## Removal of Radioactively Marked Calcium Hydroxide from the Root Canal: Influence of Volume of Irrigation and Activation

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#### Abstract

Introduction: The purpose of this study was to evaluate the amount of calcium hydroxide (Ca[OH]<sub>2</sub>) removed by irrigation with different volumes and activation methods. Methods: One hundred thirty extracted straight, singlerooted human teeth were instrumented to size 45/.04. One hundred twenty teeth were filled with radioactively marked Ca(OH)<sub>2</sub> and a gutta-percha point; 10 teeth with only gutta-percha served as a negative control. All specimens were stored in saline solution (7 days at 35°C). After storage, teeth were randomly divided into 12 groups (n = 10). The gutta-percha was taken out, and Ca(OH)<sub>2</sub> was removed either by irrigation with different volumes (0 mL, 0.5 mL, 1 mL, 2 mL, 4 mL, or 8 mL) or mechanical activation with a 2- or 4-mL volume using a file (Instr) (Flex-Master size 45/.04; VDW, Munich, Germany), a brush (CanalBrush [CB]; Coltène/Whaledent, Langenau, Germany), or passive ultrasonic irrigation (PUI, smooth wire). Irrigation was performed by alternating 40% citric acid and 3% sodium hypochlorite. Residual Ca(OH)<sub>2</sub> was measured by scintillation and expressed as a percentage of the original Ca(OH)<sub>2</sub>. Results: Increasing the irrigation volume led to a significant decrease (P < .05) of residual Ca(OH)<sub>2</sub> (0 mL [98.5%], 0.5 mL [21.7%], 1 mL [16.5%], 2 mL [12.9%], 4 mL [8.7%], 8 mL [5.0%], and negative control [0.0%]). Activation led to less residual Ca(OH)<sub>2</sub> (2 mL Instr [12.0%], 2 mL CB [11.7%], 2 mL PUI [9.1%], 4 mL Instr [8.5%], 4 mL CB [7.4%], and 4 mL PUI [6.2%]), with significant differences according to the PUI (P < .05). Conclusions: No irrigation procedure was able to remove Ca(OH)<sub>2</sub> completely. PUI was the most effective activation method. However, irrigation with an 8-mL volume was the most effective. (J Endod 2016;42:637-640)

#### Key Words

calcium hydroxide, CanalBrush, intracanal medicament, passive ultrasonic irrigation, radioactively marked glucose

The placement of an intracanal medicament is necessary when the endodontic treatment cannot be performed in 1 appointment (1). Furthermore, intracanal medication is 1 of the treatment strategies for an infected root canal system (2, 3). Calcium hydroxide suspension (CHS) is 1 of the commonly used intracanal medicaments because of its basic pH and biocompatibility (4).

Although it is 1 of the materials of choice for intracanal medicament, the complete removal of CHS from the root canal is difficult. In 1997, Margelos et al (5) showed that 25%–75% of the root canal surface was still covered with CHS depending on the irrigation regimen and the use of an endodontic instrument. Furthermore, they reported a chemical interaction of this residual material with a zinc oxide– eugenol sealer, inhibiting its chelate formation and resulting in brittle consistency and a granular structure. In 1999, Lambrianidis et al (6) found that a considerable part of the root canal wall (25%–45%) remained touched by CHS depending on the methods of calcium hydroxide removal. Only a recent study (7) reported that CHS could be completely removed using a special device based on a broad spectrum of sound waves.

Regarding the choice of a particular irrigation solution for the removal of calcium hydroxide, chelators worked best in most of the studies (8–12). Besides this, 2 different strategies can be performed for better removal of CHS: increasing the volume of the irrigation solution, a topic that was rarely examined in the past, and using different modes of activation of the solution. One problem with most of the existing studies is that the measurement of residual material is based on different scoring systems or a quantitative measurement of the surface of the residual calcium hydroxide and not the volume (5, 6, 8–20). Contrary to this, in the present study, a full quantitative measurement of the residual volume of the intracanal medicament was chosen by using radioactively marked CHS.

The aim of the present study was to perform a full quantitative measurement of the residual CHS after removal with different volumes of irrigation and different modes of activation. The 2-fold null hypothesis tested was that there is no significant difference regarding different volumes of irrigation and different activation methods.

#### Teeth

### **Materials and Methods**

One hundred thirty freshly extracted human teeth with a single straight root were selected for this study. The teeth were stored in 0.1% chloramine-T solution for 2 to 4 weeks and kept frozen until usage. A number of 10 teeth per group was chosen

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for practical reasons and because in former studies this number was sufficient to detect significant differences.

#### **Root Canal Instrumentation**

Before canal instrumentation, the teeth were trimmed to equal lengths of 15 mm. All canals were rotary instrumented to size 45/.04 with FlexMaster instruments (VDW, Munich, Germany) at a working length of 1 mm from the anatomic apex (working length = 14 mm). Irrigation was performed with 40% citric acid (Pharmacy Universitätsklinikum Erlangen, Erlangen, Germany) and 3% sodium hypochlorite (NaOCl) (Hedinger, Stuttgart, Germany) between each instrumentation. A final irrigation with 2 mL 40% citric acid, 2 mL 3% NaOCl, and 2 mL 70% ethanol (Pharmacy Universitätsklinikum Erlangen) was performed for each specimen. Then, the root canals were dried with paper points. Finally, the apical end of each specimen was covered with flowable composite (Grandio Flow WO; VOCO, Cuxhaven, Germany) and light cured (40 seconds at 800 mW/cm<sup>2</sup>; Elipar Trilight, 3M ESPE, Seefeld, Germany) to avoid a possible loss of intracanal medicament through the apex. Grooves were cut on the buccal and lingual surface, paying close attention not to perforate the root canal. This procedure was needed to be able to perform the freeze fracturing process later on.

#### **Intracanal Medication Preparation and Placement**

For each specimen, 197 mg CHS (Calxvl; OCO, Dirmstein, Germany) was mixed with 5  $\mu$ L<sup>14</sup>C-glucose (D-[14C(U)]-Glucose; PerkinElmer, Boston, MA). From each mixture, 3 samples of 20 mg each were mixed with 10 mL scintillation liquid (Rotiszint eco plus; Carl Roth, Karlsruhe, Germany) for measurement within a scintillation counter (LKB Wallac 1409 Liquid Scintillation Counter; EG&G Berthold, Bad Wildbach, Germany). The 3 values of the samples were averaged to serve as a standard for radioactive labeling. Before placement of the CHS, each tooth was weighed with a precision balance (R160P; Sartorius, Göttingen, Germany) together with the respective gutta-percha point size 40/.02 (Antaeos Guttapercha 525, VDW) cut to a length of 14 mm. The gutta-percha served to push the CHS tight to the canal wall. The radioactively labeled CHS was placed in the root canal with a Lentulo spiral (Lentulo size 25; Dentsply Maillefer, Ballaigues, Switzerland). Afterward, the gutta-percha point was placed. Excess CHS was wiped off with a foam pellet. Then, each sample was weighed again for the calculation of the amount of CHS. Ten teeth filled only with a gutta-percha point served as the negative control group. After this, flowable composite was placed over the medicated root canal (Grandio Flow WO) and light cured (40 seconds at 800 mW/cm<sup>2</sup>, Elipar Trilight). Each sample was stored in saline solution (Jonosteril; Fresenius Kabi, Bad Homburg, Germany) in a separate cup at 35°C for 7 days.

#### **CHS Removal**

The teeth were divided into 12 groups of 10 teeth each. After storage, the coronal flowable composite was removed with a scaler. The gutta-percha point was removed, and the root canal was irrigated following different irrigation regimens (0 mL, 0.5 mL, 1 mL, 2 mL, 4 mL, or 8 mL; Table 1) using a 30-G endodontic needle (Transcoject, Neumünster, Germany). For the 0.5-mL group, irrigation with 0.25 mL 40% citric acid was followed by 0.25 mL 3% NaOCl. For the other groups, 0.5-mL increments were performed (alternating citric acid with NaOCl) until the whole irrigation volume was achieved. Care was taken so that the needle did not exceed a length of 2 mm short of the working length. In the groups with activation, the latter was performed 2 times for 20 seconds, once following the first increment of irrigation and again after the second. The master apical file (FlexMaster instrument size 45/.04) was used at 350 rpm to the working length. The Ca-

#### TABLE 1. Division into Different Groups within the Present Study

1	
Removal method	Total irrigation volume (mL)
Irrigation	0 (positive control)
-	0.5
	1
	2
	4
	8
Activation and irrigation	
FlexMaster	2
CanalBrush	
PUI	
FlexMaster	4
CanalBrush	
PUI	

PUI, passive ultrasonic irrigation.

Ten teeth without calcium hydroxide suspension served as the negative control group.

nalBrush size M (Coltène/Whaledent, Langenau, Germany) was used in rotary motion at 350 rpm to the working length. Passive ultrasonic irrigation (PUI) was performed with a Piezon Master 600 (power setting level 4, endodontic mode; EMS, Nyon, Switzerland) equipped with a smooth wire at 1 mm short of the working length.

Finally, the root canals were dried with paper points (40/.02 Paperpoints, Coltène/Whaledent). The specimens of the control groups were directly referred to the freeze fracturing process without further treatment.

#### **Residual CHS Measurement**

All specimens were freeze fractured following the method by Petschelt and Oberschachtsiek (21) and placed into scintillation flasks together with 10 mL scintillation liquid (Rotiszint eco plus). Measurements were performed with a liquid scintillation counter (LKB Wallac 1409 Liquid Scintillation Counter) after 10 days of storage to allow complete dissolution of the radioactively marked substance. Measurements of the respective standards were also taken at this time. The amount of residual radioactivity was calculated relative to the radioactivity that was put into the root canals. Measurements were expressed as a percentage relative to the original suspension.

#### **Statistical Analysis**

Data were analyzed with IBM SPSS 21.0 (IBM SPSS Inc, Chicago, IL) using Kolmogorov-Smirnov tests, 1-way and 2-way analysis of variance, and Student-Newman-Keuls (SNK) post hoc tests ( $\alpha = 0.05$ ).

#### Results

Data of groups were normally distributed (Kolmogorov-Smirnov tests, P > .05). Thus, parametric tests could be applied. The negative control group had a count rate of 0.000116% (range, 0.000091%-0.000205%) of the original suspension, whereas the positive control group had a count rate of 98.55% (range, 97.6%-99.8%). Two-way analysis of variance revealed a highly significant influence of both irrigation volume and activation method (both P < .001) but not of their combination (P = .585). All of the groups with different irrigation volume and no activation showed significant differences to all other respective groups (SNK, P < .05). The amount of irrigation liquid showed a negative proportionality to the residual CHS (Pearson correlation coefficient = -0.826, P < .001). PUI showed the greatest effect on the reduction of residual CHS. PUI was the only activation method with significant influence on results with both irrigation volumes tested (SNK, P < .05). Results of different experimental groups are shown in Figures 1 and 2.

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