

## Research Article

## A comparative evaluation of irrigation methods for calcium hydroxide removal from the root canal: An in vitro study

Masako Nakano<sup>1</sup>, Yuko Yamamoto<sup>1\*</sup>, Ami Misawa<sup>2</sup>, Shunjiro Yamakawa<sup>1</sup>, Takumasa Yoshida<sup>1</sup>, Yasushi Yamazaki<sup>1</sup>, Noriyasu Hosoya<sup>1</sup>

<sup>1</sup>Department of Endodontology, Tsurumi University School of Dental Medicine, Japan

<sup>2</sup>Department of Endodontics, NYU College of Dentistry, New York University, USA

\***Corresponding Author:** Yuko Yamamoto, Department of Endodontology, Tsurumi University School of Dental Medicine, Japan

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### Abstract

**Background:** The lingering COVID-19 pandemic poses significant challenges in endodontic management. Calcium hydroxide is being widely used as a long-term root canal medicament due to delays in treatment completion, however, the best choice with great efficiency for removal has yet to be developed up to the present. This study aimed to help establish a reliable irrigation protocol by comparing the removal proportions in the root canal system.

**Methods:** Thirty extracted maxillary central incisors were used. The root canals were enlarged, filled with Ca(OH)<sub>2</sub>, sealed, then divided into six groups. Group 1: irrigation needle with syringe (NS) with NaClO. Group 2: intracanal negative pressure needle (INP) with

NaClO. Group 3: ultrasonic irrigation (UI) with NaClO. Group 4: NS with NaClO+EDTA. Group 5: INP with NaClO+EDTA. Group 6: UI with NaClO+EDTA. Removal proportions were statistically analyzed.

**Results:** Residual Ca(OH)<sub>2</sub> was observed in all groups tested. Significant differences were noted between Group 1 and all other groups but not among Groups 2 to 6. In INP, the removal proportions were greater when the combined solutions were used compared to when NaClO alone was used. No significant differences were observed between INP and UI in any regions of the canal.

**Conclusions:** None of the irrigation protocols achieved complete removal of Ca(OH)<sub>2</sub>. When either the iNP

needle or ultrasonic tip was used, the sole use of NaClO achieved high removal efficiency. Contrarily, when an irrigation needle with a syringe was used, the combined use of NaClO and EDTA was considered preferable.

**Keywords:** Calcium hydroxide  $\text{Ca(OH)}_2$ ; Root canal irrigation; Intracanal negative pressure needle; Passive ultrasonic irrigation

## 1. Introduction

The lingering COVID-19 pandemic poses significant challenges in endodontic patient management and its treatment procedures. Long term calcium hydroxide placement due to delays in treatment completion is being re-evaluated [1,2].  $\text{Ca(OH)}_2$  is widely used as a root canal medicament and has been confirmed to provide low tissue irritation, promote fibroblast and osteoblast activation and heal the apical region [3,4].  $\text{Ca(OH)}_2$  has long-lasting antimicrobial effects due to its strong alkalinity [5]. Though there are several rebutting reports [6,7] the survivability of teeth with long-term  $\text{Ca(OH)}_2$  dressing does not appear to pose a detrimental effect on tooth loss [8]. However, canals need dense and homogenous dressing because of its non-volatility, which can be removed only with difficulty [5,9,10]. Eliminating  $\text{Ca(OH)}_2$  is a necessary step in preparation prior to obturation, as its remnants adversely affect adhesion. Poor seal in the apical region can increase the risk of apical periodontitis occurrence or recurrence [11,12]. Particularly when a resin-based sealer is used, closure of dental tubules and inhibition of dentin decalcification can weaken the adhesion of the sealer [13]. Various methods to remove  $\text{Ca(OH)}_2$  dressing from the canal system have been reported. These include chemical cleaning solutions irrigated with sodium hypochlorite, EDTA, citric acid, lactic acid or other chemicals [14-16], and mechanical cleaning with hand files, Ni-Ti rotary files, ultrasonic files or irrigation needles, as well as numerous combinations of

these methods and other techniques [17-21]. With any of these methods, however, its low water solubility hampers development of solid way of thorough removal. This study was performed to help establish a safe and reliable removal protocols of  $\text{Ca(OH)}_2$  dressing. This study was approved by the ethics committee at Tsurumi University School of Dental Medicine (No.1041).

## 2. Materials and Methods

Thirty freshly extracted human maxillary central incisors were used. Tooth selection criteria included a single root canal with a complete root apex, no visible root caries, fractures, or cracks on examination with a  $\times 3$  magnifying loop. The crown of each tooth was removed by sectioning 15 mm from the anatomical root apex. Apical patency was checked before instrumentation using #10 K-files (Mani, Tochigi, Japan). The root canals were prepared by K3 XF 3 NiTi rotary instruments (SybronEndo, West Collins, CA) up to #60/0.04 with the manufacturer's instruction, with a working length of 14 mm. Longitudinal grooves were formed and each tooth was split along the groove [20-22] with plaster pliers (YDM, Tokyo, Japan). The two halves were then reattached with cyanoacrylate. We determined that there was no space between the fragments with the magnifying loop. Each test canal was filled with  $\text{Ca(OH)}_2$  (Calcipex II, Nishika, Yamaguchi, Japan) which was delivered directly into the canals by a plastic syringe and delivery tip according to the manufacturer's instruction until the  $\text{Ca(OH)}_2$  paste extrusion was visually confirmed from the apex. The canals were then sealed with hydraulic temporary restorative (Cavition, GC, Tokyo, Japan) (2-mm thickness). All the samples were stored for 1 week in a 37°C at 100% relative humidity. The test samples were randomly divided into six experimental groups five teeth each according to the solution and irrigation protocols. All the apices were sealed with sticky wax

and irrigated for 30s. In Group 1 (NS), a flat-tipped 23G irrigation needle attached to a syringe was inserted 12 mm into the canal and irrigation was performed using 4.5 mL of NaClO. A spillage of the irrigant was aspirated at the canal orifice. In Group 2 (INP), Intracanal irrigation was performed using an intracanal negative pressure needle, iNP 60 needle (Mikuni Kogyo Co., LTD., Nagano, Japan) which was connected to a dental unit aspirator. A needle tip with an outer diameter of 0.55 mm was selected and inserted 12 mm into the canal. NaClO was added into the canal as needed to keep the canal full with the solution.

In Group 3 (UI), an ultrasonic tip (SC point 4-19, Osada Electric Co., LTD., Tokyo, Japan) for intracanal debris removal was attached to an ultrasonic therapy apparatus (OE-10W, Osada Electric Co., LTD., Tokyo, Japan) and inserted to the canal. A spillage of the irrigant was aspirated at the canal orifice at power level 2, minimizing contact with the canal wall. The solution was added as needed to keep the canal full. In Group 4, irrigation protocol was the same as in Group 1, but with NaClO, then EDTA, followed by NaClO. In Groups 5 and 6, irrigation protocol was the same as in Groups 2 and 3, respectively, but with NaClO, EDTA, followed by NaClO. In all groups, a total of 4.5 ml of the solution was used in 30s. During irrigation, operators moved the instrument up and down in the canal in the range of several millimeters.

Group 1. a flat-tipped irrigation needle+syringe (NS) with 4.5 ml of NaClO for 30s

Group 2. INP needle irrigation (INP) with 4.5 ml of NaClO for 30s

Group 3. Ultrasonic irrigation (UI) with 4.5 ml of NaClO for 30s

Group 4. NS with 1.5 ml of NaClO, 1.5 ml of EDTA, followed by 1.5 ml of NaClO for 10s each (30s in total)

Group 5. INP with 1.5 ml of NaClO, 1.5 ml of EDTA, followed by 1.5 ml of NaClO for 10s each (30s in total)

Group 6. UI with 1.5 ml of NaClO, 1.5 ml of EDTA, followed by 1.5 ml of NaClO for 10s each (30s in total)

The irrigation solution was 6% NaClO. In Group 4-6, NaClO was used in combination with 15% EDTA (Morhonline for dental use, Showa Yakuhiin Kako, Tokyo, Japan).

Following irrigation, all the test samples were separated along the grooves, and standardized photographic images were acquired for the remaining Ca(OH)<sub>2</sub> on the canal wall. The area of Ca(OH)<sub>2</sub> remnants on coronal, middle and apical third was determined using the image analysis software (ImageJ, National Institutes of Health, Bethesda, MD, USA). The percentage area of removed Ca(OH)<sub>2</sub> was calculated according to the following equation. All the canals were confirmed to be dressed entirely with Ca(OH)<sub>2</sub> by the above software which was drawn as a denominator:

$$\% \text{ Area of Ca(OH)}_2 = (100-a) / 100 \times 100$$

The mean percentage area of removed Ca(OH)<sub>2</sub> for each group and region was acquired as well. The removal proportions of the groups and canal regions were statistically tested by two-way factorial analysis of variance and Scheffe's test, with a significance level of 5%.

### 3. Results

Residual Ca(OH)<sub>2</sub> was observed on the entire canal walls of all groups tested. The average removal proportion was 46.68±12.84% in Group 1, 77.48±5.76% in Group 2, 67.03±12.64% in Group 3, 67.91±5.35% in Group 4, 78.23±7.17% in Group 5 and 74.99±6.85% in Group 6. Significant differences were noted between Group 1 (NS + NaClO) and all other groups (Group 1 vs.2, 1 vs.3, 1 vs.4, 1 vs.5, 1 vs.6) but not among Groups 2 to 6. As of the use of an irrigation needle

attached to a syringe, the removal proportions were far greater when the combined solutions, NaClO and EDTA were used compared to when NaClO alone was used (Group 1 vs. 4). No significant differences were observed between the use of the intracanal negative pressure needle (Group 2 vs.5) and ultrasonic tip (Group 3 vs. 6) in any regions of the canal. As demonstrated in Table 1, in the coronal root third, significant differences were observed between Group 1 (NS + NaClO) and all other groups were observed. In the middle third, significant

differences were found between Groups 1 (NS + NaClO) and 2 (INP + NaClO), and between Groups 1 (NS + NaClO) and 5 (INP + NaClO + EDTA). In the apical third, significant differences were found between Group 1 (NS + NaClO) and Groups 2 (INP + NaClO), 1 and 5 (INP + NaClO + EDTA), and 1 and 6 (UI + NaClO + EDTA). No significant differences among any of the three regions were noted between the use of the intracanal needle and ultrasonic tip (Group 2 vs.3, 5 vs 6) (Table 1).

Region \ Group	1	2	3	4	5	6
Coronal third	38.61±17.38	76.53±6.59	74.72±10.67	64.61±11.39	77.30±10.41	75.41±7.87
Middle third	53.72±14.48	82.81±6.78	61.35±19.55	79.93±4.77	79.93±4.77	77.25±12.21
Apical third	48.48±17.15	73.50±2.77	64.98±14.38	59.15±9.03	72.15±5.05	72.27±5.95

**Table 1:** Mean and SD percentage of Ca(OH)<sub>2</sub> removed from each canal region with 6 different protocols.

## 4. Discussion

### 4.1 Experimental method

In conventional comparisons of canal medicament removal in extracted teeth by evaluating canal walls, some experimental protocols in which roots are separated after flushing, dislodged remnants often limits accurate measurements. To overcome this disadvantage, we therefore split and reassembled the root in advance then separated it again preserving the condition of the canals to make a precise evaluation [20-22]. To standardize a way to place the Ca(OH)<sub>2</sub> paste homogeneously into the canals without making a dead space, we used a plastic syringe and delivery tip to deliver the medicament evenly throughout the canal until an extrusion of Ca(OH)<sub>2</sub> beyond the apex was clearly observed.

### 4.2 None of the protocols achieved thorough removal

The results indicate that none of the irrigation protocols tested in this study, with or without passive ultrasonic irrigation, or intracanal negative pressure needle irrigation with NaClO and EDTA, was not able to remove Ca(OH)<sub>2</sub> dressing thoroughly from the entire canal in any of the 30 teeth after 30s irrigation procedure with a total of 4.5ml of solutions.

### 4.3 The combined use of solutions improved removal efficiency in NS

When irrigation was performed by a flat-tipped 23-G irrigation needle attached to a syringe, the sole use of NaClO removed Ca(OH)<sub>2</sub> dressing least effectively in all the irrigation protocols, however, the combined use of NaOCl and EDTA distinctively improved the removal efficiency. As optimal irrigation could be

achieved by combined two or more chemical irrigating solution in a specific order to attain the goals of irrigation with proper safety and effectiveness [23], the most common and simplest irrigation conducted by a needle attached to a syringe can preferably be applied with NaClO and EDTA to enhance its cleaning ability. In a research, smaller size needles could not be the best option in reaching the deeper most part of the root canal while some studies showed that 30-G needles could reach out to get close to the working length and were more efficacious in cleaning the apical areas of root canal [23]. When it comes to needle designs, there was no significant advantage between the flat and beveled or notched needles among open-ended needles [23]. Thus, we opted for 23-G irrigation needle that could not penetrate deeply into the prepared test canals. In the middle and apical root third, the combined use of NaClO and EDTA significantly increased the removal efficiency compared to NaClO alone in NS, but not in INP or UI. Previous studies demonstrated that Ca(OH)<sub>2</sub> was not completely removed with 5% NaClO irrigation alone [24] and chelators appear to be more effective than NaClO for Ca(OH)<sub>2</sub> removal [25]. This effect may be explained by the formation of complexes between ethylenediaminetetraacetic acid, EDTA and calcium ions [26,27]. Our findings indicate that under the condition of 30s of irrigation time and 4.5 ml of irrigation volume, not only mechanical agitation but also chemical activity of the irrigants positively affected the efficacy of Ca(OH)<sub>2</sub> removal. Recently, however, the erosive effects of these chelators on dentin have been verified for a possible increase in the fracture susceptibility of roots [28]. EDTA has also been reported to have limitations and negatively affect the building ability of macrophages proportional to its concentration [29]. Different solutions, such as citric acid [30], phosphoric acid [31], glycolic acid [32] and 70%

ethanol [33], along with combinations, have been used to remove Ca(OH)<sub>2</sub> dressing. Glycolic acid exhibited higher efficacy than EDTA at 10% concentration while 70% ethanol is advantageous to improve removal from the apical root third compared with 2.5% NaClO or 17% EDTA or EDTA-T. However, up until today, none of the solutions that totally removes the intracanal Ca(OH)<sub>2</sub> dressing from the apical root third has yet to be corroborated. The sole use of NaClO displayed reasonable performance when either INP or UI was used. Even though a thorough removal was not achieved, the iNP needle showed unvarying high performance in irrigation with or without EDTA (Group 2 vs. 5). The ultrasonic tip showed a similar trend as the iNP needle (Group 3 vs. 6). Both irrigation devices exhibited almost equally high performance under the same condition that NaClO alone or the combined use of NaClO and EDTA (Group 2 vs. 3, 5 vs. 6) was used. This might suggest that Ca(OH)<sub>2</sub> dressing can be eliminated to some degree by using NaClO alone when each of the two devices was used. 35% or more of the canals remain untouched by endodontic instruments so the root canal irrigants should ideally be reached to the apical regions to flush out debris, kill microbes, remove microbial by-products and the smear layer [34]. To overcome the difficulty in delivering irrigants into the apical area without inadvertent injection beyond the foramen, the apical negative pressure technique has been introduced [35,36]. The iNP needle [37] is 32-mm long with an external and internal diameter of 0.64 and 0.44mm, respectively. The open tip of the iNP needle has an external and internal diameter of 0.55 and 0.44 mm, respectively. The inner diameter should allow a high volumetric flow rate and the outer diameter placement at 2 mm from the working length when the canal is enlarged to #60/0.04. A greater pressure differential can be achieved by the iNP needle, which

could be explained by the needle lumen. Meanwhile, the ultrasonic tip used in this study was made of stainless steel with an end diameter of 0.25 mm, a taper of 0.05, and a length of 19 mm. Although the tip does not shape a considerable amount of hard tissue, direct contact with canal walls was minimized not to affect the root canal morphology. As can be seen from the results, in the middle and apical root third, the ultrasonic tip with the combined use of solutions showed enhanced removal proportions compared to when NaClO alone was used. This is because the use of ultrasonic tip for agitating solutions induces cavitation and acoustic streaming and so the solution circulates into all aspects of the root canal system [36,38]. In the middle third, however, an ultrasonic tip using NaClO alone showed poor removal proportions. We surmise that the insoluble Ca(OH)<sub>2</sub> piled up and stayed in the middle of the canal soon after the solution flow got rid of some portion of Ca(OH)<sub>2</sub> dressing from the canal walls. Given all this, the ultrasonic activation still could be one of the efficient techniques to achieve the cleaner root canal system. One of the most concerning characteristics in the nature of dental procedures during the coronavirus pandemic is the use of high-speed handpieces and ultrasonic tips, which generate aerosols of saliva particles, blood and other fluids [39-44]. Judging from the present situation, the use of dental equipment including negative pressure device could be a safer option rather than ultrasonic device. The limitation of this study was that the standardized 4.5ml of solution volume and 30 s of irrigation time does not reveal overall trends, however, these findings could be valid indicators to capture general pattern which helps set new protocols to be tested in the future studies.

## Conclusions

Within the limitation of this in vitro study, thorough removal of Ca(OH)<sub>2</sub> dressing from the root canal system was not achieved by any tested methods. The combined use of NaClO and EDTA improved removal efficiency when a needle attached to syringe was used. The sole use of NaClO displayed reasonable performance when either INP or UI was used.

## Author Contributions

Conceptualization, N.H., M.N.; methodology, M.N., N.H.; software, T.Y., N.H.; validation, A.M., Yu.Y.; formal analysis, M.N., N.H.; investigation, Ya.Y, T.Y; resources, N.H., Ya.Y.; data curation, Yu.Y., S.Y.; writing, original draft preparation, M.N., Yu.Y., A.M.; writing, review and editing, Yu.Y., N.H.; visualization, S.Y., Yu.Y; supervision, N.H., Ya.Y.; project administration, N.H.; funding acquisition, N.H. All authors have read and agreed to the published version of the manuscript.

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## Institutional Review Board Statement

This study involving human specimen was conducted according to the guidelines of the Declaration of Helsinki, and was approved by the ethics committee at Tsurumi University School of Dental Medicine (Approval No.1041)

## Conflicts of Interest

The authors declare no conflict of interest.

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