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A Comparison of Two Techniques for the Removal of Calcium Hydroxide and Nano Calcium Hydroxide intracanal medicaments from Root canals: (In-Vitro Study)

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Abstract

Intracanal medicaments are recommended in root canal treatment in several clinical situations. However, complete removal of medicaments before the root canal filling is important because it may interfere with the ability of sealers penetration to the dentinal tubules. Aim**:** to evaluate the efficacy of two irrigation techniques in removing calcium hydroxide and nano calcium hydroxide intracanal medicaments from the root canal using a scanning electron microscope. Forty extracted single-rooted teeth were prepared using Protaper Gold rotary instruments up to a master apical file size F4. Teeth samples were divided randomly into two groups (n=20), according to the intracanal dressing. The root canals were filled with calcium hydroxide paste (Group I) and nano calcium hydroxide intracanal medicament paste (Group II). The removal of medicaments was performed after seven days with either conventional syringe irrigation technique (Subgroup A) or passive ultrasonic irrigation technique (Subgroup B) (n=10). Passive ultrasonic irrigation demonstrated the lowest mean values, while the conventional irrigation group exhibited significantly higher scores ($p<0.05$). When the root canal thirds were compared, the highest mean values were observed in the apical thirds. PUI was associated with significantly fewer medicament residues in the coronal, middle, and apical third compared to the conventional irrigation technique.

Keywords: Calcium hydroxide, nano calcium hydroxide removal, passive ultrasonic irrigation, irrigation, intracanal medicament.

Full-length article **Corresponding Author*, e-mail: *Ranatobar1992@gmail.com*

1. Introduction

The main goal of root canal treatment is to reduce or eliminate bacteria and their by-products from the root canal system [1]. Although instrumentation procedures have improved over the years, none of the existing techniques achieve a completely cleaned root canal system [2-3]. Therefore, the use of intracanal medicament has been advocated to enhance the disinfection process [4-5]. Calcium hydroxide [$Ca(OH)_2$] has been widely accepted as the most frequently used intracanal medicament owing to its good antimicrobial properties against the majority of endodontic pathogens, biocompatibility, tissue dissolving capability, potency to inhibit inflammation and osteoclastic activity, and ability to induce mineralized tissue formation and lipopolysaccharides inactivation [6]. Despite the mentioned benefits of $Ca(OH)_2$, it cannot inhibit several resistant pathogens [7]. Nano calcium hydroxide (NCH) particles are reported for higher levels of antimicrobial activity than conventional $Ca(OH)_2$ and deeper penetration into dentinal tubules [8-9].

All interappointment dressings must remove completely from the root canal before filling to avoid the negative interference between the medicament and the root filling material, which may increase apical leakage. It has been reported that residual $Ca(OH)_2$ on the root canal walls may increase apical leakage of gutta-percha root fillings when a zinc oxide eugenol sealer is used [10]. In addition, the intracanal medicament residues may change the physical properties of endodontic sealers, reducing the flow and setting time and preventing the penetration of sealers into dentinal tubules [11-12]. Thus, adequate removal of intracanal medicaments before obturation is mandatory

[13].Several techniques have been used to remove intracanal medicaments from the root canal system, including hand files [14], sonic activations [15], passive ultrasonic irrigation, and [16] nickel-titanium(NiTi) rotary instruments [17-18].

Mechanical instrumentation with a master apical file (MAF) and copious irrigation with sodium hypochlorite (NaOCl).and EDTA is the most frequently described method for removing medicaments from the root canal [19]. However, there is still no consensus regarding the best method for removing $Ca(OH)_2$. Continuous passive ultrasonic irrigation (PUI) uses an ultrasonically activated file inside the root canal with a continuous irrigant supply. It has suggested for optimizing intracanal medicament removal and considered one of the most universal and well-established irrigation methods. To date, several studies have been proposed to evaluate different techniques in removing $Ca(OH)_2$ intracanal medicament. Still, the literature has less signified the evaluation of various irrigation techniques in removing nano calcium hydroxide. Therefore, this scanning electron microscopic study aimed to evaluate the effectiveness of the different irrigation techniques in removing nano calcium hydroxide (NCH) and conventional calcium hydroxide $Ca(OH)_2$ from the root canal wall.

2. Materials and Methods

2.1. Ethical approval

The faculty of Dentistry at Misr International University's research ethics committee authorized the study with an approval number of 16/9/2020.

2.2. Inclusion criteria

Maxillary central incisors that were freshly extracted for periodontal reasons also with complete root formation and single canal with <10° curvature, no visible caries, fracture, cracks, resorption, and calcifications. Buccolingual and mesiodistal radiographs assessed canal morphology.

2.3. Exclusion criteria

Open apex tooth, teeth having more than one canal, fractures, crack lines, resorptive defects, and calcification.

2.4. Preparation of nano calcium hydroxide paste

Nano calcium hydroxide was prepared mechanically by particle size reduction attained through a ball miller, as Zhang W et al. described [20]. The powder was milled using a ball mill machine (planetary-ball-mill-pm—400) for ten h, speed 350 rpm, and 3 min intervals. Propylene glycol (PG) was used as a vehicle to improve the consistency of the paste.

2.5. Preparation of conventional calcium hydroxide paste

1.35 gm calcium hydroxide powder was added to 5 ml of Distilled water with sonication and stirring for one h to get a concentration of 27%w/v. Then, 0.5 gm of propylene glycol (PG) was sprinkled gently and gradually over the solution at a mild temperature until it reached a paste consistency.

2.6. Specimen preparation

Forty maxillary central incisors (n=40) under inclusion criteria were collected from the MIU teeth bank for this study. All teeth decontaminated by immersion in 5.25% NaOCl for 5 min, and then cleaned using an ultrasonic scalar to remove any surface deposits and calculus. Later, teeth were decorated at the level of cement-enamel junction using a saw under water coolant to obtain a standardized length of 15mm. After the access cavity's preparation, the tooth's height was measured by inserting a size 10 k file into the root canal until the tip of the instrument was visible only at the apical foramen. All root canals were prepared using Protaper Gold rotary files to size F4 master apical file (40, 0.02). After preparation, root canals were irrigated with a sequence of 5ml EDTA (17%), 5ml NaOCl (1%), and 5ml saline solution, then dried with paper points. Afterward, teeth samples were randomly allocated into two experimental groups (n=20) according to the intracanal dressing. In the first group, root canals were filled with calcium hydroxide paste (Group I) using Metapex tip. In contrast, root canals in the second group were filled with nano calcium hydroxide paste (Group II). All apices were sealed with sticky wax to simulate the clinical condition. The access cavities were sealed with a cotton pellet and temporary filling. Subsequently, the specimens were stored for seven days at 37°C and 100% humidity to simulate the clinical situation. After this period, each group classified into two subgroups according to a technique used to remove medicament.

2.7. Subgroup A (conventional irrigation) [n=10]

The removal of medicament performed with traditional irrigation using a plastic syringe with a 30-G needle. Sequentially, the canals were first rinsed with 1 mL 0.5% NaOCl, followed by the manual use of the last file used in the instrumentation procedure (MAF = F4) to the WL. The canals were irrigated again with 1mL 0.5% NaOCl and 3mL 17% EDTA for 60 seconds. A final rinse with 3 mL 0.5% NaOCl followed by saline ended the irrigation protocol. Generally, conventional irrigation standardized the needle depth for each canal by placing the rubber stopper 1–2 mm short of the WL.

2.8. Subgroup B (passive ultrasonic irrigation) [n=10]

The removal of medicament was performed with the ultrasonic continuous irrigation using ultrasonic file size 25 (Acteon Satelec). The needle was used in conjunction with a piezo-electric ultrasonic energy generating unit (Satelec, P5 Newtorn; Acteon, Merignac Cedex, France) to provide the energy for tip oscillation. Sequentially, the canals were first irrigated with 1mL 0.5%NaOCl followed by a continuous irrigation flow with 3 mL 17% EDTA that was maintained and agitated for 60 seconds using the ProUltra Piezo Flow device. In contrast, the high-volume suction was placed at the access opening to recover used irrigant. The ProUltra needle was inserted into the canal not greater than75%of the WL, as recommended by the manufacturer. The inactive needle was inserted into the canal to the premeasured depth, and the irrigant flow was started before activation of the suction mode. During activation, the needle was passively moved up-and-down to ensure it did not bind to the root canal wall. A final rinse with 3 mL 0.5% NaOCl followed by saline ended the irrigation protocol.

2.9. Scanning electron microscopic evaluation

Teeth samples of each subgroup after removing the intracanal medicaments were prepared for scanning electron microscope (n=10). The teeth samples were sectioned along their long axis in a buccolingual direction into two halves

using a hammer and chisel to prevent canal contamination. For scanning electron microscopic analysis, Teeth halves were individually dehydrated, fixed on aluminum stubs (Silverpoint: Agar Scientific Ltd, Stansted, Essex, UK), sputter coated with 20-nm platinum, and viewed with scanning electron microscope at magnification 1000x (Ultra55; Carl Zeiss NTS Gmbh, Oberkochen, Germany) to evaluate the amount of intracanal medicament on the canal walls. The sections were assessed independently using a 5-grade scale in a blind manner [21].

- Score 1: 80%–100% removal of $Ca(OH)_2$ (total cleanliness).
- Score 2: 60%–80% removal of $Ca(OH)_2$ (great cleanliness).
- Score 3: 40%–60% removal of $Ca(OH)_2$ (partial cleanliness).
- Score 4: 20%–40% removal of $Ca(OH)_2$ (light cleanliness).
- Score 5: 0%–20% removal of Ca(OH)2 (no cleanliness)

3. Results and discussion

3.1. Results

Statistical analysis and descriptive data between the groups and subgroups were illustrated in (Table 1, Table 2, Table 3, and Table 4. In addition, Figure 1. The residues of each intracanal medicament were evaluated using a 5-step scale method. Three different calibrated investigators examined and scored the microphotographs. Mean and standard deviation for remaining intracanal medicament scores calculated for each group and statistically evaluated for quantitative assessment of each intracanal medicament in the coronal, middle, and apical thirds. The low mean scale values represent fewer remnants on the dentinal tubules, while high mean scale values represent more remnants on the dentinal tubules. Friedman test was used to compare the root canal thirds, and the Whitney test was used to compare the two removal techniques, Conventional syringe irrigation (subgroup A) and Passive ultrasonic irrigation (subgroup B). When the root sections were compared, regardless of the irrigation technique. The lowest scale values observed in the coronal thirds and the highest in the apical thirds. (Table 1, Table 2, and Figure 1). According to the Friedman and Whitney test results, passive ultrasonic irrigation groups demonstrated the lowest scale values (total cleanliness) with a significant difference in both intracanal medicaments removal results than the conventional syringe irrigation groups. The traditional irrigation groups exhibited higher scores (large amounts of remnants). (Table 2, Table 3, and Figure 1).

3.2. Discussion

To our knowledge, no study has compared the effectiveness of conventional and passive ultrasonic irrigation in removing nano calcium hydroxide from the root canal system. Therefore, the main goal of our study was to compare the effectiveness of conventional needle irrigation and passive ultrasonic irrigation in combination with NaOCl and EDTA irrigant solutions in removing calcium hydroxide and nano calcium hydroxide intracanal medicaments from the root canal wall. Several methods used to measure remnants of the medicaments on the root canals such as direct visualization, digital microscope, scanning electron microscope, and volumetric analysis using cone beam computed tomography. In several studies, the removal efficiency of different techniques was assessed by the percentage ratio of medicament-coated surface area to the total canal surface area.

However, it evaluates only the superficial layer of the remnants and does not allow for three-dimensional evaluation. The present study used scanning electron microscopy at 1000x magnification to evaluate the intracanal medicament residues. Studies have reported that scanning electron microscope is a reliable method in examining and assessing the removal of intracanal medicaments from the root canal system [22-24]. In the present study, NaOCl and EDTA were used as irrigation solutions for the removal of intracanal medicaments, in agreement with the studies of Lee et al. [25] and Rodig et al. [26], which reported that the cleaning efficacy of an irrigation technique depends not only on the mechanical agitation and volume of the irrigation solution. Also on the chemical activity of the irrigant. The most commonly described method for removing intracanal medicaments is instrumentation, NaOCl, and EDTA irrigation solutions. However, it has been reported that removing intracanal medicaments from the apical root canal wall is inefficient and tedious when this method is used [27].

This could be explained as the instrumentation and irrigation alone cannot completely clean the entire canal wall, removing the intracanal medicaments only with a file and irrigation. Remnants will remain in canal extensions and root canal complexities. Therefore, irrigant agitation is the only way to remove the medicament residues from inaccessible areas [16]. During passive ultrasonic irrigation, acoustic streaming and cavitation occur, causing a specific streaming pattern within the root canal from the apical to the coronal. Micro cavitation (small voids) shaking solution inside the canal and improving the removal of the medicament residues as well as enhancing the penetration of the irrigant solutions liquid into the apical third of root canal system [28]. The results of the present study showed that passive ultrasonic irrigation with continuous irrigation was more effective in removing intracanal medicaments than conventional syringe irrigation. The SEM observations showed that the largest number of teeth with a score of one belongs to subgroup B (PUI).

Regarding the effect of subgroups, there was a significant difference between subgroup B (PUI) and subgroup A (CSI) in-group I (calcium hydroxide) and group II (nano calcium hydroxide) in all root canal thirds. PUI groups demonstrated the lowest scale values (cleanest canals). At same time, conventional syringe irrigation group exhibited significantly higher scores (high amounts of remnants). Similar to these findings, several previous studies showed that calcium hydroxide medicament removal was superior to passive ultrasonic irrigation compared with conventional syringe irrigation. A survey conducted by Tasdemir et al. [27] showed ultrasonic agitation of NaOCl left significantly less calcium hydroxide than an irrigant flush alone. In addition, Silva et al. [29] showed that use of PUI was associated with fewer residues compared to rotary file and control group. Using an additional file did not lead to better removal of the medicament. A possible explanation for our results is higher velocity and volume of irrigant flow created by PUI, which enhances its efficiency in flushing out loose medicament remnants from the root canal walls.

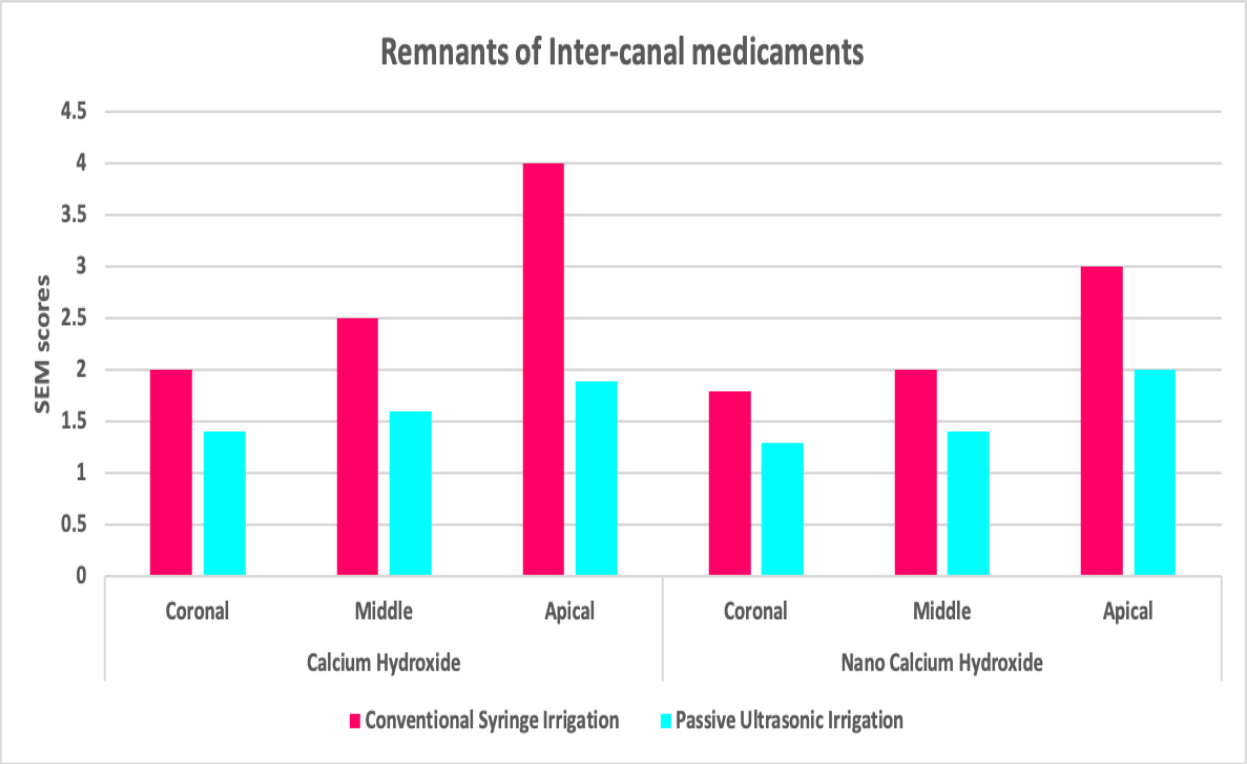

Figure 1: Bar chart comparing the two irrigation techniques (CSI and PUI) in removing the two-intracanal medicaments (Calcium hydroxide and Nano calcium hydroxide) in different root canal thirds.

Table 1: The mean and standard deviation (SD) values of the Conventional syringe irrigation technique (subgroup A) in the removal of calcium hydroxide and Nano calcium hydroxide medicaments in different root canal thirds (Friedman test).

Variables	Conventional Syringe Irrigation						P value
	Calcium Hydroxide			Nano Calcium Hydroxide			
	Mean	SD	Median	Mean	SD	Median	
Coronal	2.00 [b]	0.47	2	1.80 [b]	0.42	2	0.329ns
Middle	2.50 [b]	0.97	2	2.00 [b]	0.47	2	0.197ns
Apical	4.00 [a]	0.67	4	3.00 [a]	0.94	3	0.140ns
p-value	0.001*			<0.001*			

Means with different letters in the same column indicate statistically significant difference *; significant (p<0.05). ns; non-significant (p>0.05).

Table 2: The mean and standard deviation (SD) values of passive ultrasonic irrigation technique (Subgroup B) in the removal of calcium hydroxide and Nano calcium hydroxide medicaments in different root canal thirds (Friedman test).

Variables	Passive Ultrasonic Irrigation						P value
	Calcium Hydroxide			Nano Calcium Hydroxide			
	Mean	SD	Median	Mean	SD	Median	
Coronal	1.40 [a]	0.52	1	1.30 [a]	0.48	1	0.648ns
Middle	1.60 [a]	0.84	1	1.40 [a]	0.52	1	0.728ns
Apical	1.90 [a]	0.57	2	2.00 [a]	0.94	2	0.808ns
p-value	0.223ns			0.050ns			

Means with different letters in the same column indicate a statistically significant difference (p<0.05). ns; non-significant (p>0.05).

Table 3: The mean and standard deviation values between conventional syringe irrigation and passive ultrasonic irrigation techniques in the calcium hydroxide group (Mann Whitney test).

Variables	Calcium Hydroxide								
	Coronal			Middle			Apical		
	Mean	SD	Median	Mean	SD	Median	Mean	SD	Median
Conventional Syringe Irrigation	2.00 [a]	0.47	2	2.50 [a]	0.97	2	4.00 [a]	0.67	4
Passive ultrasonic irrigation	1.40 [b]	0.52	1	1.60 [b]	0.84	1	1.90 [b]	0.57	2
p-value	0.018*			0.039*			<0.001*		

Means with different letters in the same column indicate statistically significant difference *; significant ($p<0.05$).

Table 4: The mean and standard deviation between conventional syringe irrigation and passive ultrasonic irrigation techniques in the nano calcium hydroxide group (Mann Whitney test).

Variables	Nano Calcium Hydroxide								
	Coronal			Middle			Apical		
	Mean	SD	Median	Mean	SD	Median	Mean	SD	Median
Conventional Syringe irrigation	1.80 [a]	0.42	2	2.00 [a]	0.47	2	3.00 [a]	0.94	3
Passive ultrasonic irrigation	1.30 [b]	0.48	1	1.40 [b]	0.52	1	2.00 [b]	0.94	2
p-value	0.028*			0.018*			0.045*		

Means with different letters in the same column indicate statistically significant difference *; significant ($p<0.05$).

The efficiency of passive ultrasonic depends on the activation duration and is enhanced with fresh irrigant replacement [30]. Conversely, the flushing action from syringe irrigation is relatively weak and dependent not only on the anatomy of the root canal but also on the depth of placement and the diameter of the needle [16]. PUI is based on the energy transmission from an ultrasonically oscillating instrument to the irrigant in the root canal. During PUI, acoustic microstreaming and cavitation can occur, which causes a streaming pattern within the root canal from the apical to the coronal. Because of this microstreaming, more dentin debris and intracanal medicament remnants can be removed from the root canal than from the syringe delivery of the irrigant, even from remote places in the root canal. When the root thirds were compared, regardless of the removal technique, the lowest mean scale values were observed in the coronal thirds. The middle thirds exhibited higher mean values compared with the coronal thirds. The highest mean values were observed in the apical thirds.

By several previous studies, remnants of intracanal medicaments were found in the apical region regardless of the medicament used [17-22-29-31]. A survey conducted by Salgado et al. [15] reported that regarding the root canal sections, the results of the apical third were typically worse than the coronal and middle due to the difficulty of cleaning this region. In line with the results of the aforementioned study, Nandini et al. [13] revealed that the remaining intracanal medicament was found to be packed apically, mainly of about 2-3mm. This could be attributed to the normal anatomical morphology of the conical root canal system. The larger coronal diameter as compared with the middle and apical diameters facilitates the irrigation and remnants removal from the coronal third. Moreover, the canal medicament residues tend to accumulate apically during the removal procedure, especially when apical anatomical variations are present; in addition, the limited insertion of the irrigation cannula and the ultrasonic tip being placed 2 mm short of the working length leaves this area without the direct effect of irrigant solution [17].

A study conducted by Teixeira et al. [32] demonstrated that the cleaning efficacy of irrigants had been effective mostly on the cervical and middle thirds compared to the apical third. The authors explained that the size of the root canal in the coronal and middle thirds allowed for better circulation and action of irrigating solution making the complete removal of debris possible. While in the apical third, the reduced dimension of root canal make air bubbles frequently remain trapped and prevent total filling with the irrigant that leads to inadequate removal of debris at this region in spite of the irrigating needle going as deep as mm short of the working length. In our study, considering the apical root third, the mean values of the groups in which PUI was used for the removal of medicament were significantly lower than the groups in which CSI was used. Our study showed that ultrasonically activated irrigation was superior to syringe irrigation. In accordance with Yücel et al. [22] who showed that PUI created a much cleaner apical area compared with conventional needle irrigation.

Also, knee et al.17 stated that there was a tendency for the debris to be located at the apical third and showed that

irrigants alone (NaOCl+EDTA) could not penetrate well into the apical third, while mechanical means of removal (PUI) showed significantly better results due to the acoustic streaming effect. The vibration of the ultrasonic insert produces an acoustic stream that generates a shear stress that is able to dislocate the debris inside the instrumented root canal system. The ultrasonic file vibrates at a frequency of 25-30 kHz, creating micro cavitation (small voids), which are able to propagate inside the irrigant solution and improve the penetration of the liquid into the apical third of the root canal system. The results of the current study showed that no difference found between calcium hydroxide and nano calcium hydroxide in terms of paste removal. Although the nano-size particles of NCH might be easily dislodged during irrigation, they may penetrate deep into the canal complexities (lateral canals and isthmuses). Conventional needle irrigation was insufficient to remove NCH and $Ca(OH)_2$ from the root canal system. Passive ultrasonic irrigation systems improved the removal of both medicaments, resulting in cleaner root canal walls.

4. Conclusion

Compared to conventional irrigation, PUI was associated with significantly fewer medicament residues in the coronal, middle, and apical third. Due to canal morphology, all groups tended to locate debris in the apical third. The limited insertion of the irrigation cannula and the ultrasonic tip, placed 2 mm short of the working length, left this area without the direct effect of the irrigant solution. Irrespective of the irrigation technique, no significant difference found between calcium hydroxide and nano calcium hydroxide regarding paste removal. The root canal third and the removal technique were the only significant factors.

Conflict of interest

The authors deny any conflict of interest related to this study.

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