



Comparative Evaluation of Metapex Removal Using Chelating Agents with and Without Ultrasonic Activation -An in-vitro CBCT volumetric analysis.

1. Basawaraj Biradar, Professor, Department of Conservative Dentistry and Endodontics, Rural Dental College, PIMS, Loni, Ahilyanagar, Maharashtra-India
2. Sakshi Gandhi, Postgraduate student, Department of Conservative Dentistry and Endodontics, Rural Dental College, PIMS, Loni, Ahilyanagar, Maharashtra-India
3. Aparna Palekar, HOD and Professor, Department of Conservative Dentistry and Endodontics, Rural Dental College, PIMS, Loni, Ahilyanagar, Maharashtra-India
4. Mukund.V.Singh, Professor, Department of Conservative Dentistry and Endodontics, Rural Dental College, PIMS, Loni, Ahilyanagar, Maharashtra-India

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| KEYWORDS | ABSTRACT: |
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| Metapex, Chitosan, ethylenediaminetetraacetic acid, Cone-beam computed tomography, Ultrasonic activation | <p>BACKGROUND: Effective removal of intracanal medicaments is essential before obturation to ensure optimal sealer adhesion and treatment success.</p> <p>AIM: Comparatively evaluate the effectiveness of 17% EDTA and 0.2% chitosan in removing Metapex intracanal medicament with and without ultrasonic activation.</p> <p>MATERIAL & METHODS: Forty single-rooted extracted human teeth were decoronated and instrumented using standard rotary techniques. In this in vitro study, root canals were filled with Metapex intracanal medicament, sealed with Cavit, and stored at 37 °C with 100% humidity for 7 days. CBCT imaging was performed after embedding samples in wax rims to simulate soft tissue, and volumetric analysis of medicament was carried out using software. Samples were then allocated into four subgroups for medicament removal: (A1) 0.2% chitosan with ultrasonic activation, (A2) 17% EDTA with ultrasonic activation, (A3) 0.2% chitosan without activation, and (A4) 17% EDTA without activation. A second CBCT will be done and the volume of remaining material in each tooth was estimated as before.</p> <p>RESULTS Post-hoc analysis showed that both 0.2% Chitosan and 17% EDTA demonstrated significantly greater Metapex removal when used with ultrasonic activation compared to when used alone ($p < 0.001$). No significant difference was observed between the two irrigants when ultrasonically activated ($p = 1.000$), indicating that ultrasonic activation, rather than the type of chelating agent, was the key factor in enhancing medicament removal.</p> <p>CONCLUSION</p> |



Ultrasonic activation significantly enhances the removal of Metapex compared to conventional irrigation alone. The choice of chelating agent becomes less critical when activation is used, highlighting ultrasonic activation as the key determinant in effective intracanal medicament removal.

Introduction

Microorganisms are the main causative agents, in pulp and periradicular diseases.^[1] Chemomechanical preparation can greatly clean the root canal system, but it cannot totally rid it of microorganisms.^[2] Intracanal medications are mainly recommended to remove any remaining bacteria in between treatments and to serve as a physiochemical barrier.^[3]

Complete removal of the medication from the root canals is required prior to obturation because any remaining medication may impair the root canals' ability to seal.^[4] For the best root canal obturation, intracanal medications must be totally removed prior to the last filling because any remaining medication can stick to the dentinal wall and prevent root canal sealers from penetrating and adhering, which could compromise the effectiveness of treatment.^[5,6] Copious irrigation with sodium hypochlorite and EDTA is commonly used for medicament removal; however, it is often inadequate for complete elimination. Therefore, additional chelating agents like EDTA, chitosan, and citric acid are incorporated into standard irrigation protocols to enhance effectiveness.^[7]

The use of chelating agents and other irrigation techniques have been investigated to improve the clearance of intracanal medications. While chitosan, a naturally occurring biopolymer, has drawn interest because of its chelating and antibacterial qualities, ethylenediaminetetraacetic acid (EDTA) is extensively utilized because it can demineralize dentin and make debris removal easier.^[8,9] Additionally, ultrasonic activation (UA) has been shown to improve the effectiveness of irrigants by increasing their penetration and agitation within the root canal system.^[10]

The effectiveness of medicament removal can be assessed using cone-beam computed tomography (CBCT), which provides high-resolution three-dimensional imaging for volumetric analysis of residual materials. This *in vitro* study aims to evaluate and compare the removal efficiency of metapex intracanal medicaments using two calcium chelators (chitosan and

EDTA), with and without ultrasonic activation, through CBCT volumetric analysis.

MATERIALS AND METHODS:

Sample Preparation:

For this study, forty freshly extracted single-rooted permanent teeth were chosen. [Fig 1] Teeth that exhibited additional canals, fractures, significant canal curvature, or signs of internal and external resorption, as identified through radiographic evaluation, were excluded. The selected teeth were carefully cleaned to remove any tissue remnants and debris using a scaler, disinfected in 5.25% sodium hypochlorite for one hour, and then stored in saline for later use. The teeth were then decoronated using a diamond disk, and the length of each tooth was standardized to 14 mm.

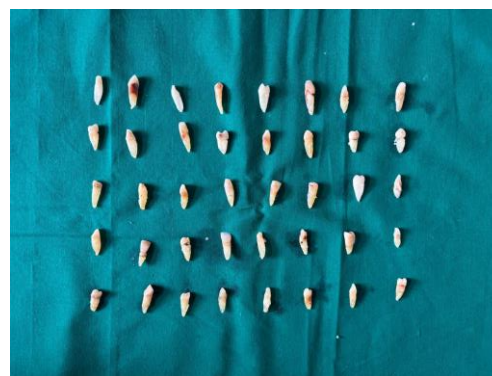


Fig 1: Single rooted sample teeth

METHODOLOGY

- Root canals were negotiated with a #10 K-file (Mani, Japan) until the tip was visualized at the apical foramen. The working length was determined radiographically at 1 mm short of the apical foramen using a K-file. The root canals were then instrumented up to size #30 with a taper of 0.06 using Hero Gold rotary files. [Fig 2] After each change of file, the canals were lubricated with 17% ethylenediaminetetraacetic acid (EDTA) gel (PRIME RC Help), and irrigation was performed with 10 mL of 3% sodium hypochlorite solution using a 27-gauge needle (DISPO VAN).

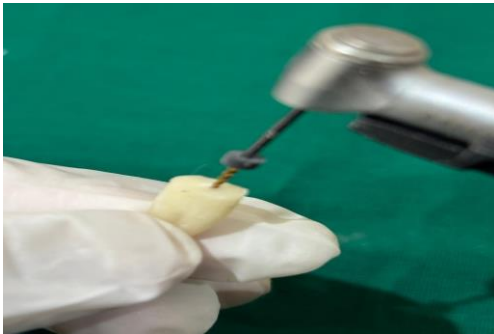


Fig 2 : Root canal Preparation

- The irrigation needle was inserted as deep as possible into the canal without binding. A final irrigation with 10 mL of 17% EDTA solution was performed in all canals to remove the smear layer. The canals were then rinsed with distilled water to eliminate any residual chemicals. Finally, the canals were dried using paper points (Dentsply-Maillefer, Ballaigues, Switzerland).

The samples were then filled with Metapex intracanal medicament.[Fig 3,4]Excess material was removed using wet cotton.A cotton pellet was placed in the access cavity, and the cavity was sealed with a temporary restorative material Cavit (3M).



Fig 3: Metapex Intracanal Medicament



Fig 4: Placement of Metapex in root canal

The samples were subsequently stored in an incubator at 37°C and 100% relative humidity for 7 days.For CBCT imaging, the teeth were randomly embedded in U-shaped wax rims, with 2 mm of modeling wax placed around each tooth to simulate soft tissue and improve image contrast.[Fig 5]

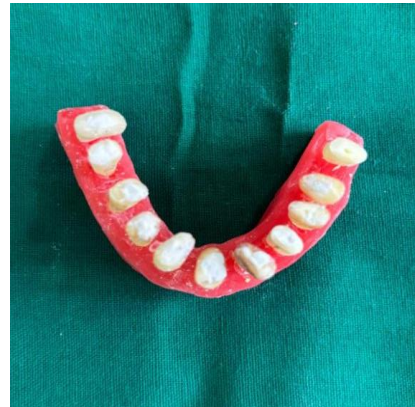


Fig 5: Samples arranged on wax rim

Following CBCT acquisition, the volume of the material within the root canal of each tooth was calculated using dedicated imaging software (Planmeca Romexis).[Fig 6]



Fig 6: CBCT imaging

The samples were subsequently divided into four experimental groups based on the irrigant used for Metapex removal and the application of ultrasonic activation:[Fig 7,8]



Fig 7 : Removal of Metapex intracanal medicament using ultrasonic agitation with an irrigating solution.



Fig 8 : Removal of Metapex intracanal medicament using conventional syringe irrigation without ultrasonic activation.

Group A1: Metapex was removed using 20 mL of 0.2% chitosan solution with ultrasonic agitation for 4 minutes.

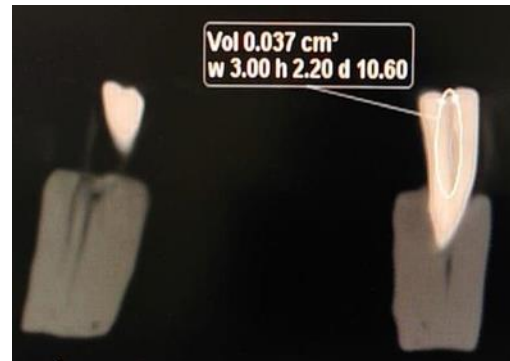
- Group A2: Metapex was removed using 20 mL of 17% EDTA solution with ultrasonic agitation for 4 minutes.

- Group A3: Metapex was removed using 20 mL of 0.2% chitosan solution for 4 minutes without ultrasonic activation.

- Group A4: Metapex was removed using 20 mL of 17% EDTA solution for 4 minutes without ultrasonic activation.

Subsequently, a final rinse was performed with 1 mL of distilled water. A second CBCT scan was then acquired, and the volume of the remaining intracanal material in each tooth was calculated using the same imaging software and methodology as described previously.[Fig 9]

A]



B]



Fig 9 : [A,B,] Volume calculation of intracanal medicament using CBCT software analysis.

Results Table 1- Normality assessment

| Group | Statistic | df | p-value |
|-----------------------|-----------|----|---------|
| Chitosan + Ultrasonic | 0.921 | 10 | 0.364 |
| EDTA + Ultrasonic | 0.956 | 10 | 0.740 |
| Chitosan | 0.677 | 10 | <0.001 |
| EDTA | 0.93 | 10 | 0.452 |

The Shapiro–Wilk normality test revealed that three groups—Chitosan + Ultrasonic, EDTA + Ultrasonic, and EDTA—showed normally distributed data with p-values greater than 0.05, while the Chitosan group alone demonstrated a significant deviation from normality ($p < 0.001$). Due to this lack of uniform normality across groups, non-parametric statistical methods were selected for further group comparison.



Table 2 - Comparison of % of material removed between four groups

| Group | Mean ± SD | Median (IQR) | χ^2 value | p-value |
|-----------------------|--------------|--------------|----------------|---------|
| Chitosan + Ultrasonic | 96.90 ± 0.96 | 97.09 (1.12) | 32.482 | <0.001* |
| EDTA + Ultrasonic | 95.31 ± 1.61 | 95.14 (3.12) | | |
| Chitosan | 90.21 ± 2.86 | 90.92 (1.65) | | |
| EDTA | 84.92 ± 5.24 | 86.15 (9.55) | | |

Kruskal Wallis test; * indicates a significant difference at $p \leq 0.05$

The Kruskal–Wallis test demonstrated a statistically significant difference in the percentage of residual metapex removed among the four groups ($\chi^2 = 32.482$, $p < 0.001$). The Chitosan + Ultrasonic group showed the highest removal efficiency ($96.90 \pm 0.96\%$), followed by EDTA + Ultrasonic ($95.31 \pm 1.61\%$), whereas the lowest removal was seen in the EDTA group ($84.92 \pm 5.24\%$). These findings suggest that ultrasonic activation greatly enhances the ability of irrigants to eliminate metapex medicament from the root canal system.

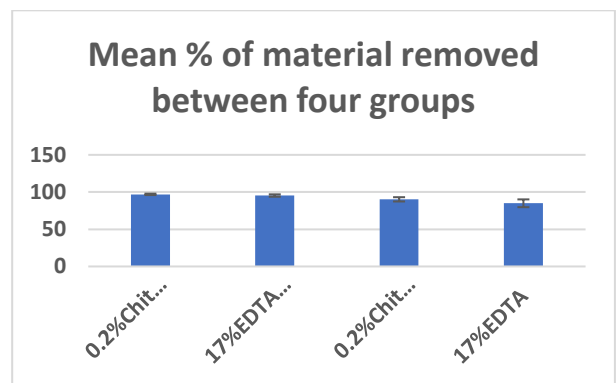
Table 3 - Pairwise comparison of % of material removed between four groups

| Group | Mean difference | p-value |
|--|-----------------|---------|
| Chitosan + Ultrasonic vs EDTA + Ultrasonic | 1.60 | 1.000 |
| Chitosan + Ultrasonic vs Chitosan | 6.69 | 0.001* |
| Chitosan + Ultrasonic vs EDTA | 11.98 | <0.001* |
| EDTA + Ultrasonic vs Chitosan | 5.10 | 0.062 |
| EDTA + Ultrasonic vs EDTA | 10.38 | <0.001* |

| | | |
|------------------|------|-------|
| Chitosan vs EDTA | 5.29 | 1.000 |
|------------------|------|-------|

Post hoc Bonferroni test; * indicates a significant difference at $p \leq 0.05$

Post hoc Bonferroni comparisons revealed significant differences between Chitosan + Ultrasonic and both Chitosan alone ($p = 0.001$) and EDTA alone ($p < 0.001$), as well as between EDTA + Ultrasonic and EDTA alone ($p < 0.001$). However, differences between Chitosan + Ultrasonic and EDTA + Ultrasonic ($p = 1.000$), and between Chitosan and EDTA alone ($p = 1.000$), were not statistically significant. Overall, the results indicate that the key factor contributing to improved metapex removal is ultrasonic activation, irrespective of the irrigant used independently.



Discussion

Effective removal of intracanal medicaments before obturation is a critical step in endodontic treatment, as residual medicaments may interfere with sealer adhesion, compromise the penetration of sealers into dentinal tubules, and ultimately affect the quality of the apical seal^[1, 2]. Calcium hydroxide–based medicaments such as Metapex are widely used because of their antimicrobial properties and ability to promote periapical healing^[5, 6]. However, complete removal of these medicaments from the root canal system remains challenging due to the complex anatomy of root canals and the tendency of medicaments to adhere to canal walls and penetrate dentinal tubules^[3, 11].

In the present study, cone beam computed tomography (CBCT) was used to evaluate the volumetric removal of Metapex from root canals. CBCT provides three-dimensional imaging that enables accurate volumetric assessment of intracanal materials and allows reliable comparison of medicament volumes before and after



irrigation protocols^[4,12]. This method overcomes the limitations of conventional two-dimensional radiographs, which may produce distortion, superimposition of anatomical structures, and inaccurate estimation of residual materials^[13]. Therefore, CBCT-based volumetric analysis has become an effective and reliable tool for evaluating intracanal medicament removal in *in vitro* studies^[4,13].

The present study also investigated the role of ultrasonic activation in enhancing the effectiveness of irrigating solutions. Ultrasonic activation has been widely reported to improve irrigant performance by generating acoustic streaming and cavitation, which increase irrigant penetration into canal irregularities, lateral canals, and dentinal tubules^[10,14]. This hydrodynamic agitation enhances the mechanical disruption and displacement of medicament remnants that may otherwise remain attached to canal walls when conventional syringe irrigation is used^[15].

Among the groups evaluated in this study, 0.2% chitosan with ultrasonic activation demonstrated the highest efficiency in removing Metapex, showing the least residual medicament volume. The superior performance of chitosan may be attributed to its chelating properties, which facilitate the removal of inorganic components and smear layer from dentinal surfaces^[7,16]. Chitosan has also been reported to possess favorable biocompatibility and effective chelating ability, making it a promising alternative to conventional chelating agents in endodontics^[16,18]. When combined with ultrasonic activation, the enhanced irrigant penetration and acoustic streaming further improve its cleaning efficacy within complex root canal anatomy^[14].

The group treated with 17% EDTA with ultrasonic activation also demonstrated significant removal of Metapex, although its effectiveness was slightly lower compared with activated chitosan. EDTA is a well-established chelating agent widely used in endodontics for smear layer removal and for facilitating cleaning of the root canal system^[3,17]. However, differences in the chelating mechanism and interaction with dentin may explain the comparatively lower removal efficiency observed when compared with chitosan in the present study^[16].

The 0.2% chitosan without ultrasonic activation group exhibited moderate removal efficiency, suggesting that

the chemical action of chitosan alone may not be sufficient to completely eliminate intracanal medicaments from the canal system. These findings highlight the importance of mechanical agitation in enhancing irrigant effectiveness, as agitation promotes deeper penetration and improved contact between the irrigant and canal walls^[14,18].

In contrast, the 17% EDTA without ultrasonic activation group demonstrated the highest residual medicament volume, indicating the least effective removal protocol. Conventional syringe irrigation is often insufficient to deliver irrigants effectively into complex canal irregularities such as fins, isthmuses, and apical ramifications, leading to incomplete removal of intracanal medicaments^[15,19].

The findings of the present study therefore emphasize that both the choice of irrigating solution and the use of activation techniques play a crucial role in improving intracanal medicament removal. Ultrasonic activation significantly enhances cleaning efficiency by increasing irrigant penetration, improving mechanical disruption of medicament remnants, and facilitating their removal from complex canal anatomy^[10,14,20].

From a clinical perspective, the results suggest that the combination of 0.2% chitosan with ultrasonic activation may provide superior removal of calcium hydroxide-based intracanal medicaments, thereby improving canal cleanliness before obturation and potentially enhancing the long-term success of endodontic treatment.

Conclusion

Within the limitations of this *in-vitro* CBCT volumetric analysis, Chitosan with ultrasonic activation was found to be the most effective in removing Metapex intracanal medicament, followed by EDTA with ultrasonic activation and Chitosan without activation. EDTA without ultrasonic activation showed the least removal efficiency.

Thus, the use of chelation combined with ultrasonic activation can be recommended for improved intracanal medicament removal prior to obturation.

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