficacy. Other biological features of calcium hydroxide include the capacity to dissolve tissue, the ability to prevent tooth resorption, and the ability to stimulate the growth of hard tissue during repair. These outcomes have led to the use of calcium hydroxide in numerous therapeutic settings[1].

Its alkaline pH and capacity to quickly dissociate into calcium and hydroxyl ions are the key causes of this material's clinical efficacy. Calcium hydroxide is placed into the canal and left there for different durations, which can vary from 7 days for intracanal asepsis to a longer period of 6 to 24 months for apexification[2].

Complete elimination of $Ca(OH)_2$ from the root canal is always important before the permanent obturation is performed. It was found that incomplete removal of calcium hydroxide may lead to an increase in the apical leakage in endodontically treated teeth, poor adhesion, and penetration of resin sealers to dentin tubules. Additionally, the calcium remnants can potentially interact with zinc oxide eugenol sealers, causing them to become brittle and granular. Therefore, it is of utmost importance to ensure the thorough removal of calcium hydroxide from the root canal before proceeding with the obturation process[2].

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Article Received on: 03.09.2023 Revised on: 09.10.2023 Accepted on: 18.11.2023

Various methods have been advocated for this purpose such as employing various irrigants and irrigation procedures[3], instrumentation of the root canal with the Master Apical[4], Passive Ultrasonic Irrigation, endodontic microbrush, etc. The Passive Ultrasonic Irrigation (PUI) technique entails the use of an oscillating file or a smooth wire to transmit acoustic energy to an irrigant within the root canal. This energy is transmitted through ultrasonic waves, which can generate cavitation and acoustic streaming effects within the irrigant[5].

A new endodontic microbrush called the Canal Brush (Roeko Canal BrushTM, Coltène/Whaledent, Langenau, Germany) has just been released. Three different sizes are offered for this extremely flexible microbrush, which is made entirely of polypropylene. Removing debris from root canal extensions and abnormalities is thought to be most efficient when small, flexible Canal Brushes are used in conjunction with irrigation treatments[6].

A new root canal debridement tool called the "F-file" (Plastic Endo, LLC) made of endodontic polymer has recently been introduced. To stir up the irrigant in the canal, a fine diamond-coated plastic rotary file that is single-use, sterile, and disposable is utilized. The 20 mm tip of the polymer rotary finishing file has a 0.04 taper. The tip size and taper improve the effectiveness of its debridement[7].

EDTA is commonly used as an irrigant to remove the smear layer. It is combined with a cationic detergent to improve its diffusion and effectiveness.[8-12] Other products such as Citric acid have also been investigated for their capacity to dissolve the inorganic constituents of the smear layer as well as for intracanal medication.

This study aims to assess the efficiency of different irrigants and agitation techniques employed in the removal of calcium hydroxide medication from root canals.

2. Material and Methods

In this study, N=84 recently extracted mandibular single-rooted, single-canal premolars without any visible root caries, fractures, cracks, internal or external resorption, or calcification as examined under 4x magnification, and a fully developed root apex was used. To eliminate organic material, the teeth were kept in 3% NaOCl for two days at room temperature after extraction. The teeth were subjected to ultrasonic scaling to remove any adherent tissue debris, followed by thorough rinsing with distilled water, followed by immersion in 10% formalin solution until they were ready for use.

By Schneider's classification (1971), the root canals classified as straight were included in the study, i.e., roots with a curvature not exceeding five degrees were used. Pre-operative radiographs showing buccolingual and mesiodistal anatomy were taken to verify the root structure. Root canals where the first file (size 20) fit snugly at the apex were considered.

To prepare the teeth, they were decoronated using a low-speed diamond disc with water cooling, resulting in a root length of approximately 16 mm. A size #10 K-file was inserted into the root canal to ensure it was visible at the apical foramen. For shaping the root canal, initially, a glide path was created using #25 K-file followed by a complete sequence of Protaper (Dentsply Maillefer, Ballaigues, Switzerland) rotary instrumentation from SX to F3 was used. The canals were irrigated with 2 ml of 5.25% sodium hypochlorite (NaOCl) using a 30-gauge needle held 2 mm short of the apex after each instrument. EDTA (RC Help, Prime Dental, India) was the lubricant used. Post instrumentation, the samples were dried using sterile paper points and then flushed for 1 minute with 5 ml of 17% EDTA (Dent Wash, Prime Dental, India), followed by 1 minute with 5 ml of 5.25% NaOCl. After complete drying of the root canal, intracanal medication $Ca(OH)_2$ was placed.

On a glass slab $Ca(OH)_2$ powder and normal saline were combined in a ratio of 1:1.5. The $Ca(OH)_2$ paste was inserted into the canal using a lentulospiral until it protruded from the canal opening. The procedure was repeated until the entire canal was filled. This was verified radiographically.

The specimens were maintained at 37°C with 100% relative humidity for one week after temporarily sealing the canal orifice. After a week, the specimens were divided into 4 groups based on irrigation agitation/ techniques used for intracanal calcium hydroxide removal. Each group further had 2 subgroups based on the chelating agent used.

Group 1: Irrigant Agitated using Master Apical File

ProTaper F3 rotary file, sized at #30 with a taper of 0.09 was used as the MAF at a constant speed of 300 rpm and a rotational force of 2.6 Ncm. $Ca(OH)_2$ was removed along the working length, with 5ml irrigant (5.25% NaOCl +10% Citric acid; 5.25% NaOCl +17 % EDTA) was used for one minute.

Group 2: Irrigant Agitated using Passive Ultrasonic Irrigation

The stainless steel IrriSafe with Suprasson P5 Booster ultrasonic unit (size #20/25 mm file; Satelec, Acteon, Merignac France) was used for irrigation. Its non-cutting file was used 1 mm short of the working length with five ml of irrigant (5.25% NaOCl +10% Citric acid; 5.25% NaOCl+17% EDTA) for one minute.

Group 3: Irrigant Agitated using CanalBrush

A CanalBrush with an apical diameter of 0.3mm and a speed of 600 rpm was used along with a handpiece and prepared till the working length. It was employed in a circular motion with five ml of irrigant (5.25% NaOCl +10% Citric acid; 5.25% NaOCl+17% EDTA) for one minute.

Group 4: Irrigant Agitated using F-file

In a controlled low-speed handpiece, the F-file was operated at a speed of 800 rpm passively in the canal in a circular motion along the dentinal walls with a cyclic axial motion (up and down) with five ml of irrigant (5.25% NaOCl +10% Citric acid; 5.25% NaOCl+17% EDTA) for one minute.

For all samples, the irrigation procedure calls for adding 5ml of 5.25% NaOCl for 60 seconds, followed by 60 seconds of agitation. After drying with sterile paper points, the next step is to add 5ml of 10% Citric acid/17% EDTA for 60 seconds, followed by 60 seconds of agitation. Finally, flushing it with 10 ml of saline to complete the process.

The grooves were made on both the outer (buccal) and inner (lingual) surfaces of the teeth using a water-cooled diamond bur. The teeth were bisected along their longitudinal axes, from the buccal side to the lingual side, using a surgical chisel. The processed samples were dehydrated and secured onto aluminum mounts, and a thin layer of gold through sputtering was coated. The samples were examined using a Scanning Electron Microscope (SEM) (JEOL, Tokyo, Japan). The apical third of the root canal (5 mm from the apex) was evaluated using 1000x magnification.

Three examiners after reliability calibration evaluated the cleanliness of the root canal walls using the following criteria: Score 0 indicated the absence of residues, Score 1 indicated minimal residues covering up to 20% of the surface, Score 2 represented moderate residues covering up to 60% of the surface, and Score 3 signified the presence of significant residues covering more than 60% of the surface.

The data was subjected to statistical analysis using nonparametric tests, the Kruskal-Wallis and Mann-Whitney tests, with a predetermined significance level of ($p<0.05$).

3. Results

The scores (cleanliness of the root canal walls criteria), and statistical comparisons of $Ca(OH)_2$ residues in the apical third of the root canal for each experimental group, utilizing two different chelating agents, are presented

Table 1. Scores and Mean Rank regarding the presence of $Ca(OH)_2$ Residues using 10% Citric Acid and 17% EDTA Chelating Agents

Agitation Technique	Scores				p-value
Group 1- Master Apical File					
(a) 10% Citric Acid	0	0	1	9	0.32
(b) 17% EDTA	0	0	0	10	
Group 2- Passive Ultrasonic Irrigation					
(a) 10% Citric Acid	0	5	5	0	0.66
(b) 17% EDTA	0	4	6	0	
Group 3- Canal Brush					
(a) 10% Citric Acid	0	0	2	8	0.54
(b) 17% EDTA	0	0	1	9	
Group 4- F File					
(a) 10% Citric Acid	0	4	5	1	0.7
(b) 17% EDTA	0	3	6	1	

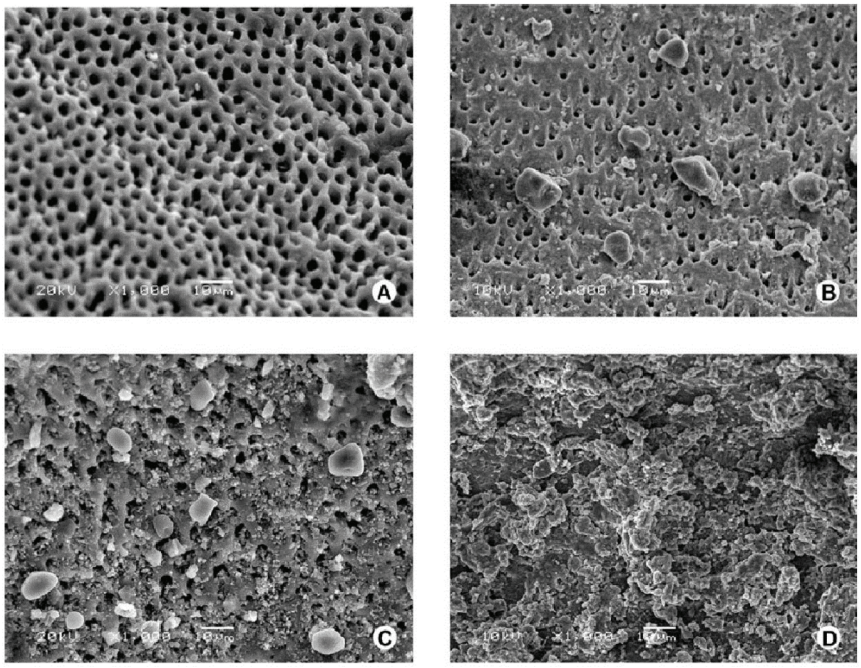


Figure 1. SEM images representative of scores attribution. **A** = Score 0: absence of residues; **B** = Score 1: small amount of residues (up to 20% of the surface covered), **C** = Score 2: moderate amount of residues (20% to 60% of the surface covered), and **D** = Score 3: large amount of residues (more than 60% of the surface covered).

in Table 1. $Ca(OH)_2$ dressing could not to eliminated by any technique. There was no statistically significant difference observed between the performance of 10% Citric Acid and 17% EDTA in each group (Figure 1). Passive Ultrasonic Irrigation and F File proved to be more effective than Canal Brush and Master Apical File in removing $Ca(OH)_2$ residues ($p<0.05$) regardless of the chelating agent utilized.

Table 2 represents the statistical comparison among the groups for each of the chelating agents. In the apical third, no difference was found ($p>0.05$) between the chelating agents, regardless of the agitation technique used. SEM images representative of $Ca(OH)_2$ residues using 10% Citric Acid and 17% EDTA for each of the agitation techniques are shown in Figure 2.

4. Discussion

Successful endodontic treatment consists of chemo-mechanical debridement and root canal cleaning, using the right intracanal medications and irrigating agents.[13] The commonly used substances are formocresol, cresatin, phenolic compounds, aldehydes, antibiotics, steroids, and calcium hydroxide[14].

$Ca(OH)_2$ has antimicrobial capability, tissue dissolving ability, and lipopolysaccharide degradation. It is also biocompatible[13]. The breakdown of calcium hydroxide paste into calcium and hydroxyl ions is important for its efficacy.[15] Regardless of the intended application, calcium hydroxide must be thoroughly removed from the root canal space before it is obturated[16]. Due to its interference with the adaption of different types of root canal sealers such as ZnOE, silicone, epoxy resin, and methacrylate resin-based sealers, incomplete removal of calcium hydroxide might have detrimental implications on the outcome of endodontic therapy. Calcium hydroxide remnants can lessen canal permeability by encouraging the production of calcium carbonate particles and interfering with endodontic sealers' capacity to effectively seal canals. The bonding abilities of these sealers to root canal dentine have also been observed to be diminished[17].

Therefore, this study was undertaken with an emphasis on diverse irrigant agitation techniques and irrigants aimed at achieving a more efficient and effective removal of calcium hydroxide from the root canal walls. Digital photos, Stereo microscopes, SEM, Micro-CT, and spiral CT were used to determine to assess the residues

Table 2. A comparative analysis between the groups to evaluate the presence of $Ca(OH)_2$ residues on the walls of root canals for each of the chelating agents

Chelating agents	Agitation Techniques	p-value
(a) 10% Citric Acid	G(MAF) vs. G(PUI)*	**0.0005***
	G(MAF) vs. G(CB)	0.542
	G(MAF) vs. G(FF)*	**0.001***
	G(PUI) vs. G(CB)*	**0.0005***
	G(PUI) vs. G(FF)	0.522
	G(CB) vs. G(FF)*	**0.001***
(b) 17% EDTA	G(MAF) vs. G(PUI)*	**0.0005***
	G(MAF) vs. G(CB)	0.317
	G(MAF) vs. G(FF)*	**0.0005***
	G(PUI) vs. G(CB)*	**0.0005***
	G(PUI) vs. G(FF)	0.483
	G(CB) vs. G(FF)*	**0.001***

*Indicates statistically significant difference ($p<0.05$)

in the canal[2,13,18-20]. In our investigation, root canal walls with calcium hydroxide residue were assessed using Kuga *et al.*'s established scoring standards while under 1,000x magnification utilizing scanning electron microscopy[21].

According to the SEM findings of the present study, the samples agitated using Passive Ultrasonic Irrigation and the F file showed enhanced calcium hydroxide removal from the apical third in comparison to the Master Apical File and Canal Brush. PUI works on the principle of transfer of energy from an ultrasonically oscillating instrument to the irrigation solution present within the root canal[5]. This technique's effectiveness in dislodging loose calcium hydroxide from the root canals can be attributed to the increased speed and volume of irrigant flow generates. The energy is conveyed through ultrasonic waves, giving rise to phenomena such as acoustic streaming and cavitation of the irrigant[13].

The purpose of the F-file is to agitate the irrigant and remove dentinal wall debris without further widening the canal. It represents an innovative rotary finishing file designed for single use, featuring a distinctive design with a diamond abrasive embedded in a non-toxic polymer[7]. When it is in axial motion (moving up and down), it displaces a larger volume of fluid compared to when it is stationary. This motion prompts rapid fluid flowing through the restricted space between the file and the canal wall[7]. This could be one of the factors contributing to F file's superior calcium hydroxide removal efficiency when compared to other irrigant agitation methods.

The current investigation demonstrated that CanalBrush and Master Apical File have lower efficacy of $Ca(OH)_2$ medicament elimination from the apical third of the root. The probable cause for its low efficiency is the lack of taper in the CanalBrush design. As a result, a parallel-form tool is compelled to spin into a root canal's tapering space. The bristle distortion may be severe in the narrowest portion of the cone or the apical third of the root canal, and its impact on the canal walls may cease quickly, leaving the apical third without further brush action[6]. This could be the reason why, as compared to

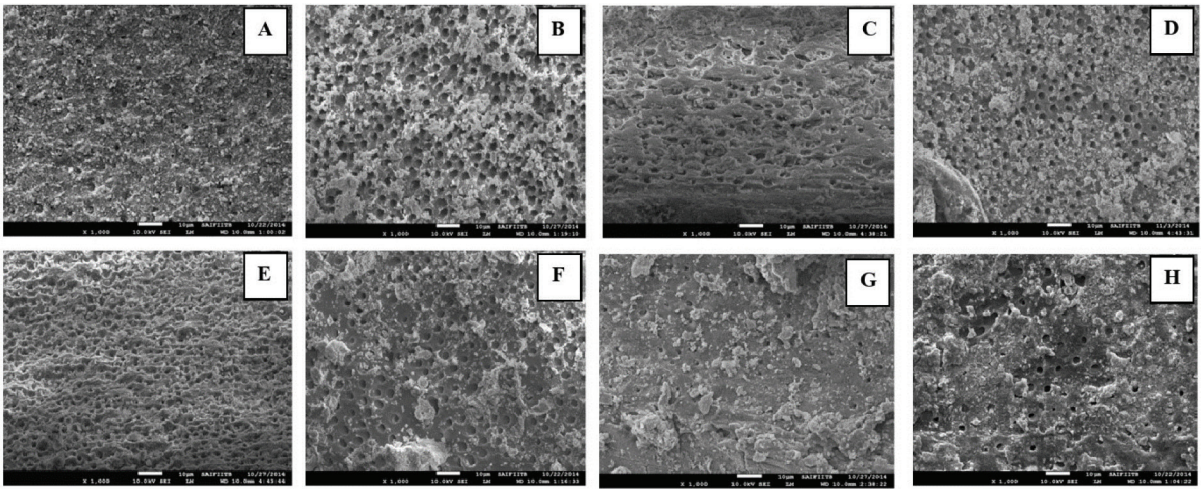


Figure 2. SEM micrographs representative of GI (**A** = 10% Crtic Acid; **E** = 17% EDTA), G2 (**B** = 10% Citric Acid; **F** = 17% EDTA), G3 (**C** = 10% Citric Acid; **G** = 17% EDTA) and G4 (**D** = 10% Citric Acid; **H** = EDTA).

the other irrigant agitation procedures, CanalBrush removed less calcium hydroxide from the canal. MAF uses mechanical instrumentation is ineffective and does not agitate the irrigant thoroughly[22].

The two most effective chelating agents, 10% citric acid and 17% EDTA were utilized in this investigation to remove calcium hydroxide[2,4,17-19,23]. The findings of this study suggest that 10% Citric acid had a somewhat higher effectiveness than 17% EDTA in calcium hydroxide removal from the root canal, however, this difference was insignificant.

The findings from this current study align with the outcomes of various in vitro studies conducted by S. Anitha Rao *et al.*[17], Nikhil Vineeta *et al.*[23], Lara Maalouf *et al.*[24]. Sevinç Aktemur Türker *et al.*[25] Fuat Ahmetoğlu *et al.*[26], Luciana Jorge Moraes Silva *et al.*[27], Ismail Davut Capar *et al.*[28], Anda Kfir *et al.*[29]. David Donnermeyer *et al.*[30], N.C. Tavella E Silva *et al.*[31] and Lu Shi *et al.*[32]. This study also concludes the higher calcium hydroxide medicament removal ability of Passive Ultrasonic Irrigation and F file when compared to CanalBrush and Master Apical File.

5. Conclusion

Complete removal of $Ca(OH)_2$ from the root canals could not be achieved using any irrigants or agitation methods. In the apical third of the root, PUI and F files proved to be more effective than MAF and CanalBrush in the removal of $Ca(OH)_2$. No significant difference was noted in the removal of $Ca(OH)_2$ while using 10% Citric Acid or 17% EDTA as chelating agents. It is crucial to note that the findings from this in vitro study should not be directly extrapolated to all clinical scenarios. Clinical outcomes can be influenced by various factors such as the quantity and duration of exposure to irrigating solutions, the type of calcium hydroxide vehicle used, and other pertinent variables.

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