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Research Article

RETRIEVABILITY OF CALCIUM HYDROXIDE INTRACANAL MEDICAMENT WITH THREE DIFFERENT CHELATORS FROM ROOT CANAL: AN IN VITRO CBCT

Yogesh Ahirrao., Sana Mohani*., Sunanda Gaddalay., Anita Kale., Sonali Gite., Praveen Dhore and Abhishek Badade

Department of Conservative Dentistry, MIDSR Dental College, Vishwanathpuram, Ambajogai Road, Latur

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A B S T R A C T

Aim: This study compared amount of aqueous-based and oil-based calcium hydroxide [Ca(OH)2] remaining in the canal after removal with three different chelators 17% EDTA, 0.2% Chitosan and 7% Maleic acid in combination with sonic agitation.

Materials and Method: Cleaning and shaping of root canals of 30 single rooted premolar was done and canals were filled either with oil based $Ca(OH)_2$ or aqueous- based $Ca(OH)_2$. Volumetric analysis was performed utilizing cone beam-computed tomography (CBCT) after 7 days of incubation. $Ca(OH)_2$ was removed using either 17% EDTA , 0.2% Chitosan and 7% Maleic acid in combination with sonic agitation. Volumetric analysis was repeated and percentage difference was calculated and statistically analysed by one-way ANOVA and post-hoc tukey test.

Results: All the three chelators failed to remove aqueous-based as well as oil-based $Ca(OH)_2$ completely from the root canal. Aqueous-based $Ca(OH)_2$ was easier to be removed than oil-based $Ca(OH)_2$.0.2% Chitosan, 7%Maleic acid and 17% EDTA were ound to remove the aqueous based calcium hydroxide efficiently ($P > 0.05$), whereas 0.2% chitosan was found to perform significantly better than 7% Maleic acid followed by 17% EDTA in removing oil based calcium hydroxide preparation.($P<0.05$)

Conclusion: Combination of 0.2% Chitosan and agitation results in lower amount of $Ca(OH)_2$ remnants than 7% Maleic acid followed by 17% EDTA irrespective of type of vehicle present in the mix.

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INTRODUCTION

The presence of microrganism within the root canal plays important role in causing endodontic infections [1].The main aim of root canal therapy is getting a microbial diminution and elimination of their by products from the root canal[2]. However, biomechanical preparation cannot completely eliminate the microorganism from the root canal[3]. Hence, the use of intracanal medicaments is necessary.[4]

Calcium hydroxide $Ca(OH)_2$ is the most popular intracanal medicament introduced by Herman in 1930[5]. It has good antimicrobial property which is used to achieve disinfection of root canals and many other goals such as healing periapical inflammation, arresting inflammatory root resorption, root fractures and preventing the reinfection of the root canal system through the interappointment period[6-8].

$Ca(OH)_2$ medicament that has been applied to the root canal should be removed before obturation. Any $Ca(OH)_2$ residue remain on the root canal walls negatively affects the quality of obturation. *In vitro* studies have shown that any $Ca(OH)_2$ residue can hinder the penetration of sealers into the dentinal tubules, hinder the bonding of resin sealer adhesion to the dentin, increase the apical leakage of root canal treated teeth, and potentially interact with zinc oxide-eugenol sealers which make them granular and brittle.[9-10]

Thus, complete and predictable removal of $Ca(OH)_2$ before obturation is necessary and several methods such as ultrasonic, sonic, canal brush, etc. and irrigating agents including NaOCl, EDTA, their combination, maleic acid, etc. have been used for removal of $Ca(OH)_2$ from root canal.[11-14] The mechanical agitation provided by ultrasonic(Passive Ultrasonic Irrigation) and sonic (Endoactivator) instrumentation or a rotary file together with irrigation enhance the removal of CH from the root canal wall. However, there is no general consensus among the researchers regarding the best method for the removal of Calcium hydroxide.[15]

Maleic acid with concentrations of 5% and 7% can be used as alternatives of 17% EDTA for removal of smear layer. It has anti‑microbial activity against *E. faecalis*, is less cytotoxic than EDTA, and effectively remove smear layer than 17%EDTA[16-18].Chitosan, a natural polysaccharide prepared by the deacetylation of chitin. It is endowed with properties of

Corresponding author:* **Sana Mohani
Department of Conservative Dentistry, MIDSR Dental College, Vishwanathpuram, Ambajogai Road, Latur

biocompatibility, biodegradability, bio-adhesion, and cytoxicity. It also has high chelating capacity for different metal ions in acidic condition which helps to remove smear layer and used in dentistry as a barrier membrane for periodontal therapy and as oral mucosal delivery agent for chlorhexidine.[19]

Various methods have been employed to measure the remaining calcium hydroxide residues in the root canal such as stereomicroscopy, scanning electron microscopy, spiral computed tomography (CT), etc.[11] Recently, cone beam computed tomography (CBCT) has been found to be more superior. In this method, three‑dimensional volume measurements are possible without sectioning the specimens. It is more faster and accurate which avoid loss of the specimen during process.

The aim of this *in vitro* study was to assess the efficiency of three calcium chelators, 17% EDTA solution, 0.2% Chitosan and 7% Maleic acid in combination with sonic agitation, in the removal of $Ca(OH)_2$ when mixed with two different vehicles. The volume of Ca(OH)2 removed was analyzed with cone beam computed tomography (CBCT).

MATERIALS AND METHOD

Thirty extracted single rooted mandibular premolars free of cracks, fracture or any other defects were selected. Access preparation was done and the root canals were subjected to biomechanical prepration with the crown down technique using ProTaper files (Densply-Mailiefer, Ballaigues, Switzerland) till F4. 2 ml of 5.25% NaOCl was used as an irrigant after each instrument and 5ml of 17% EDTA for final flush. Canals were dried with paper points (Densply-Mailiefer, Ballaigues, Switzerland).

Two formulations of $Ca(OH)_2$ with different vehicles were selected for the study. Metapex (Meta Dental Corp. Ltd., Elmburst, NY), a commercially available product is composed $Ca(OH)_2$, silicone oil, and iodoform. RC Cal(PRIME DENTAL PRODUCTS PVT LTD) is a formulation of calcium hydroxide and barium sulphate in ready to use radioapaque water soluble paste form.

The teeth were divided into two groups of 15 teeth each.

Group1: Metapex (Oil based $Ca(OH)_2$) was injected into the root canal until the material extruded through the apex.
Group 2: Rc Cal (Water based $Ca(OH)_2$) was injected into the canals until material extruded through the apex.

Teeth were kept in wet sponge during placement of $Ca(OH)_2$ formulations. Excess material was wiped off with moist cotton. The access cavities were temporarily sealed with a cotton pellet and Cavit. Teeth were stored at 37°C and 100% relative humidity for 7 days. Subsequently the teeth were mounted in a modeling wax for CBCT analysis.

After CBCT analysis,the volume of the filled material in each tooth were estimated in coronal section using On Demand 3D software (Cybermed inc. Korea). The teeth in each group were kept in wet sponge and further randomly divided into three subgroups on the basis of chelators used for retrieval of calcium hydroxide. 30-G endodontic needle was used in up-down motion for irrigation at 2 mm from the working length. Sonic agitation was done with Endo Activator (Denstply Sirona) with size of #25/0.04 taper for 1min.

The teeth in each group were further randomly divided into three subgroups on the basis of chealators used for removal.

Group 1a (n = 5): Metapex retrieved with 1 ml of 17% EDTA + sonic agitation for 1 min + final rinse with 1ml of distilled water.
Group 1b (n = 5): Metapex retrieved with 1 ml of 0.2%Chitosan+ sonic agitation for 1min + final rinse with 1ml distilled water.
Group 1c (n = 5): Metapex retrieved with 1 ml of 7% Maleic acid+ sonic agitation agitation for 1 min + final rinse with 1ml of distilled water.
Group 2a (n = 5):RC Cal retrieved with 1 ml of 17% EDTA+ sonic agitation for 1 min + final rinse with 1 ml of distilled water.
Group 2b (n = 5): RC Cal retrieved with 1 ml 0.2% Chitosan +sonic agitation for 1 min + final rinse with 1 ml of distilled water.
Group 2c (n = 5): RC Cal retrieved with 1 ml of 0.2% Chitosan solution + sonic agitation 1 min + final rinse with 1ml of distilled water.

For the preparation of 0.2% Chitosan solution, 0.2 g of Chitosan was diluted with 100 ml of 1% acetic acid and the mixture was stirred for 2 hour using a magnetic stirrer. The pH of was adjusted to 3.2 using digital ph meter. A second CBCT was done and the volume of remaining material in each tooth was estimated as before.

Outcome assessment

The calculation of $Ca(OH)_2$ volume for each specimen was done using OnDemand 3D software (Cybermed Inc.,Korea). Each dataset was segmented using a uniform grayscale threshold (OnDemand 3D software) to visualize and calculate the volume of remaining Ca(OH)2 material. The removal efficiency was calculated as [(a-b) 100/a], where "a" was the volume of material packed in the root canal and "b" was the volume remaining after retrieval. Volumes of $Ca(OH)_2$ were expressed as cubic mm. The data was statistically analyzed by one way ANOVA and post hoc tukey test.

RESULTS

All the three chelators (0.2% Chitosan,7% Maleic acid and 17% EDTA) removed the aqueous-based $Ca(OH)_2$ significantly better ($P < 0.05$) than oil-based $Ca(OH)_2$. Aqueous based $Ca(OH)_2$ was easier to remove than oil based $Ca(OH)_2$.

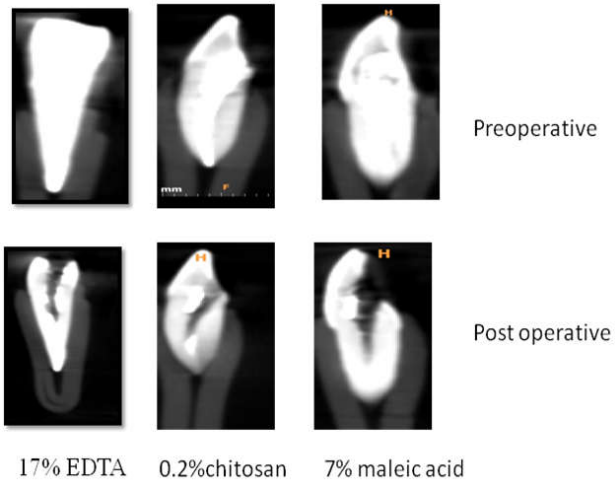

Figure 1 CBCT Images of Metapex (Oil based Calcium hydroxide)

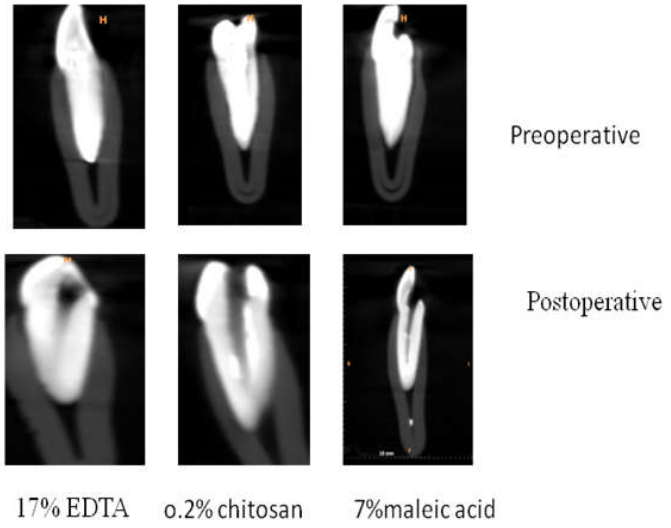

Figure 2 CBCT Images of RC Cal (Water based Calcium hydroxide)

The mean percentage of oil-based Ca $(OH)_2$ removed by 0.2% Chitosan (87.06 ± 1.68) was significantly better than 7% maleic acid (77.6% ±1.15) and 17% EDTA (62.18±1.16). 0.2% chitosan was found to perform significantly better than 7% Maleic acid and 17% EDTA in removing oil-based $Ca(OH)_2$ preparation ($P<0.05$). 7% Maleic acid perform significantly better than 17% EDTA in removing oil based Ca $(OH)_2$ ($P<0.05$).

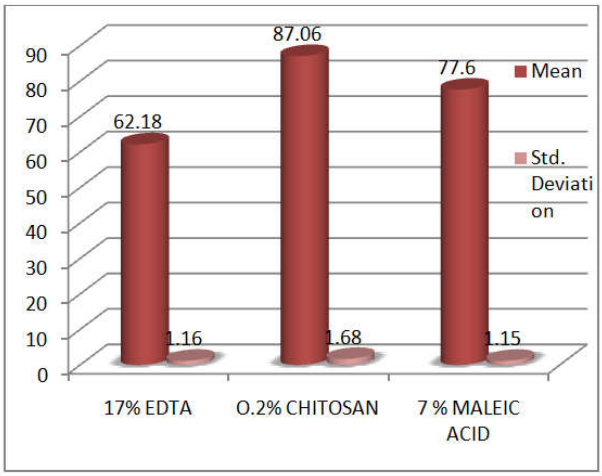

Table 1 Comparison of percentage of Metapex (Oil based Calcium hydroxide) retrieval using three calcium chelators

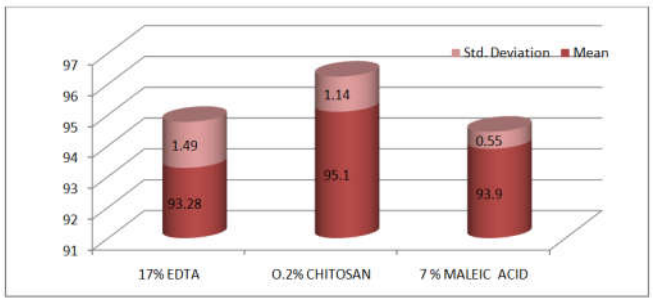

Table 2 Comparison of percentage of RC Cal (Water based Calcium hydroxide) retrieval using three calcium chelators

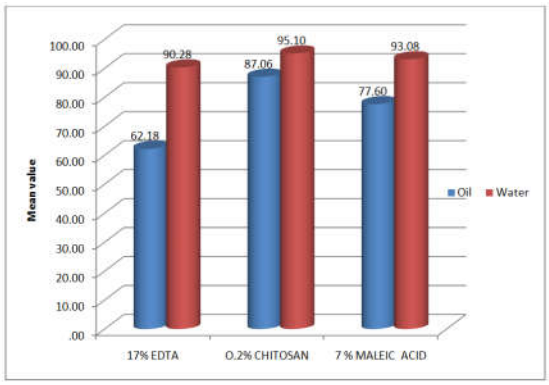

Table 3 Comparison between Metapex (Oil based)and RC Cal (Water based) calcium hydroxide retrieval using three different chelators

The mean percentage of aqueous-based $Ca(OH)_2$ removed by 0.2% Chitosan (95.1±1.14) show no significant difference from 7% maleic acid (93.9±0.55) and 17% EDTA (93.28± 1.49). All the three chealators show no significant difference in removing aqueous based $Ca(OH)_2$ ($P>0.05$) and they were equally efficient in removing aqueous based $Ca(OH)_2$.

DISCUSSION

Intracanal medicaments have been recommended with the goal to eliminate bacteria from the root canal, prevent bacterial proliferation between appointments, and to act as physiochemical barriers preventing root canal reinfection[20]. The vehicle used to mix calcium hydroxide paste has an influencing factor in the complete retrieval root canal.[21]The differences in velocity of ionic dissolution is directly related to the vehicle used. The lower the viscosity of paste higher is its ionic dissolution.[22]

Oily vehicles are non water soluble substances that promote the lowest solubility of the paste within the tissues, and residues of pastes containing this vehicle may remain within the root canal for a longer duration than the pastes containing aqueous or viscous vehicles and they are difficult to remove than aqueous based.[23]Hence, in this study, removal of silicone oil-based $Ca(OH)_2$ efficacy was investigated using different irrigants.

Removal of $Ca(OH)_2$ is accomplished through several irrigants including saline, NaOCl, several chelating agents such as EDTA, citric acid, maleic acid, EDTA-T and combination of NaOCl and EDTA.. Silva *et al*[24] concluded in their study that 0.2% Chitosan provided best results in relation to smear layer removal when compared with different concentrations of chitosan. Hence we used 0.2% Chitosan in our study. 0.2% Chitosan had also shown similar chelation activity as15% of EDTA and 10% citric acid.

The removal of calcium hydroxide has been investigated using different techniques. Canal irregularities may be inaccessible for conventional irrigation procedures, and calcium hydroxide may remain in these extensions. The other techniques like sonic agitation can be explained by the fact that the higher velocity and volume of irrigant flow can be achieved. The Endoactivator uses sonic energy to irrigate root canal. The main function of the Endoactivator is to produce vigorous intracanal fluid agitation through its swirling movement and cavitation. This hydrodynamic mechanism of activation serves to improve the penetration, circulation, and flow of irrigant

into the difficult-to-reach areas of the root canal system. Hence we have used Endoactivator in our study.[22]

Various studies conducted to analyze the removal of intracanal $Ca(OH)_2$ consisted two-dimensional imaging obtained after longitudinal sectioning to measure the surface area covered with the remaining intracanal medicament .In the current study, volume analysis was done with CBCT as it provides accurate results than surface area measurement. Advantages of the CBCT technique are a lower radiation dose, a shorter time and no loss of tooth specimen.[25]Nandani *et al.,*[11] Ballal*et al.*[26] and Wiseman *et al*.[27] used computed tomography.

The results of the current study demonstrate that none of the chelating agent were able to remove the $Ca(OH)_2$ completely irrespective to the type of vehicle used This is in accordance to the Nandini *et al.,*[11]Kenee *et al*.,[28] Kontakiotis *et al.*[29] and in contrast to De Faria *et al*.[30] Majority of the $Ca(OH)_2$ was found to be retained in apical third region which substantiate the findings of Nandini *et al.*[11] However, the presence of calcium hydroxide residual act as an apical barrier and advocated for it prolonged antimicrobial activity. Nevertheless, it is preferable to remove CH from apical third because of possibility to increase apical leakage when contacted with tissue fluids .[31]

In present study all the three chelators 17% EDTA, 0.2% Chitosan and 7% Maleic acid removed aqueous-based $Ca(OH)_2$ more effectively than oil-based $Ca(OH)_2$. Silicone oil, which was the oily vehicle present, might have resisted its dissolution and removal from the root canal. However, 0.2% Chitosan performed better than 7% Maleic acid followed by 17% EDTA solution in removal of oil-based $Ca(OH)_2$. This could be because of improved penetration of 0.2% Chitosan into the silicone oil and causing chelation of calcium ion in water. Silva *et al.* demonstrated in their study that the chelation effect of Chitosan is due to its own properties rather than because of 1% acetic acid which was used during preparation of 0.2% Chitosan[32].

CONCLUSION

- Within limitation of study, none of the chelators in combination with sonic agitation was able to completely remove the calcium hydroxide.
- 0.2% Chitosan, 17% EDTA and 7% Maleic acid were found to remove the aqueous-based calcium hydroxide efficiently.
- 0.2% Chitosan was found to perform better than 7% Maleic acid followed by 17% EDTA in removing oil-based calcium hydroxide preparations.
- Combination of sonic agitation with chelator results in cleaner canal for both aqueous-based, as well as oil-based $Ca(OH)_2$.
- The vehicle used to prepare calcium hydroxide influences its retrieval.
- Oil-based calcium hydroxide is more difficult to remove than aqueous based calcium hydroxide.
- Regardless of the vehicle used in the present study, remnants were found mainly in the apical region and also on the root canal walls.

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