

Evaluation of Calcium Hydroxide Removal Efficiency of Different Concentrations and Forms of Sodium Hypochlorite

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ABSTRACT

The aim of the present study was to compare different forms of sodium hypochlorite for the removal of calcium hydroxide from root canals. Sixty single-rooted mandibular premolars with single canals were prepared using ProTaper Next file up to size X3. Each root canal was filled with calcium hydroxide using #30 Lentulo (Dentsply, Mailfer, Switzerland) to the working length and stored in 100% humidity at 37 °C for 7 days. Afterwards, the teeth were divided into 4 groups according to the irrigation protocol to be used for the removal of calcium hydroxide (n=10): Group 1, 2.5% NaOCl; Group 2, 5.25% NaOCl; Group 3, NaOCl gel; Group 4, Distilled water (control). Samples were split longitudinally, photos of halves were taken at 10× magnification and were analyzed using the Corel DRAW Graphics Suite X4 v14.0 (Corel Corporation, CA, USA) software to calculate the percentage of surfaces with residual calcium hydroxide. Chi-square test was used for making comparisons among the groups at a significance level of $p < 0.05$. There were significant differences between the 2.5% NaOCl solution, 5.25% NaOCl solution, NaOCl gel and the control groups ($p < 0.001$). Gel form of NaOCl is an appropriate and effective method for removing calcium hydroxide from the root canals. Gel form of NaOCl, whose popularity has increased in recent years, was more effective than other groups in removing calcium hydroxide from the root canal. However, none of the 2.5% NaOCl solution, 5.25% NaOCl solution and NaOCl gel were effective in removing the calcium hydroxide completely.

Key Words: Calcium hydroxide removal, Sodium hypochlorite gel, Irrigation solution

Introduction

The microorganisms that are located in the infected root canal can be eliminated through chemomechanical preparation only by 50-70% (1). In order to ensure the bacterial elimination and prevent the recurrence of root canal infection, it is recommended to use intra-canal medicaments following the chemomechanical preparation in multiple session endodontic treatment (2). Calcium hydroxide is the most frequently used intracanal medicament due to its strong antibacterial effect and biocompatibility as well as its properties of dissolving the organic tissue, inducing the formation of hard tissue and inhibiting the osteoclastic activity (3).

In addition to all the advantages of calcium hydroxide, there are certain limitations that negatively influence the success of the root canal treatment. Previous studies reported that calcium hydroxide could not be fully removed from the root canal regardless of the method or irrigation solution used (4, 5). Calcium hydroxide residues

remaining in the dentin tubules prevent the penetration of the sealer into the dentin, thereby reducing the bond strength (6) and increasing microleakage (7). The most commonly used method for removing calcium hydroxide from root canals is agitation with the master apical file of various irrigation solutions along with the instrumentation (8).

Sodium hypochlorite (NaOCl) is the most commonly used irrigation solution during root canal therapy. In addition to its advantages such as antimicrobial activity and organic tissue dissolving ability, it has cytotoxic effects on the periradicular tissues (9). When it is extruded from apical to periradicular tissues during the root canal treatment, it damages endothelial and fibroblast cells, and may lead to development of facial nerve palsy, allergic reactions and necrosis (10). Therefore, researchers have started to search for a more biocompatible irrigant.

The use of NaOCl in gel form has been suggested as a potential option in recent years. Previous studies reported that the solution and gel forms of

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NaOCl had a similar effect on dentin (11, 12). In the study by Zand et al. (11) on the smear layer removal activity of the solution and gel forms of NaOCl, and the study by Garcia et al. (12) evaluating the effects of both forms on dentin microhardness, it was reported that they exhibited similar effects. Therefore, it is believed that the gel form can be used as an alternative to the solution form, especially in cases where apical extrusion risk is high, and the development of the root tip is not completed.

In the literature, there has been no study evaluating the calcium hydroxide removal efficiency of the gel form of NaOCl so far. Therefore, calcium hydroxide removal efficiency of NaOCl solution in different concentrations and the gel form of 5.25% NaOCl was evaluated in this study.

Material and Methods

The study protocol was approved by The Research Ethics Committee (full name of the ethics committee), presenting with statement number 2020/03-12. In this study, 60 single rooted mandibular premolar teeth extracted for orthodontic reasons were used. Teeth without caries and teeth with a single root, a single canal and complete root development were included in the study group. The mesio-distal and buccopalatal radiographs were taken from the teeth, and it was checked whether the teeth had a single canal. The teeth were decoronated with a slow-speed diamond saw under liquid cooling, to obtain the roots in standardized lengths of 12 mm.

Root Canal Preparation: A 10 K-type file was introduced into the root canal until its tip was seen from the apical foramen. Working length was established 1mm shorter from this length. The root canals were instrumented with ProTaper Next file up to X3. At each change of instrument, irrigation was performed with 2mL of 5.25% NaOCl (Imicrly, Konya, Turkey) using a NaviTip irrigation needle for 1 min. After instrumentation, the root canals were irrigated with 2 mL of 17% EDTA (Imident™ Med., Konya, Turkey) for 1 min. The root canals were then rinsed with 5 mL of distilled water and dried with paper points. Afterwards, the calcium hydroxide powder was mixed with distilled water at a 1:1 ratio on cymene paper using a plastic spatula. The mixture that was obtained in the paste consistence was placed until it was visible from the apical foramen, with the help of #30 Lentulo (Dentsply, Mailifer, Switzerland) mounted on an endodontic motor

and operated at the speed of 800 rpm. Root canal access were covered with the temporary filling material (Cavit, 3M ESPE, Germany). Digital radiographic images of each sample were taken, and it was confirmed that the canal space was filled completely with calcium hydroxide. All samples were stored at 37 °C and 100% humidity for a period of 7 days. The samples were randomly divided into 4 groups according to the irrigation protocol (n=10);

Group 1 (2.5% NaOCl): The root canals were irrigated with 5 mL of 2.5% NaOCl.

Group 2 (5.25% NaOCl): The root canals were irrigated with 5 mL of 5.25% NaOCl.

Group 3 (NaOCl gel): Gel form of 5.25% NaOCl (Chloraxid gel, Cercamed, Stalowo Wolo, Poland) was used. Instruments were coated with NaOCl gel during instrumentation. During instrumentation, canals were irrigated with 5 mL saline.

Group 4 (Control): The root canals were irrigated with 5 mL distilled water.

The irrigation procedure was performed with a NaviTip irrigation needle that was placed 2 mm short of the working length in all groups. In the removal of calcium hydroxide, instrumentation was performed with ProTaper Next X3 and #30 Hedstrom files until no medicament was noticeable on the files. The root canals were then rinsed with 5 mL of 17% EDTA for a period of 2 minutes. All the canals were irrigated with 5 mL distilled water, and then they were dried with absorbent paper points.

In order to obtain a buccolingual section, two grooves were made on the buccal and lingual surface of the root with the help of the diamond discs, along the long axis of the root, not extending to the inner wall of the canal. With the help of a spatula, the root was divided into two parts without touching the root canal. The amount of the remnant calcium hydroxide on the root canal walls was evaluated under a stereo microscope with 10× magnification. The photos were viewed on Corel DRAW Graphics Suite X4 v14.0 (Corel Corporation. CA, USA) software. Frames of 2x2 mm were placed in each root canal lumen. The residues of calcium hydroxide in the root canal lumen were evaluated separately for each of the three regions, namely coronal, middle and apical, according to the region where they were located. The data were obtained by proportioning the total number of frames in each region to the number of frames with calcium hydroxide. The amount of calcium hydroxide

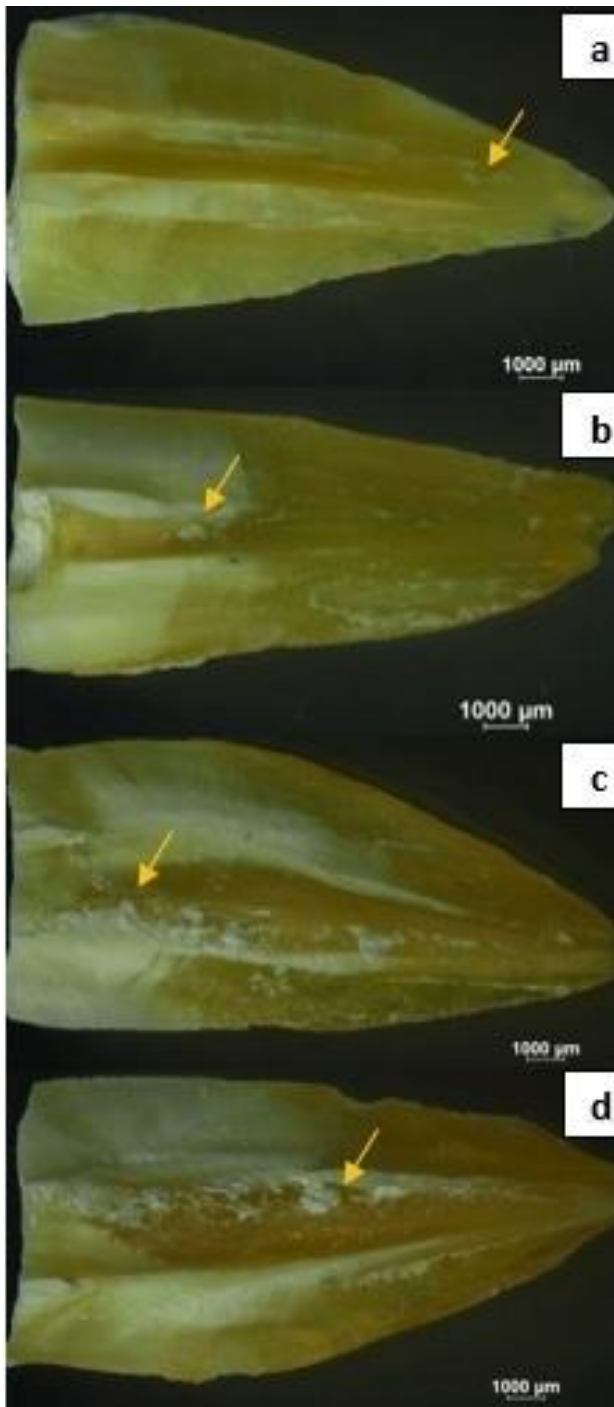


Fig. 1. Scores of calcium hydroxide remnants: (a) score 1 (group sodium hypochlorite gel at apical region); (b) score 2 (group 5.25 % sodium hypochlorite solution at middle region); (c) score 3 (group 2.5 % sodium hypochlorite solution at coronal region); (d) score 4 (group serum solution at middle region)

remaining in root canals, was scored independently by 2 operators calibrated and blunted according to the scoring system proposed by Ok et al. (6) (Fig.1).

Statistical Method: The data were analysed by using IBM SPSS V23. Chi-square test was used for

making comparisons among the groups. The results of the analysis were presented as frequency (percentage) for the categorical data. $P < 0.05$ was considered statistically significant.

Results

There were significant differences between the 2.5% NaOCl solution, 5.25% NaOCl solution, NaOCl gel groups and the control group ($p < 0.001$). The comparisons made between the groups are presented in Table 1. While the hydroxide paste remained in the root canal by 21-50% in the 53.3% of 2.5% NaOCl solution group, in the 66.7% of 5.25% NaOCl solution group, and the 66.7% of NaOCl gel group and the percentage of hydroxide paste in the root canal was 21-50% in 68.3% of the serum group.

There was a statistically significant difference between the distributions of the amounts of remaining calcium hydroxide according to the root regions ($p < 0.001$) (Table 2). While calcium hydroxide paste remained in the root canal by 21-50% in 61.3% of the apical region, the percentage of calcium hydroxide paste was 11-20% in 65% of the coronal region and in 52.5% of the middle region.

In 2.5% NaOCl solution group, there was a statistically significant difference between the distributions of calcium hydroxide remaining in the root canal according to the root regions ($p < 0.001$) (Table 3). While calcium hydroxide paste remained in the root canal by 21-50% in 85% of the apical and 60% of the middle regions, the percentage of calcium hydroxide paste remaining in the root canal was 11-20% in 75% of the coronal region. In the 5.25% NaOCl solution group, there was a statistically significant difference in the distributions of calcium hydroxide remaining in the root canal according to the root regions ($p < 0.001$). While calcium hydroxide paste remained in the root canal by 21-50% in 75% of the apical region, the percentage of calcium hydroxide paste remaining was 11-20% in 95% of the middle region and 80% of the coronal region. In the NaOCl gel group, there was a statistically significant difference between the distributions of the amounts of remaining calcium hydroxide according to the root regions ($p = 0.011$). The calcium hydroxide paste remained in the root canal by 11-20% in 80% of the apical region, in 55% of the coronal region, and in 65% of the middle region. In the control group, there was a statistically significant difference in the distributions of the amounts of remaining calcium

Table 1. Comparison of Calcium Hydroxide Remnants According To Solution Groups

	%2.5 NaOCl solution	%5.25 NaOCl solution	%5.25 NaOCl gel	Serum	Total	Statistical Analysis	P value
Remnants of Calcium Hydroxide							
Score 1	2 (3.3)a	5 (8.3)a	17 (28.3)b	---	24 (10)		
Score 2	23 (38.3)a	40 (66.7)b	40 (66.7)b	15 (25)a	118 (49.2)	$\chi^2=90.506$	<0.001
Score 3	32 (53.3)a	15 (25)b	3 (5)c	41 (68.3)a	91 (37.9)		
Score4	3 (5)	---	---	4 (6.7)	7 (2.9)		

* χ^2 : Chi-square statistical analysis. a-c: There is no difference between groups with the same letter

Table 2. Comparison of Calcium Hydroxide Remnants According To Root Canal Regions

	Apical	Coronal	Middle	Total	Statistical Analysis	p value
Remnants of Calcium Hydroxide						
Score 1	1 (1.3)a	15 (18.8)b	8 (10)b	24 (10)	$\chi^2=52.795$	<0.001
Score 2	24 (30)a	52 (65)b	42 (52.5)b	118 (49.2)		
Score 3	49 (61.3)a	13 (16.3)b	29 (36.3)c	91 (37.9)		
Score 4	6 (7.5)	---	1 (1.3)	7 (2.9)		

* χ^2 : Chi-square Statistical Analysis. a-c: There Is No Difference Between Groups With The Same Letter

hydroxide according to the root regions ($p=0.012$). While calcium hydroxide paste remained in the root canal by 21-50% in 70% of the apical region and 85% of the middle region, the percentage of calcium hydroxide paste remaining in the root canal was 21-50% in 50% of the coronal region, and by 11-20% in 50% of the coronal region. Representative images demonstrating in Figure 1.

Discussion

The Ca (OH)₂ residues remaining in the root canal wall affect the success of root canal treatment negatively. Previous studies reported that the removal of Ca (OH)₂ before root canal obturation increased the penetration of the root canal sealers into the dentin tubules as well as the bond strength between the dentin and sealer (13, 14). Therefore, the Ca (OH)₂ removal activity of NaOCl in different concentrations and forms was evaluated in this study. According to our results, the gel form of NaOCl was more effective compared to the 2.5% and 5.25% NaOCl solutions in terms of the removal of Ca (OH)₂. In addition, differences were obtained in the coronal,

middle and apical regions of the root in terms of the removal of Ca(OH)₂. In all groups, there was a greater amount of Ca (OH)₂ residue in the apical region of the root compared to the coronal and middle regions.

NaOCl is the most common irrigation solution in root canal treatments. NaOCl can dissolve organic tissue during chemomechanical preparation, has a broad-spectrum antimicrobial activity, and can easily diffuse into dentin walls with low surface tension (15). However, despite all these advantages, it has a highly toxic effect on living tissues. Its cytotoxic effects are of clinical concern, particularly when extruded into periapical tissues (10). It is used in root canal treatments in concentrations varying between 0.5% and 5.25%. Previous studies reported that the toxicity and concentration of NaOCl were directly proportional, and it was recommended to be used at the lowest effective concentration (16,17,18).

There are many studies in the literature evaluating the efficacy of NaOCl in different concentrations (16,17,18,19,20). Goldsmith et al. (16) reported that diluting NaOCl up to 2.2% did not create a significant decrease on the tissue dissolving effect;

Table 3. Comparison of Calcium Hydroxide Remnants According To Root Regions of The Solution Groups

Solution Groups	Remnants of Calcium Hydroxide	Apical	Coronal	Middle	Total	Statistical Analysis	P value
%2.5 NaOCl solution	Score 1	---	2 (10)	---	2 (3.3)	$\chi^2=34.133$	<0.001
	Score 2	---	15 (75)	8 (40)	23 (38.3)		
	Score 3	17 (85)a	3 (15)b	12 (60)a	32 (53.3)		
	Score4	3 (15)	---	---	3 (5)		
%5.25 NaOCl solution	Score 1	---	4 (20)	1 (5)	5 (8.3)	$\chi^2=43.35$	<0.001
	Score 2	5 (25)a	16 (80)b	19 (95)b	40 (66.7)		
	Score 3	15 (75)	---	---	15 (25)		
%5.25 NaOCl gel	Score 1	1 (5)a	9 (45)b	7 (35)ab	17 (28.3)	$\chi^2=13.068$	0.011
	Score 2	16 (80)	11 (55)	13 (65)	40 (66.7)		
	Score 3	3 (15)	---	---	3 (5)		
Serum	Score 2	3 (15)ab	10 (50)b	2 (10)a	15 (25)	$\chi^2=12.905$	0.012
	Score 3	14 (70)	10 (50)	17 (85)	41 (68.3)		
	Score4	3 (15)	---	1 (5)	4 (6.7)		

* χ^2 : Chi-square Statistical Analysis. a-b: There Is No Difference Between Groups With The Same Letter

however, the effect of 0.5% NaOCl solution was insufficient. Zehnder et al. (17) reported that NaOCl was effective on necrotic pulpal tissue even at concentrations as low as 1%. Baumgartner and Cuenin (18) reported that 0.5%, 1%, 2.5% and 5.25% concentrations of NaOCl could completely remove pre dentin and pulpal tissue residues from the unformed canal wall. Ayhan et al. (19) compared the antimicrobial efficacy of 0.5% and 5.25% NaOCl on different microorganisms and found that the 0.5% concentration had a significantly lower effect. Siqueira et al. (20) also stated that 1%, 2.5% and 5.25% NaOCl were effective in the elimination of *Enterococcus faecalis*, and this effect increased with the increase in concentration. In the literature, there have been no studies evaluating the Ca (OH)₂ removal efficiency of 2.5% and 5.25% NaOCl solution and the gel form of NaOCl so far. Therefore, NaOCl was used in 2.5% solution and gel forms, with increasing popularity in recent years.

The gel form of NaOCl was more effective compared to the 2.5% and 5.25% NaOCl solutions in terms of the removal of calcium hydroxide. Hydroxide paste remained in the root canal by 21-50% in the 53.3% of 2.5% NaOCl solution group, in the 66.7% of 5.25% NaOCl solution group, and in the 66.7% of the NaOCl gel group. There is no study in the literature to compare the results obtained in the present study. Instruments were coated with NaOCl gel during instrumentation and used inside the root canals. Accordingly, we believe that NaOCl was more effective in removing calcium hydroxide since it

had a greater contact with root canal walls. Turkun et al. (21) reported that, in addition to its cytotoxic and irritating properties, tissue dissolving and antibacterial effects of NaOCl were significantly reduced when used at lower concentrations. In the present study, 2.5% NaOCl removed less calcium hydroxide compared to 5.25% NaOCl, and the data obtained were consistent with the literature. However, we believe that further in-vitro studies are required on the Ca (OH)₂ removal capacity of NaOCl solutions at low concentrations through various activation techniques.

An important point that should be evaluated in the present study was that there was a greater amount of calcium hydroxide residues in the apical third of the root than the coronal and middle thirds in all groups. The results obtained were consistent with the studies in the literature (6, 7, 22). The reason why the irrigation solutions exhibited less efficacy in the apical region could be attributed to the narrower diameter of the apical region of the canal, and thus, the decrease in the volume of solution reaching this region (23). This is due to the fact that the effectiveness of the solutions used in endodontics depends on the maximum contact with the canal walls. In addition, the anatomical structure and stenosis of the root canal may cause air bubbles to remain in the apical region during irrigation and prevent the irrigation to extend to the apical region. As a result, complete clearance of the apical region may not be achieved, and this would prevent an optimum treatment (24). Besides, when the root

regions were evaluated according to the groups in the present study, it was found that the gel form of NaOCl was more effective in the apical region compared to the solution forms. We believe that this may result from the fact that the solutions may have been prevented from reaching the apical region due to air bubbles formed during irrigation; however, the gel form is in greater contact with the canal walls directly through the file.

One of the most limitation of this study was that the residual Ca (OH)₂ on the dentin walls was determined under the stereomicroscope. In the literature, stereomicroscope, SEM and Micro-CT were used to calculate the residual Ca (OH)₂ remaining in the root canal (6,7,14,22,23). However, when using a stereomicroscope, only superficial residues can be measured, and residues in dentinal tubules cannot be calculated (6). Therefore, further studies with a three-dimensional view or advanced microscopy techniques are needed in the evaluation of residual Ca (OH)₂ residues.

Within the limitations of this study, Ca (OH)₂ could not be removed completely in any experimental group. The gel form of NaOCl was more effective compared to the 2.5% and 5.25% NaOCl solutions in terms of the removal of Ca (OH)₂. In addition, there was a greater amount of Ca (OH)₂ residue in the apical third of the root compared to the coronal and middle thirds in all groups. In all groups, the gel form of NaOCl was more effective in removing Ca (OH)₂ in the apical region. In conclusion, we believe that the gel form of NaOCl is an appropriate and effective method for removing Ca (OH)₂ from the root canals.

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