ORIGINAL ARTICLE
DOI:10.5152/EssentDent.2025.24013

ISTANBUL UNIVERSITY CERRAHPASA

Evaluation of Calcium Hydroxide Removal from Simulated Internal Resorption Cavities by Different Irrigation Techniques

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Cite this article as: Kürkçü O, Haznedaroğlu F, Kutlu Basmacı G. Evaluation of calcium hydroxide removal from simulated internal resorption cavities by different irrigation techniques. *Essent Dent.* 2025; 4, 0013, doi: 10.5152/EssentDent.2025.24013.

Abstract

Background: Calcium hydroxide (CH) is a material that is very difficult to remove, especially in complex root canal anatomies and pathologic formations such as internal root resorption (IRR). The purpose of this study is to compare ultrasonic and sonic irrigation activation systems, as well as XP-endo Finisher, in terms of their efficiency in removing 2 forms of CH from simulated IRR cavities.

Methods: Fifteen human maxillary central incisors were used for this study. The teeth were cut from their crowns to standardize the root lengths at 15 mm. After preparing root canals, the roots were split into 2 halves vertically using a diamond disc, and artificial resorption cavities were prepared 5 mm above the root apex. Two forms of CH were applied into the root canals and resorption cavities. Tested methods were manual irrigation, XP-endo Finisher, EDDY, and passive ultrasonic activation (PUI). After the experiments, samples were evaluated under a stereomicroscope at ×20 magnification and scored via pixel count.

Results: Residual CH was observed in all samples after experiments. Passive ultrasonic activation revealed the best median scores, yet there was no significant difference between PUI and EDDY ($P > .05$). Passive ultrasonic activation was statistically superior to XP-endo Finisher ($P < .05$). Passive ultrasonic activation, EDDY, and XP-endo Finisher revealed superior results compared to manual irrigation ($P < .05$). There was no significant difference between CH prepared with distilled water and its injectable form (UltraCal XS) in terms of removability from simulated IRR cavities.

Conclusion: None of the tested methods was able to remove CH completely from the simulated internal root resorption cavities.

Keywords: Activation, calcium hydroxide, irrigation, pixel count, simulated internal root resorption cavity

What is already known on this topic?

- Calcium hydroxide is an intracanal medicament with proven efficacy, but it is highly controversial in terms of its removal from canals, especially irregular anatomical structures such as internal resorption cavities
- It has been proven in the literature, including the results of this study, that manual irrigation alone is insufficient to remove calcium hydroxide from internal resorption cavities.

What this study adds on this topic?

- Although it is impossible to completely remove calcium hydroxide from internal resorption cavities no matter which activation method is used, this study shows that EDDY sonic activation is as effective as passive ultrasonic activation in removing calcium hydroxide from internal resorption cavities.
- Considering the results of this study, it is very important to utilize irrigation activation methods with proven efficacy in the literature, such as passive ultrasonic activation, EDDY sonic activation and XP-Endo Finisher, to remove calcium hydroxide from irregular anatomical irragularities such as internal resorption cavities.

INTRODUCTION

Root resorption is an inflammatory process of the tooth that activates odontoclastic cells and leads to the loss of hard tissue.[1,2] Root resorptions are defined as internal or external depending on the localization of the resorption cavity.[1] Internal root resorptions (IRRs) are related to conditions such as trauma, inflammations, pulp infection, and autotransplantation.[3,4] Although IRR lesions are hard to diagnose because of their

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Received: May 6, 2024
Revision Requested: July 16, 2024
Last Revision Received: August 23, 2024
Accepted: November 18, 2024
Publication Date: January 20, 2025

asymptomatic nature, it is important to treat the disease in the early stages in order to prevent further hard tissue loss and even perforation of the root.[1,4] In addition to that, early diagnosis and treatment of IRR provide a better prognosis.[5]

Complex anatomy of IRR lesions causes difficulties and challenges during nonsurgical root canal treatment, such as debridement and proper filling of the pulp space, since instruments are not able to touch all the surfaces of the lesions.[3,6] Thus, the irregular structure of IRR lesions makes using irrigant agitation methods and calcium hydroxide (CH) placement necessary for effective disinfection, bleeding control, and removal of granulation tissue.[3,6,7] However, the complete removal of CH from root canal walls is another challenge that must be overcome. Because residual CH affects the physical properties of endodontic sealers, prevents their penetration into dentinal tubules, and finally may lead to the failure of root canal treatment.[8,9] Furthermore, residual CH may show solubility and dimensional instability, affecting the sealing of the root canal filling, which may result in treatment failure.[10] Calcium hydroxide that is not removed from irregular areas in root canals, such as IRR cavities, can also cause electronic apex locators to show inaccurate results.[11] For these reasons, it is very important to remove CH from root canals as much as possible.

Conventional irrigation techniques do not provide effective CH removal from root canal walls and IRR cavities. For this reason, additional devices and techniques have been studied and presented by developers over the years to maximize the effect of irrigation solutions such as sodium hypochlorite (NaOCl) and ethylene diamine tetraacetic acid (EDTA).[7,9,12]

Passive ultrasonic activation (PUI) is commonly used for the activation of irrigation solutions and considered more efficient than conventional irrigation with a syringe.[13] Ultrasonic tips vibrate at high frequency, so they create cavitations and acoustic transmissions in the irrigation solutions, which ensure more efficient cleaning of the root canal system.[13,14]

EDDY is a sonic activation device with a polyamide polymer tip, which has a size 25 and 0.04 taper.[6] EDDY has 6000 Hz frequency, which allows oscillation at high amplitude, causing cavitations and acoustic transmission according to the manufacturer.[6,15]

XP-endo Finisher is an instrument used for irrigant agitation, made from nickel-titanium alloy called MaxWire. It has a size 25 with zero taper. The alloy of this instrument changes its phase from martensitic to austenitic at body temperature and adapts itself to the root canal's morphology.[6,15,16]

The purpose of this study is to compare 4 different irrigation techniques in terms of their efficiency in removing 2 forms of CH from simulated internal resorption cavities. These 4 irrigation techniques are manual irrigation with syringe and needle, mechanical agitation with XP-endo Finisher, sonic activation with EDDY, and PUI. The null hypothesis tested was that XP-endo Finisher would be as efficient as the other tested methods.

MATERIALS AND METHODS

The manuscript of this laboratory study has been written according to Preferred Reporting Items for Laboratory Studies in Endodontology (PRILE) 2021 guidelines (Figure 1).[17] This study's research protocol was approved by the İstanbul

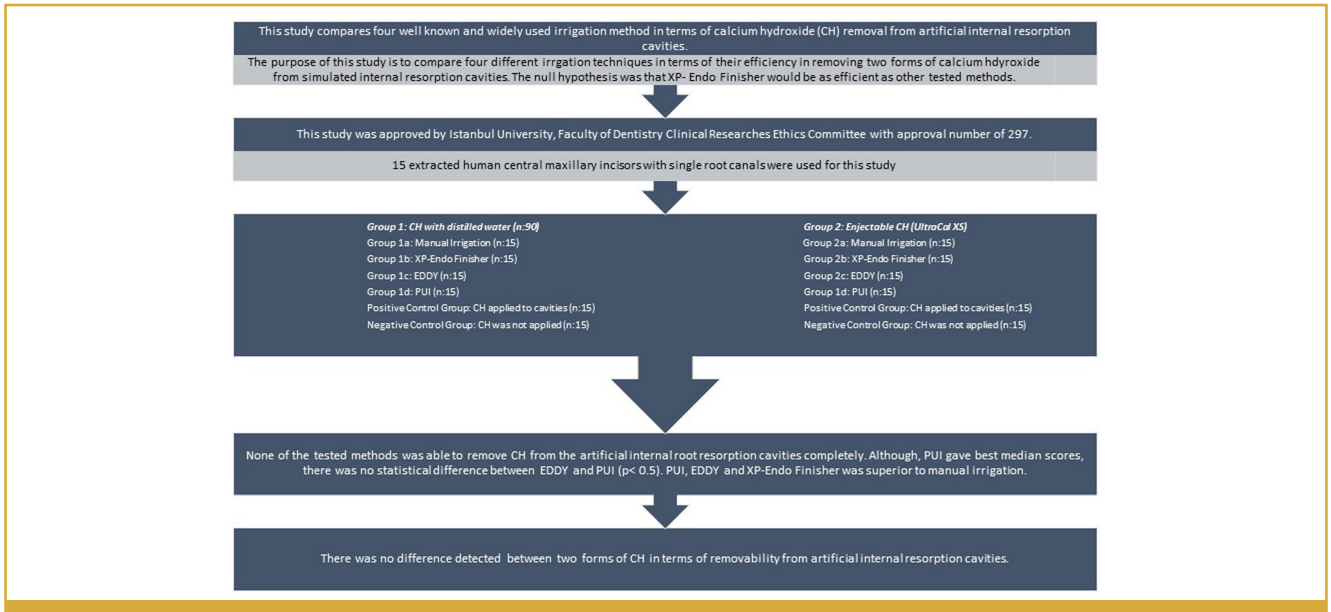

Figure 1. PRILE 2021 flowchart for the current study.

University, Faculty of Dentistry Ethics Committee Board (Approval no: 297, Date: June 7, 2018). Consent forms were obtained from the individuals from whom the samples were obtained.

Selection of Samples

Fifteen extracted human central maxillary incisors with a single root canal were used for this study. The samples were examined under an operational microscope, and radiographs were taken from buccolingual and mesiodistal directions. Teeth with caries, calcifications, cracks, fractures, root canal treatments, or resorptions were excluded. Samples were stored in purified filtered water until the time they were used for the experiment. The same 15 samples were used in all experimental and control groups using a special system.

Preparation of Samples

The crowns of teeth were partially removed and the length of roots was standardized at 15 mm. After the preparation of endodontic access cavities, a size 20 K-file (Dentsply Maillefer, Ballaigues, Switzerland) was inserted into root canals until it was visible at the apical foramen, and then working lengths were determined by subtracting 1 mm from that measurement. Root canals were instrumented using a Reciproc R50 file (size 50, taper 0.05, VDW, Munich, Germany), and root canal space was kept full with 5.25% NaOCl solution during instrumentation in order to keep the root canal space moist. The irrigant was refreshed with 2.5 mL NaOCl using a 29-Gauge needle (Endo-Eze, Ultradent, South Jordan, UT) after every 3 mm advancement. After mechanical preparation, root canals were irrigated with 5 mL 5.25% NaOCl, 5 mL distilled water (dw), 5 mL 17% EDTA, and finally 10 mL dw. Then, root canals were dried with paper points.

After mechanical preparation of the root canals, samples were split along their long axis in the bucco-lingual direction using a diamond disk (4" × 0.12" × 1/2 Diamond Wafering Blade Sintered, Metkon) attached to an Isomet device (Isomet 4000, Buehler, IL, ABD). Separation of the samples was done in a single move by centering the root canals as much as possible. In this way, the re-adaptation of the straight root walls to each other was aimed to be ensured. Samples where the root canals could not be centered or the root halves could not be re-adapted to each other were excluded from the study. Afterward, artificial IRR cavities positioned 5 mm above the anatomical apex in both halves of the root canals were prepared using a diamond round bur. These artificial IRR cavities were prepared with a diameter of 1.6 mm and a depth of 0.8 mm. The root halves were then re-adapted to each other and fixed with a thin layer of utility wax along the cut line. Afterward, samples were embedded in acrylic molds with a collapsible screw system. This system, which enables the adaptation of 2 root halves and artificial internal resorption cavities to each other, can be defined as a modification to Bremente's Muffle System.[18] Representative images of this system are shown in Figure 2. In this way, it

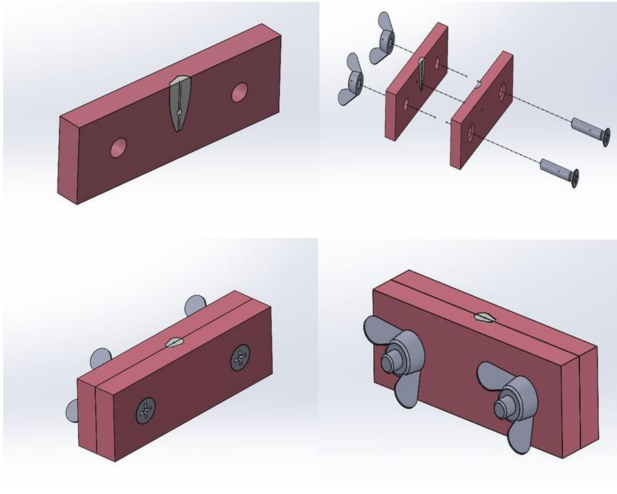

Figure 2. Representative image of the system.

was ensured that the experiments were carried out using the same samples for all groups, resulting in 60 samples for each main group (n=120), 15 samples for each positive control group (n=30), and 15 samples for 1 negative control group, 165 in total.

In order to mimic the irregular structure of the IRR cavities, 37% orthophosphoric acid (Dia-Etch, DiaDent, Seoul, Korea) was applied to the artificial internal resorption cavities for 30 seconds. The bonding agent (3M ESPE, Seefeld, Germany) was applied with a micro applicator (Micro Brush, WI, USA) to the dentin surfaces of both parts of the models facing each other, except root canal walls, and polymerized using a curing light (Valo, Ultradent, SJ, USA) for 20 seconds. In this way, the leakage of irrigation solutions and CH from the root canals of the samples along the cut line was prevented. These procedures were repeated for every application after cleaning the root canals and resorption cavities. Root canals and resorption cavities were cleaned using compressed air-water spray, a microbrush, and an excavator.

Groups and Irrigation Protocol

In this study, there are 11 groups in total, including 1 negative control group and 2 positive control groups. Four experimental groups (Conventional Irrigation, Xp-endo Finisher, EDDY, and PUI) were created for each of the 2 main groups. Experimental and control groups are shown in Figure 3. Fifteen samples were used for all control and experimental groups.

In the first main group, CH powder (Sultan, Sultan Healthcare, Hackensack, NJ) was mixed with dw at a ratio of 1:1.5 and placed into root canals and artificial resorption cavities using paper points. In the second main group, UltraCal XS (Ultradent, South Jordan, UT) was placed in root canals and artificial resorption cavities using a Navi-Tip 29 G needle (Ultradent,

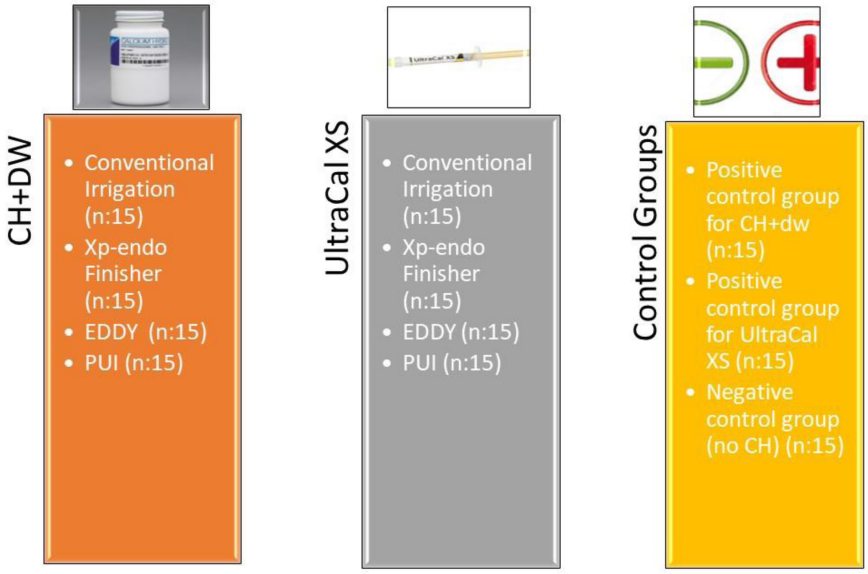

Figure 3. Experimental and control groups of the study.

South Jordan, UT). In the positive control groups, CH placement was done but no irrigation or activation was performed. In the first positive control group, CH powder and dw were mixed and applied to root canals and artificial resorption cavities (n=15). In the second positive control group, UltraCal was applied to root canals and artificial resorption cavities (n=15). In the negative control group, the samples were left empty and CH was not applied (n=15). The samples exist in the system in 2 halves of the root canal form. In this way, it was easy to visually photograph that the canal and the artificial simulated internal resorption cavities were completely filled with CH. Photographs were taken for each sample, and representative images of the experimental and control groups are shown in Figure 4. After CH placement, samples were closed with a screw system and sealed with temporary restorative material (Cavit-W, 3M Espe, Seefeld, Germany). Samples were kept at 37°C and 100% humidity for 1 week.

At the end of 1 week, irrigation and agitation procedures were performed after removing temporary restoration material and loosening CH in the root canals with a #15 K-file. Firstly, irrigation with 2 mL of 5.25% NaOCl was done, and 20 seconds

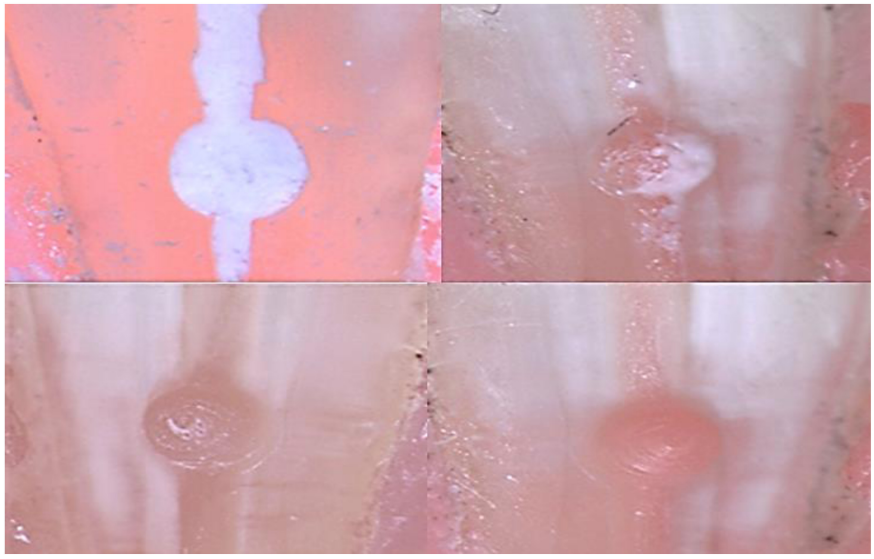

Figure 4. Representative images of samples from experimental and control groups.

agitation was performed except for manual irrigation. This process was repeated 3 times, and 6 mL of NaOCl was used in total. After NaOCl irrigation, root canals were flushed with 2 mL of dw. Afterward, root canals were irrigated with 2 mL of 17% EDTA and then agitated for 20 seconds. This process was repeated 3 times, and 6 mL of EDTA was used in total. Finally, root canals were flushed with 10 mL of dw and dried with paper points. As a consequence, 6 mL of NaOCl, 6 mL of EDTA, and 12 mL dw were used for each sample. Agitation time was 2 minutes (1 minute for NaOCl and 1 minute for EDTA) for all samples except for manual irrigation. In manual irrigation groups, irrigation was performed similarly to other groups, but no agitation was performed except needle movement. Radiographs were taken from all samples to confirm that no leakage occurred during irrigation or agitation processes. All experiments were performed at 20°C, and each instrument was used 3 times before they were discarded.

Agitation of irrigation solutions was performed for each group as follows:

- Manual irrigation (n = 15 for group CH + dw and n = 15 for group UltraCal):

Twenty-nine G Navi-Tip needle was inserted into root canals 1 mm shorter than the working length. Irrigation was done with in-and-out movements that had 5 mm of amplitude.

- XP-endo Finisher (n = 15 for group CH+dw and n = 15 for group UltraCal):

XP-endo Finisher kit attached to the VDW Silver Reciproc engine (VDW, Munich, Germany) was used at a speed of 800 rpm and a torque of 1 N. The kit was used at the working length with 7-8 mm vertical movements as the manufacturer advised.

- EDDY (n = 15 for group CH+dw and n = 15 for group UltraCal):

EDDY tip (size 25, 0.04 taper) attached to Kavo Sonic Borden system (Kavo, Charlotte, USA) was inserted into root canals 1 mm shorter than the working length and used with 2-4 mm vertical movements at a frequency of 6000 Hz as the manufacturer advised.

- PUI (n = 15 for group CH+dw and n = 15 for group UltraCal):

Ultrasonic tip with a size of 15 and 0.02 taper (ESI instrument; EMS, Le Sentier, Switzerland) attached to a piezoelectric ultrasonic unit (EMS, Switzerland) inserted into root canals 1 mm shorter than the working length and used with a power setting of 6.

After irrigation and agitation procedures were performed, screw systems were opened and artificial resorption cavities were photographed with Nikon Coolpix digital camera (Nikon, Tokyo, Japan) attached to a stereomicroscope (BX60, Olympus, Tokyo, Japan) at ×20 magnification. All photos were taken with a standardized angle. Evaluation of CH remnants was done by pixel count using Adobe Photoshop Programme 23.5.2 and recorded as the percentage of overall surface area. Magnetic Losso Tool and histogram were used to determine the border of resorption cavities and pixels with CH remnants.

Python 3.10 program was used in the statistical evaluation of the findings. Friedman and Dunn's tests were performed to determine the differences between tested methods. The Wilcoxon test was used to compare the 2 forms of CH. Significance was evaluated at the $P < .05$ level.

RESULTS

Wilcoxon test revealed that there was no significant difference between 2 forms of CH in terms of removability ($P > .05$). As a consequence, the results obtained from the comparisons made between the experimental groups of both main groups (Group CH + dw and Group UltraCal) overlapped with each other. Positive control groups confirmed that there was no CH loss from root canals and artificial IRR cavities during storage or transportation.

None of the tested methods was able to remove CH from the artificial IRR cavities completely. There was no significant difference between the positive control groups and Manual irrigation ($P > .05$), also between PUI and the negative control group ($P > .05$). XP-endo Finisher, EDDY, and PUI removed significantly more CH than Manual irrigation ($P < .05$). Although PUI had the best median scores, there was no significant difference between PUI and EDDY ($P > .05$). There was a significant difference between PUI and XP-endo Finisher ($P < .05$). There was no significant difference between EDDY and XP-endo Finisher although EDDY revealed better

Table 1. ***P*** **Values Obtained Using Dunn's Test for the Positive and Negative Control Groups of Group CH + dw and the manual irrigation, EDDY, and PUI Experimental Groups**

Groups	CH + dw–Manual Irrigation	CH + dw–XP-endo Finisher	CH + dw–EDDY	CH + dw–PUI	Negative Control	Positive Control
CH + dw–Manual irrigation	1	**.049**	**.0007**	**.0003**	**3.00E−10**	.1158
CH + dw –XP-endo Finisher	**.049**	1	.1621	**.0319**	**.000015**	**.00405**
CH + dw–EDDY	**.0007**	.1621	1	.4545	**.0034**	**8.00E−07**
CH + dw–PUI	**.000039**	**.03191**	.4545	1	**.029225**	**1.00E−08**

CH, calcium hydroxide; dw, distilled water; PUI, passive ultrasonic activation. Values in bold indicate statistical significance.

Table 2. ***P*-Values Obtained Using Dunn's Test for the Positive and Negative Control Groups of Group UltraCal and the manual irrigation, EDDY, and PUI Experimental Groups**

Groups	UltraCal–Manual Irrigation	UltraCal–XP-endo Finisher	UltraCal–EDDY	UltraCal–PUI	Negative Control	Positive Control
UltraCal–Manual Irrigation	1	**.0456**	**.0012**	**.000007**	**2.00E-10**	.16431
UltraCal–XP-endo Finisher	**.0456**	1	.216	**.0133**	**.000011**	**.0007**
UltraCal–EDDY	**.0012**	.2160	1	.216	**.0016**	**3.00E-06**
UltraCal–PUI	**.000007**	**.0133**	.216	1	.0564	**4.50E-09**

PUI, passive ultrasonic activation. Values in bold indicate statistical significance.

median scores ($P > .05$). In both group CH+dw and group UltraCal, statistical analyses revealed similar results for tested methods except for small median differences. The statistical analysis and *P* values of the sub-experimental groups of the 2 main groups (CH+dw and UltraCal) among themselves and with the control groups are shown in Tables 1 and 2.

DISCUSSION

In cases of IRR, the application of CH as an intracanal medicament is required, along with agitation of irrigation solutions.[7] It is reported that there is no successful irrigation or agitation method in terms of complete removal of CH from root canal irregularities.[13,19,20] Therefore, many agitation or activation methods and devices have been studied in recent years. [7,9,12] In this study, various activation methods such as Xp-endo Finisher, PUI, and EDDY were tested for the removal of 2 different CH pastes from simulated internal resorption cavities. The main finding of this study is that all experimental methods except manual irrigation revealed superior results compared to positive control groups. The null hypothesis was rejected as XP-endo Finisher revealed higher remaining CH scores than all experimental methods, especially PUI.

Former studies that investigated CH removal from artificial IRR cavities commonly used burs to create standardized cavities.[7,9] However, da Silveira et al[2] reported that creating artificial IRR cavities using 5% nitric acid is a better way to mimic the irregular structure of IRR lesions. Marques-da-Silva et al[6] reported that activation methods revealed better results when cavities were created by using burs in comparison to burs and nitric acid combination. In this study, artificial IRR cavities were created by using burs and etched with 37% orthophosphoric acid before each application.

According to the studies on removing CH from root canals, the combined use of NaOCl and EDTA solutions gives more successful results than using only one of them.[21] Therefore, in this study, NaOCl and EDTA were used in combination.

In most of the studies on CH removal from artificial IRR cavities or grooves, different samples were used for each experiment.[6,7,9,13,15,16] Campos et al[22] used the same samples for all tested methods. The researchers embedded the teeth they prepared in Eppendorf tubes with silicone and created a mold for repetitive applications. In this study, acrylic blocks with a screw system that can be opened and closed, inspired by the technique presented by Bramante et al[18], were used. In this way, solution leakage due to deformation that may occur in the silicone mold with repetitive applications is prevented. The procedures were performed on the same samples for all groups. With this screw system, which can be considered as a modification of the Muffle system, it is aimed to provide standardization among the experimental groups.

In this study, it was aimed to evaluate the efficiency of different irrigation methods in terms of removing 2 forms of CH from artificial IRR cavities. Results revealed that none of the tested methods was able to remove CH from artificial IRR cavities completely. This finding of the present study is consistent with previous studies.[6,7,9,13,15,16] In addition to that there was no significant difference between 2 forms of CH in terms of removability.

Studies that compare PUI, EDDY, and XP-endo Finisher in terms of removing CH from simulated IRR cavities or grooves are controversial. Marques-da-Silva et al[6] did not detect a significant difference between XP-endo Finisher and EDDY; however, they reported that both groups were significantly superior to PUI. Keskin et al[9] reported that there was no significant difference between XP-endo Finisher and PUI, and both methods gave better results than EDDY. Donnermeyer et al[15] reported that there was no significant difference between EDDY and PUI; however, both methods were superior to XP-endo Finisher. Neskovic et al[23] reported that PUI was significantly superior to agitation with XP-endo Finisher. Campos et al[22] reported that the XP-endo Finisher was significantly superior to PUI.Zhou et al[24], as a result of their meta-analysis in 2021, reported that PUI was more successful in removing intracanal medicaments, especially in the apical third, compared to the XP-endo Finisher. All researchers reported that all of the researched methods gave statistically superior results than manual irrigation. As a result, although there are differences between studies comparing XP-endo Finisher, EDDY, and PUI methods, it is understood that all 3 methods are significantly more successful than manual irrigation.

Göktürk et al[21] reported that although the effectiveness of XP-endo Finisher is not dependent on the solution temperature, it is directly related to the temperature of the environment in which it is used. While the metal composition (nickel-titanium) constituting the structure of XP-endo

Finisher is in the martensite phase at room temperature, it passes into the austenite phase at body temperature, expands, and turns into a spiral structure. It is thought that this phase change makes it easier to contact the hard-to-reach areas in the root canal and increases the agitation efficiency.[15] In the present study, all experiments were done at 20°C, the same as in the study belonging to Donnermeyer et al.[15] Despite that, Marques-da-Silva et al[6] heated the irrigation solutions and used them at 37°C. So, the temperature difference between the used irrigation solutions may be one of the reasons that these studies revealed controversial results. In addition to that, Marques-da-Silva et al.[6] used an ultrasonic tip which has size 30 and taper 0.02, while Donnermeyer et al[15] and the present study used tips that have size 15 and 0.02 taper. According to Marques-da-Silva et al[6], because of its relatively big size, the ultrasonic tip could not vibrate freely in the root canals and touched the walls. As a result of that, ultrasonic energy could not be delivered to the irrigation solutions properly and that reduced the efficiency of PUI.

EDDY has a higher vibration frequency (6000 Hz) than other sonic devices and can oscillate at high amplitudes thanks to its polymer tip.[9,15] This is thought to be one of the reasons why EDDY gave similar results to PUI.

In this study, no significant difference was detected between CH prepared with dw and its injectable form (UltraCal XS) in terms of removability. This finding is consistent with previous studies.[25,26]

In this study, all the methods tested were performed in straight root canals, which is a limitation. It was thought that it would be useful to evaluate the methods in curved root canals in future studies. Also, the simulated internal resorption cavities prepared in this study have smooth surfaces and borders and may not overlap with naturally occurring irregular surfaced cavities. Therefore, removing CH from these cavities may have relatively more successful results. This may be one of the limitations of the study.

None of the tested methods could remove CH completely from artificial IRR cavities. Passive ultrasonic activation revealed the best median scores compared to EDDY and XP-endo Finisher.

Calcium hydroxide that cannot be removed from irregular areas such as IRR cavities may have a negative effect on treatment prognosis. This study, in which additional irrigation methods were investigated to remove CH from these areas, suggested the need for these additional methods and more. Different materials and methods should be considered in future studies, such as using curved root canals and naturally occurring irregular cavities.

The lack of standardization between studies makes it difficult to create standardized methods.

Availability of Data and Materials: The data that support the findings of this study are available on request from the corresponding author.

Ethics Committee Approval: Ethics committee approval was received for this study from the Ethics Committee of İstanbul University, Faculty of Dentistry (Approval no: 297, Date: June 7, 2018).

Informed Consent: Written informed consent was obtained from participants who participated in this study.

Peer-review: Externally peer-reviewed.

Author Contributions: Concept – F.H., O.K.; Design – F.H., O.K.; Supervision – F.H.; Materials – F.H., O.K.; Data Collection and/or Processing – O.K., F.H., G.K.B.; Analysis and/or Interpretation – O.K., F.H., G.K.B.; Literature Search – O.K.; Writing Manuscript – O.K., G.K.B.; Critical Review – F.H.

Declaration of Interests: The authors have no conflict of interest to declare.

Funding: The authors declared that this study has received no financial support.

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