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Removal of calcium hydroxide dressing from the root canal system using different irrigation solutions and methods

Introduction: The aim is to compare different methods for the removal of calcium hydroxide $(Ca(OH)_2)$ from root canals.

Material and methods: 160 extracted human teeth were divided into 2 groups. In group 1 (n = 80), all root canals were prepared with hand instruments to ISO size 40 and in group 2 (n = 80) by rotary nickel-titanium files (Mtwo) to size 04/40. After rinsing, all root canals were filled with Ca(OH)₂ and the access cavity was temporized. All teeth were stored for 7 d at 37 °C and 100% humidity. After storage, in half of the specimens of both groups (n = 40) root canal irrigation without previous instrumentation was performed. In the other half (n = 40) root canals were instrumented to working length with Hedstrom file ISO size 45. All specimens were divided in subgroups (n = 10) and rinsed with 5 ml of NaCl-solution 0.9%, CHX 2%, and NaOCl 2.5% with or without ultrasonic activation, respectively. By scanning electron microscope evaluation the cleanliness of the root canal walls was scored from 1 (no Ca(OH)₂ visible) to 5 (pronounced layer of Ca(OH)₂). The data obtained were statistically evaluated by Kruskal-Wallis-test (p < 0.05).

Results: Ultrasonic-activated NaOCl removed significantly more $Ca(OH)_2$ than all other solutions or methods (p < 0.05). The instrument taper (hand instruments 2% or NiTi files 4%) as well as instrumentation before rinsing, had no significant influence (p > 0.05). For all rinsing solutions tested, the result within the respective group was independent of the localization in the root canal (p > 0.05).

Conclusion: Only passive ultrasonic activation was able to remove $Ca(OH)_2$ from the root canal sufficiently. Neither the taper of the instruments used nor instrumentation before rinsing had an significant influence on the removability of $Ca(OH)_2$.

Keywords: calcium hydroxide; instrument taper; passive ultrasonic irrigation (PUI); root canal irrigation; root canal dressing

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Introduction

Disinfection of the root canal system after preparation and before obturation is a prerequisite for successful root canal treatment [15, 27]. Due to the complexity of the root canal system, only about 50-60% of the canal wall surfaces are mechanically cleaned even with current rotary nickeltitanium (NiTi) instruments [25]. Therefore, under clinical conditions with a mechanical instrumentation and antibacterial irrigation it is only possible to remove microorganisms in 50-70% of the infected root canals, depending on the irrigation protocol [2]. If a root canal is infected with microorganisms, they may survive in dentinal tubules, ramifications, accessory canals, anastomoses, apical delta etc., where they are not accessible to mechanical instrumentation or irrigation [2]. In order to remove microorganisms from an infected root canal system, the use of root canal medication is particularly indicated in non-vital teeth [15, 27]. In the past, an abundance of substances has already been proposed as a root canal medication. But until today calcium hydroxide (Ca(OH)₂) is a popular and commonly used, widespread root canal dressing [15, 27], introduced to dentistry for this purpose by Hermann in 1920 [13].

The effectiveness of $Ca(OH)_2$ in endodontics is mainly due to its antibacterial activity to hydrolyse the lipid moiety of bacterial lipopolysaccharides (LPS) without being cytotoxic and to dissolve soft tissue in the root canal [15, 27].

Besides many positive characteristics, $Ca(OH)_2$ has some drawbacks like being insufficiently effective against E. faecalis, facultative anaerobic bacteria and yeasts [15, 27]. Another disadvantage is that it is often not possible to remove $Ca(OH)_2$ completely from the root canal system. Hence, the use of $Ca(OH)_2$ as root canal medication has been discussed controversially recently [15, 27].

Because the removal of $Ca(OH)_2$ is frequently incomplete [21], 20–45% of the root canal wall dentine is covered with residue of the dressing, even after copious irrigation [20]. The disadvantage of remaining





Total = 160 teeth

Group 2, n = 80

NiTi- instrumentation (Mtwo)

Figure 1 Experimental setup.

Ca(OH)₂ is the possible negative impact on root canal filling materials and their properties. Residual Ca(OH)₂ may reduce sealer adaptation on the root canal wall dentine [5, 34] and may hinder the penetration of sealer into dentinal tubules [7], thus reducing bondstrength of epoxy resin-based sealers [12], resin-based sealers [5], siliconbased sealers [10], and zinc-oxide eugenol-based sealers [17]. Furthermore, residual Ca(OH)₂ can reduce the processing time, shorten the setting time and increase the film thickness of the sealer [22]. In conclusion, Ca(OH)₂ may interfere with the root canal sealer, compromise the quality of the root canal filling and hence of the whole treatment [15, 27]. Finally, remaining Ca(OH)₂ may be washed out by body fluids or will be resorbed from the apical part of the root canal over time resulting in apical leakage and treatment failure [28]. Therefore, root canal sealers may show leakage after Ca(OH)₂ applications [5]. In addition, $Ca(OH)_2$ used as long-term root canal dressing may increase the risk of root fracture [1]. Thus, $Ca(OH)_2$ needs to be removed completely from the root canal system [15, 27].

With regard to the root canal taper after preparation, it was found that irrigation of root canals with a greater taper allows improved disinfection of the entire root canal system [6, 26]. However, it is still unclear whether a lager diameter of the root canal and a greater conicity, and thus a higher volume of irrigation solution also lead to improved removability of Ca(OH)2. Therefore, the aim of the present study was to investigate different methods and irrigation solutions for the removal of aqueous calcium hydroxide suspension (Ca(OH)₂) from root canals of extracted human teeth. The following null hypotheses should be tested: 1. Preparing the root canals before medication with NiTi-files with an enlarged taper (4% instead of 2%)

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Figure 2a Grade 1 = no calcium hydroxide remnants visible, all dentinal tubules are patent.

Figure 2a–e Examples of the electron microscopic evaluation of scores 1 to 5, magnification 2,500×.



Figure 2b Grade 2 = small amounts of calcium hydroxide remnants visible, some dentinal tubules are open.



Figure 2c Grade 3 = Calcium hydroxide homogeneously coats almost the entire root canal wall, only isolated dentinal tubules are open.

has no positive influence on the removability of $Ca(OH)_2$ by root canal rinsing solutions.

2. Further preparation of the root canals from ISO 40 to ISO 45 with hand instruments before root canal irrigation has no positive influence on the removability of $Ca(OH)_2$.

Materials and methods

All full-aged participants provided their written informed consent that the extracted teeth may be used for study purposes. The handling of all human samples followed strictly the "Declaration of Helsinki".

Prior to root canal treatment, all teeth were investigated under a stereomicroscope (Expert DN, Müller Optronic, Erfurt, Germany) to exclude cracks or root resorption. Buccal and proximal radiographs were taken to ensure a single root canal with an intact apical region and a single apical foramen. 160 extracted human single-rooted upper incisor teeth that showed a round root canal with a width of approximately ISO size 15 near the apex were included in this study. The root canal width was examined with silver points ISO size 15 and 20 (VDW, Munich, Germany). Patency of the canal was determined with K-files ISO size 10 (VDW). The working length was defined by measuring the length of the initial instrument (K-Files ISO size 10; VDW) visible at the major apical foramen minus 1 mm.

The total of 160 teeth was divided into two groups: group 1 included 80 teeth in which the root canals were prepared by hand instrumentation using Reamers and Hedstrom files from ISO size 15 up to ISO size 40 (VDW). Group 2 included also 80 teeth in which the root canals were instrumented by rotary NiTi-files up to size 04/40 (Mtwo; VDW) using the torque-limited endodontic motor VDW.Silver (VDW) with the settings according the manufacturer's instructions. The instruments were cleaned after 3 pecks (in-and-out move) and the root canal was irrigated with 5 ml NaOCl 2.5% during the instrumentation. After instrumentation the root canal system was irrigated with 5 ml isotonic saline solution (NaCl) 0.9% and 2 ml EDTA 17% to remove the smear layer. Afterwards, again 5 ml NaCl 0.9% were applied as final irrigant. A 30 g open needle (Miraject; Hager & Werken, Duisburg, Germany) was used to perform the irrigation during and after instrumentation, in which the needle was inserted as deep as possible into the canal but without binding. The root canals were dried by paper points afterwards.

After drying, all root canals were filled with an aqueous $Ca(OH)_2$ suspension (Calxyl blau, OCO, Dirmstein, Germany) by using a Lentulo (VDW) which reached the exact working length of each tooth. Thereafter, the access cavity was temporised by using Cavit (3M ESPE, Seefeld, Germany) and all samples were stored in an incubator (Wärme- und Trockenschrank, Heraeus, Hanau, Germany) in NaCl 0.9 % at 37 °C and 100 % humidity for 7 days.

Before removing Ca(OH)₂ from the root canals, each group (group 1 and 2) was again divided, so that 4 groups with 40 teeth resulted (groups 1.1, 1.2, 2.1, 2.2). The samples of Group 1.1 and 2.1 were rinsed only with an irrigation solution to remove Ca(OH)₂ from the root canals. In group 1.2 and 2.2 the root canals were instrumented with Hedstrom files ISO size 45 (VDW) right before irrigation to remove Ca(OH)₂ also mechanically. Four different irrigation solutions or methods were used so that 10 teeth in each group were rinsed with the same solution or method. The following irrigation solutions were used:

- A: Sodium hypochlorite 2.5% (NaOCl) (Hospital pharmacy of the University Hospital, Münster, Germany) + passive ultrasonic activation (VDW Ultra; VDW)
- B: Sodium hypochlorite 2.5 % (NaOCl)
- C: Chlorhexidine 2% (Chlorhexidindigluconat-Lösung 2%; Engelhard Arzneimittel, Niederdorfelden, Germany)
- D: Isotonic saline solution 0.9% (NaCl) (Hospital pharmacy of the University Hospital, Münster, Germany)

5 ml of the respective irrigation solution were applied in each root canal using a 30 g open needle whereby the needle was inserted as deep as possible into the canal but without binding. In all groups where NaOCl was activated this was done using a file



Figure 2d Grade 4 = the entire root canal wall is homogeneously covered with calcium hydroxide, no patent dentinal tubules.



Figure 2e Grade 5 = the complete root canal wall is covered with a distinct layer of calcium hydroxide.

size 25 (Irri-S 21/25; VDW) with a frequency of 28,000 Hz as recommended by the manufacturer. The Irri-S tip was placed 2 mm short of working length and in-and-out movements with an amplitude of 5 mm were performed. Irrigation for passive ultrasonic activation (PUA) was repeated 2 times for 30 s. Care was taken to ensure that the volume (5 ml) and the contact time of the irrigation solution (2 min) in the root canal were identical in all groups.

Thereafter, the samples were prepared for scanning electron microscopic (SEM) observation to assess the Ca(OH)₂ remnants on the root canal walls. All teeth were carefully split longitudinally to allow visualization of the root canal lumen. Afterwards, the root halves were attached to a sample plate (Provag, Oestrich-Winkel, Germany) by using an electrically conductive glue (Leit-C nach Göcke; Chemikalien Neubauer, Münster, Germany). To ensure drying of the glue, the samples were stored dustproof for 24 hours. Afterwards the specimens were coated in a thin film of gold (95 nm) using a sputtering technique to provide conductivity of each sample (Sputter Coater, Balzers Union, Balzers, Liechtenstein). The specimens were divided into 3 sections by marking them with a waterproof pen so that a coronal, middle and apical third of the root canal resulted.

Finally, the specimens were observed under the scanning electron microscope (Philips PSEM 500X, Eindhoven, Netherlands). The visual evaluation of the root canal halves was carried out at 2,500× magnification, so that the root canal areas could be visibly recorded, examined, evaluated and photographically documented.

In order to assess the $Ca(OH)_2$ remnants on the root canal walls as well as to evaluate the effectiveness of each irrigating solution and its application method, the scoring system by Hülsmann et al. [14] was modified as follows:

- **Grade 1** = no Ca(OH)₂ remnants visible, all dentinal tubules are patent.
- **Grade 2** = small amounts of $Ca(OH)_2$ remnants visible, some dentinal tubules are open.
- **Grade 3** = Ca(OH)₂ homogeneously coats almost the entire root canal wall, only isolated dentinal tubules are open.
- **Grade 4** = the entire root canal wall is homogeneously covered with Ca(OH)₂, no patent dentinal tubules.
- **Grade 5** = the complete root canal wall is covered with a distinct layer of $Ca(OH)_2$.

Statistical analyses

The grades were recorded for every single tooth at the coronal, middle and apical third of each root canal. In order to evaluate significant differences in Ca(OH)₂ removal within the different groups, the data were statistically evaluated using the Kruskal-Wallis-Test (p < 0.05) (MedCalc, Ost-

end, Belgium). The data were checked for normality before using the non-parametric test for statistical comparison.

Results

Only irrigating with NaOCl 2.5% in combination with PUA showed significantly less Ca(OH)₂ remnants on the root canal walls compared to all other irrigation solutions and methods (p < 0.05) (Table 1). Instrument taper and an additional instrumentation right before root canal irrigation had no statistically significant influence on the removability of Ca(OH)₂ from the root canal (p > 0.05) (Table 2). Thus, neither the preparation with NiTi files taper 4% instead of hand instruments taper 2% nor an additional instrumentation with a Hedstrom file ISO size 45 before rinsing could decrease the amount of Ca(OH)₂ remnants on the root canal wall significantly.

In all groups where NaOCl in combination with PUA was used for root canal irrigation the highest amount of patent dentinal tubules was observed. Hence, this irrigating protocol mostly reached grade 1 or 2 from the scoring system except for 5 teeth which were classified as grade 3 in some root canal sections. None of the other rinsing solutions or methods yielded a single tooth section rated grade 1. No statistically significant differences were observed in all other specimens without the use of NaOCl plus PUA (p > 0.05). Furthermore, no statistically significant difference between the results of the 3 root canal sections (apical, middle or coronal third) was observed, independent of irrigation method and solution (p > 0.05). The amount of Ca(OH)₂ residues was equal in all areas of the root canal.

Discussion

In this in vitro study passive ultrasonic activation of NaOCl 2.5% was significantly more effective in removing Ca(OH)₂ from the root canal walls than NaOCl without PUA or all other irrigation protocols tested (p < 0.05). In some root canals, passive ultrasonically activated NaOCl 2.5% was able to remove the Ca(OH)₂ dressing completely, while 80 Removal of calcium hydroxide dressing from the root canal system using different irrigation solutions and methods

	NaOCI + PUA	NaOCI	СНХ	NaCl
Score 1 in %	65.0	0.0	0.0	0.0
Score 2 in %	30.8	23.3	20.0	20.8
Score 3 in %	4.2	41.7	47.5	41.7
Score 4 in %	0.0	30.8	28.3	31.7
Score 5 in %	0.0	4.2	4.2	5.8

 Table 1 Percentage of each irrigation solution and method regarding the different scores.

	Hand instrumentation	Rotating NiTi-files	снх	NaCl
	Only rinsing	Instrumentation with Hedstrom file ISO size 45 right before rinsing	Only rinsing	Instrumentation with Hedstrom file ISO size 45 right before rinsing
	Group 1.1	Group 1.2	Group 2.1	Group 2.2
NaOCI + PUA	1/2/3	1/2/3	1/2/3	1/2/3
NaOCI	4	4	4	4
снх	4	4	4	4
NaCl	4	4	4	4

Table 2 Statistical evaluation of the results. Statistically significant differences to other groups show 1/2/3, outcomes without significant differences show 4; Kruskal-Wallis-Test (p < 0.05).

no other irrigation solution or method could generate root canals with completely patent dentinal tubules. This result was independent from which cone (2% or 4%) was used to prepare the root canals and whether or not the root canals were instrumented with a Hedstrom file ISO size 45 to mechanically remove Ca(OH)₂ before irrigation. Hence, the null hypotheses were therefore accepted: neither a larger taper nor instrumentation prior to root canal irrigation has a significant influence on the removability of $Ca(OH)_2$ from the root canal. To the best of our knowledge, this is the first study to evaluate the influence of the instrument taper on the removability of $Ca(OH)_2$.

Rinsing solely with NaOCl 2.5% without PUA is significantly less effective than with PUA. This finding is

in accordance with previous investigations [24, 31, 32]. Despite the superior cleaning effect of ultrasonically activated NaOCl, it had to be noted that all experimental groups showed remnants of $Ca(OH)_2$ which was also observed in other studies [8, 19–22, 24, 29, 31, 32, 34].

The overall evaluation showed no statistically significant difference in the effectiveness of the removal regarding the 3 sections (apical, middle and coronal third) of the root canal (p < 0.05), which is in line with previous findings [8, 9]. On the contrary, there are reports that the removal of Ca(OH)₂ in the apical third was more effective than the removal in the coronal part [24, 29]. In contrast, Silva et al. [31] observed a higher percentage of remaining Ca(OH)₂ in the apical.

This may be explained by the fact that $Ca(OH)_2$ may tend to accumulate apically during the removal procedure [19]. A conical morphology of the root canal with a smaller diameter in the apical region may lead to reduced irrigation efficiency in this area [4, 16].

The instrumentation using a Hedstrom file ISO size 45 before rinsing had no statistically significant effect on removing $Ca(OH)_2$ from the root canal (p > 0.05), which is in accordance with another study [31]. In contrast, Salgado et al. [30] reported an improved removal of $Ca(OH)_2$ compared to irrigation alone when a re-instrumentation was performed with a master apical file.

It is well accepted that NaOCl irrigation leaves significantly more $Ca(OH)_2$ on the root canal walls than rinsing with EDTA or citric acid [18, 33]. This might be explained by the fact that NaOCl has a limited ability to dissolve inorganic substances [3, 34], such as calcium, whereas citric acid and EDTA are decalcifying solutions and undergo neutralization reactions with Ca(OH)₂. Ballal et al. [3] stated that ultrasonically activated 17% EDTA and 10% citric acid solution were able to remove Ca(OH)₂ completely from root canals. However, other studies do not agree with these findings [32]. In the present study these rinsing solutions were not used, as the main question of this study was not to evaluate the solutions itself but to assess the effects of instrument taper and instrumentation before root canal irrigation on the removal of Ca(OH)₂.

The design of the present study which assessed the cleanliness of the complete root canal wall was the same as used in previous investigations [20, 23]. Some other studies used a groove model [11, 19, 24] to standardize the procedure as the location and size of the groove does not vary as much as the natural root canal anatomy. This might be an advantage but disadvantage at the same time, as the groove model seems to be easier and probably more reproducible in evaluating the outcomes when compared to assessing the total surface of the root canal wall. Nevertheless, the complexity of the natural

root canal anatomy and its clinical relevance cannot be replicated in the groove model. One could assume that it may be easier to remove $Ca(OH)_2$ from artificial grooves than from natural isthmuses or irregular surfaces of natural root canal walls, which could possibly lead to an overestimation of the removal efficacy of irrigation solutions.

Conclusion

From the results of this study, it can be concluded that the removal of Ca(OH)₂ is significantly more effective when PUA is used as it leads to less Ca(OH)₂ remnants on the root canal walls (p < 0.05). All irrigation protocols without PUA (NaOCl 2.5%, CHX 2%, NaCl 0.9%) did not show statistically significant differences among each other (p > 0.05). The taper of the root canal preparation before medication with $Ca(OH)_2$ did not influence the results significantly (p > 0.05). The re-instrumentation using a Hedstrom file ISO size 45 before irrigation did not affect the removability of $Ca(OH)_2$ from the root canal significantly (p > 0.05) either. Furthermore, statistically significant differences in the cleanliness of the apical, middle and coronal region of the root canal were not observed (p > 0.05).

Conflicts of interest

The authors deny any conflict of interest related to this study. This research received no external funding.

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