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ORIGINAL ARTICLE / ОРИГИНАЛНИ РАД

Efficiency of calcium hydroxide removal techniques from simulated internal root resorptions – *in vitro* study

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SUMMARY

Introduction/Objective Calcium hydroxide (CH) is the medicament of choice in endodontic treatment of internal root resorptions.
The aim of the study was to compare the effectiveness of three different techniques for CH removal from simulated internal root resorptions.

Methods Twenty-nine extracted single-root teeth were prepared using NiTi rotary files of BioRaCe system (40/.04) following irrigation. A round diamond drill was used in the making of a symmetrical standardized internal resorptions 6 mm from the apex. Three techniques for CH removal from internal resorptions were tested: modified conventional syringe irrigation (CSI), passive ultrasonic irrigation (PUI), XP-endo Finisher (XP). Resorptive cavities and apical thirds were observed under a stereomicroscope (×45) and scored (from 1 to 5), while representative samples were analysed by a scanning electron microscope. Obtained results were statistically processed by Kruskal–Wallis and Mann–Whitney U-test ($p < 0.05$).

Results The most efficient system was PUI, with 66.7% of samples rated 1 and 33.3% rated 2. The next one was XP, and the least efficient was CSI, with 33.3% of samples rated 1 (resorptive defect without medicament). There was a statistically significant difference between the PUI and CSI systems ($p < 0.05$), while there was no difference between the PUI and XP systems.

Conclusion No system completely removed the CH from the simulated internal root resorption cavities. PUI was the most effective system for removing CH. The combination of techniques provides better performance in removing CH paste residues from the canal walls.

Keywords: internal root resorption; irrigation; ultrasound; XP-endo Finisher

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INTRODUCTION

Internal root resorption is a pathological process originating from pulp tissue that, as it spreads peripherally, causes the loss of hard dental tissues [1]. This process was described for the first time in 1830 [2]. It is most often caused by a trauma or an inflammatory process [2, 3]. In most cases, this is an asymptomatic process and is accidentally detected by a radiograph on which it is presented as a sharply limited and symmetrical round radiopacity corresponding to the widening of the root canal. If there is no perforation and communication with the periodontium, endodontic therapy (biopulpectomy) is indicated [4]. The application of a medicament is mandatory, as canal instrumentation and irrigation are not enough to remove granulation tissue and resorptive cells on its periphery [5]. Also, if necrotization and infection of the pulp tissue occur, mechanical instrumentation alone is insufficient for bacteria elimination from irregular spaces of the root canal.

Calcium hydroxide is the medication of choice in endodontic treatment of internal root resorption due to its antibacterial, therapeutic, regenerative and biocompatible properties and has a beneficial effect on mineralization processes [6]. Dissociated hydroxyl ions interfere with the integrity of the bacterial membrane, disrupting the flow of nutrients and destroying phospholipids from unsaturated fatty acids [7].

The CH must be completely removed from the root canal walls prior to the obturation in order not to compromise the penetration of the sealer into the dentinal tubules and to affect the binding and physical properties of the sealer, e.g., eugenol based or mineral trioxide aggregate [7, 8, 9].

One of the most described techniques for calcium hydroxide removal is conventional syringe irrigation (CSI) [10]. Many authors point out the inefficiency of this technique [7, 9, 11, 12]. Due to the perceived shortcomings, like the lack of medication on the walls, numerous other techniques and instruments have been developed to clean root canals more efficiently. Some of these are as follows: passive ultrasonic irrigation, XP-endo Finisher (XP), Canal Brush, Rins Endo system, laser-activated irrigation (PIPS), sonic and ultrasonic irrigation activation, Endo Vac system, Self-Adjusting-Files [4, 5, 9, 13, 14, 15].

Passive ultrasonic irrigation (PUI) is based on the transfer of sound energy to the irrigant. The 25–30 kHz frequency activates the irrigant and creates cavitation bubbles. Sound waves and/or energy-released cavitation increase the penetration of irrigants into irregular spaces [12]. The XP (FKG Dentaire, La-Chaux-de Fonds, Switzerland) is a NiTi nontapered instrument (size 25). At room temperature it is straight (martensite phase), and at body temperature it changes its shape into a unique spoon shape (austenite phase) with a length of 10 mm from the top and a depth of 1.5 mm, due to its molecular memory [15]. It is designed for final cleaning and irrigation, especially for ampoule dilated and irregular canals [9].

The aim of the study was to test the effectiveness of three different techniques for calcium hydroxide removal from simulated internal root resorptions.

The null hypothesis is that there is no statistically significant difference in the efficiency of calcium hydroxide removal from simulated internal resorption between the CSI, PUI, and XP systems.

METHODS

This study used 29 single-rooted, single-canal, extracted teeth. Research was approved by the institutional ethics committee (No. 36/2, 25.02.2020). Round diamond burr with water cooling was used to prepare access cavities and instrument K# 15 was used for checking canal patency. Working length, 1 mm shorter than the apex of the root, was defined. All canals were prepared with BioRaCe (FKG Dentaire) system with apical preparation of 40/.04, with obligatory irrigation of 2% NaOCl after each instrument. Final irrigation consisted of 5 ml of 2% NaOCl and 5 ml of 10% citric acid for a period of 1 minute and 5 ml of saline. Samples were imprinted into a silicone impression material (Elite HD + putti, Zhermack, Badia Polesine, Italy) in an Eppendorf tube (Eppendorf AG, Hamburg, Germany). On the approximate root surfaces longitudinal grooves were made using a diamond disc with water cooling. The roots were cut in half with a chisel and a hammer and a round drill (0.08 mm deep and 0.16 mm in diameter) was used in the making of standardized internal resorptions 6 mm coronary from the root apex. Root halves were reassembled with a dental adhesive (OptiBond Solo Plus, Kerr, USA) and returned to silicone molds, after which all the canals were filled with an aqueous suspension of calcium hydroxide (except negative controls) and closed with temporary filling Citodur hard (DoriDent – Dr. Hirschberg GmbH, Wien, Austria). Samples were incubated in a humid environment at 37°C for seven days. After seven days, the teeth were randomly divided into three groups (n = 9), and a single tooth was used for positive and negative controls. The positive control tooth was filled with calcium hydroxide, which was not removed, and the negative control sample was not filled with medication.

Group I: **CSI (modified)** – medicament was removed from the canal with hand instruments K # 15 to # 40 (master apical file – MAF) with a syringe and an open tip needle irrigation 27G (Sinomedic, China). Total irrigation consisted of 5 ml 2% NaOCl for a period of 1 minute.

Group II: **PUI** – ultrasound apparatus was used (PB-323, W&H Dentalwerk Bürmoos GmbH, Bürmoos, Austria) – ultrasonic endodontic instrument was placed in a canal, 1 mm shorter from the working length, without contact with the canal walls. The irrigation was performed in three series of 20 seconds each. Fresh irrigant was added after each cycle. Total irrigant amount consisted of 5 ml 2% NaOCl for a period of 1 minute.

Group III: **XP** – the instrument is placed in a canal, 1 mm shorter from the working length, using X-smart endo motor (Dentsply Sirona, Ballaigues, Switzerland) with a rotation of 800 rpm and a torque of 1 Ncm. Gentle brush strokes (up and down) were performed with the instrument for 1 minute with constant irrigation, for a total of 5 ml of 2% NaOCl for a period of 1 minute.

Finally, all groups were irrigated with 5 ml of 10% citric acid for 1 minute. Irrigation of all samples was completed with 5 ml of saline.

The teeth were halved again, and internal root resorptions and apical thirds were observed on a stereomicroscope (Boeco BSZ-405, Boeckel + Co (GmbH + Co); Hamburg, Germany) with an integrated digital camera, at 45× magnification. ScopeImage 9.0 computer software (Telescope, Linz, Austria) was used to display the images. Representative samples of each group were analysed by scanning electron microscope (SEM) (Tescan FE-SEM Mira 3 XMU, Tescan a.s., Brno, Czech Republic) operated at 20 keV. The samples were coated with atomic gold layer using a sputter coater (Polaron SC503, Fisons Instruments, Ipswich, UK).

Medication removal efficacy was evaluated according to the methodology of Faria et al. [16] and Topçuoğlu et al. [13] with five grades:

1 = clean internal resorption with only a few drug particles;

2 = several small agglomerates of medication;

3 = multiple medication agglomerates covering less than 50% of the internal resorption area;

4 = more than 50% of internal resorption is covered with medication;

5 = internal resorption is completelly filled with medication.

Evaluation was performed by 3 objective observers. Obtained values were statistically processed in Kruskal–Wallis and Mann–Whitney tests. A value of $p < 0.05$ was considered statistically significant.

RESULTS

Results are shown in Table 1 and Figures 1–3.

Results of this study indicate that after passive ultrasonic irrigation, 66.7% of the samples were graded 1 and 33.3% of the samples were graded 2. After the conventional drug removal technique with MAF with permanent irrigation, 33.3% of the samples were graded 4, and 22.2% of the samples were graded 5; 11.1% of the samples were graded

Table 1. Sample ratings, depending on irrigation technique

Group	n	1	2	3	4	5
Negative c.	1	1	0	0	0	0
Positive c.	1	0	0	0	0	1
PUI	9	6	3	0	0	0
CSI	9	3	1	0	3	2
XP	9	3	2	2	2	0

PUI – passive ultrasonic irrigation; CSI – conventional syringe irrigation;
XP – XP-endo Finisher

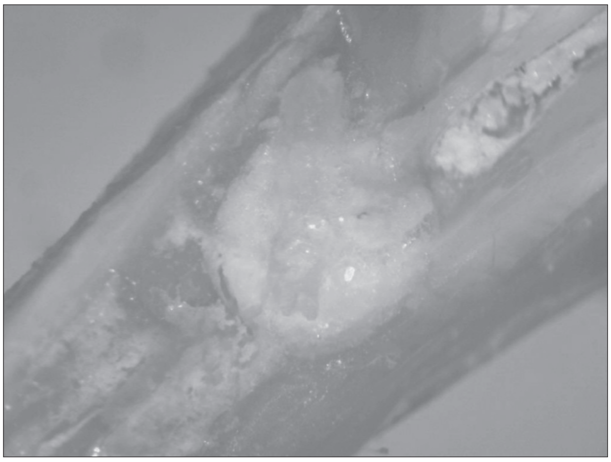

Figure 1a. Stereomicroscope image of filling of internal resorption cavity with $Ca(OH)_2$ after conventional syringe irrigation; grade 5; magnification 45×

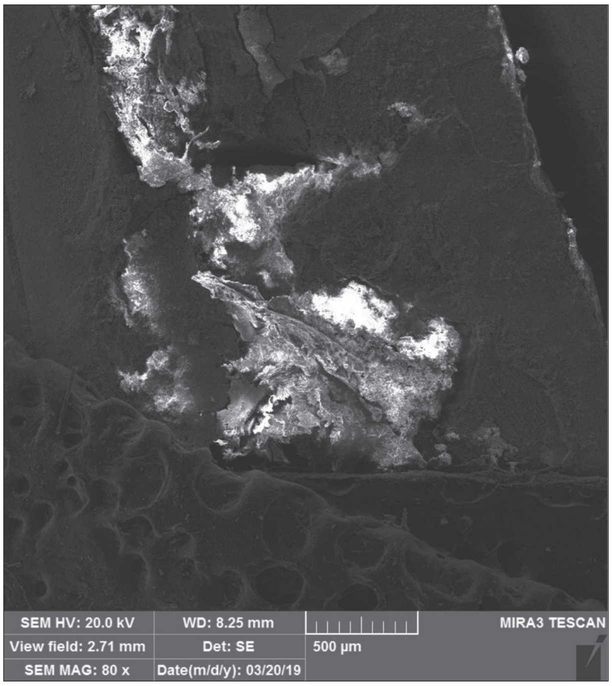

Figure 1b. Scanning electron micrograph of resorptive lacuna completely filled with $Ca(OH)_2$

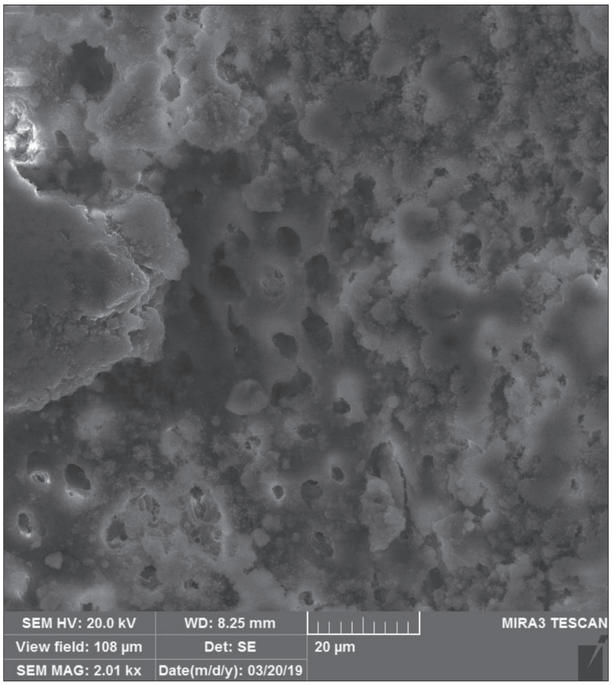

Figure 1c. Scanning electron micrograph of dentin in the apical third mainly covered with $Ca(OH)_2$ crystals

2, and 33.3% of the samples were graded 1. For XP, 33.3% of samples had a grade of 1, while other grades had 22.2% each. A statistically significant difference was observed between the conventional drug removal technique and other irrigation techniques ($p < 0.05$). There was no statistically significant difference between PUI and XP ($p > 0.05$).

DISCUSSION

No technique has completely removed the CH paste from artificial internal root resorption, which is in agreement with the findings of numerous researchers [4, 5, 9, 13]. Also, the null hypothesis was rejected because PUI was the most effective system for removing CH from resorptive cavities as well as apical thirds. PUI and XP removed the medication much more efficiently than CSI, with no statistical difference between them.

Many studies have been published on the topic of CH removal from the root canal system after inter-session medication. Some of them have tested different irrigants [8, 12, 17, 18], while others tested different irrigation systems [9, 10, 11, 13, 14, 19]. All highlight the great problem of completely removing the medication from the walls, especially from the apical third. Some authors point out that 27% of the canal surface remains covered with CH, while our earlier studies indicate that in some cases only 48% of the root canal surface is cleared of CH [11, 20].

Internal root resorption therapy is a real challenge and one of the least successful endodontic procedures. Problems occur in almost every stage of endodontic treatment: during diagnosis, removal of the entire pulp content, profuse bleeding of the granulation tissue, possible root perforation, and preparation and obturation is difficult of the canal due to irregular canal anatomy [9, 21].

The use of calcium hydroxide is essential in the endodontic treatment of internal root resorptions, as it promotes control and elimination of infection. It affects the processes of repair, remineralization and has antibacterial effects on the remaining microorganisms. Numerous studies indicate a failure to completely remove the drug from

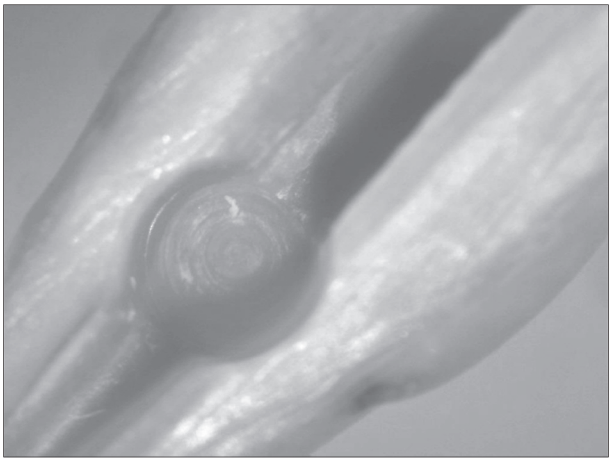

Figure 2a. Stereomicroscope image of an empty internal resorption cavity after passive ultrasonic irrigation; grade 1; magnification 45×

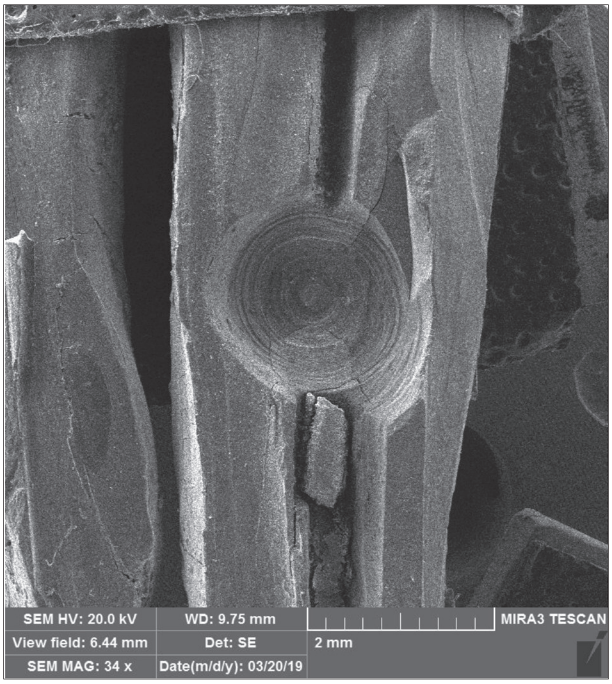

Figure 2b. Scanning electron micrograph of empty resorptive lacuna

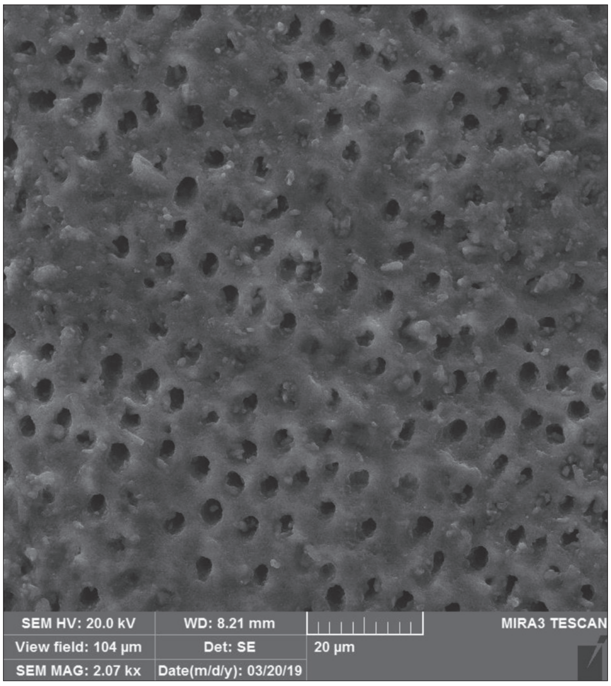

Figure 2c. Scanning electron micrograph of open dentinal tubules in the apical third

the dilated portions of the root canal or resorptive lacunae. [4, 5, 9, 13]. Today, it is known that residual CH interferes with hermetic, three-dimensional obturation and weakens it. The residual CH disturbs the binding mechanisms of different types of sealers and in the long run increases the apical leakage between the canal walls and the sealer [8]. This is especially represented with zinc-oxide eugenol-based sealers, where the newly formed calcium eugenolate alters its physical properties [8, 9]. Residual CH also affects the penetration of epoxy resin-based sealer, more so than calcium silicate-based sealer [8]. Also, there is no agreement as to whether the cleansing effect is affected by the type of vehicle in CH paste (distilled water, propylene glycol, oil etc.) [6, 14, 22]. Chou et al. [11] point out as a problem the viscosity of CH pastes with cellulose carriers with respect to the aqueous suspension, as well as the possibility that partial conversion of calcium hydroxide to calcium carbonate has occurred over time due to the reaction with carbon dioxide.

In this study, a round diamond burr was used to form simulated internal resorptions that were thus round, standardized, with the same amount of medication. In addition to the drill bit, artificial resorptions or grooves in the canal are often prepared with ultrasonic instruments or diamond discs [9, 13, 14, 15, 18]. However, internal resorption has an irregular and usually oval shape, so Da Silveira et al. [23] find it more similar to clinical conditions that the root canal interior is treated with 5% nitric acid in order to create simulated internal resorptions. Given that the efficacy of different methods of calcium hydroxide removal was investigated, artificial internal root resorption of the standardized form could not influence the evaluation of irrigation techniques, although they did not represent the complex physiological anatomy of the root canal. This is confirmed by other authors [4, 9, 18].

Different methods of quantifying calcium hydroxide residues have been described. Digital canal/resorption image analysis is most used, using non-parametric rating systems [5, 9, 18]. However, Phillips et al. [7] point out that there are concerns regarding two-dimensional quantification on uneven surfaces, so they propose chemical microtitration techniques that use high-pH calcium hydroxide. SEM analysis, helical computed tomography (CT) and micro-CT are also commonly used, but the problem of accurate detection of residual CH with respect to root canal tissue has arisen [4, 7]. The scoring system used in this study has been used in many other studies [9, 13, 14, 16, 18]. Most of them used a 3- or 4-grade system. In this study, a 5-rating system was used to describe in more detail the amount of the residual medicament on canal walls/resorption [16].

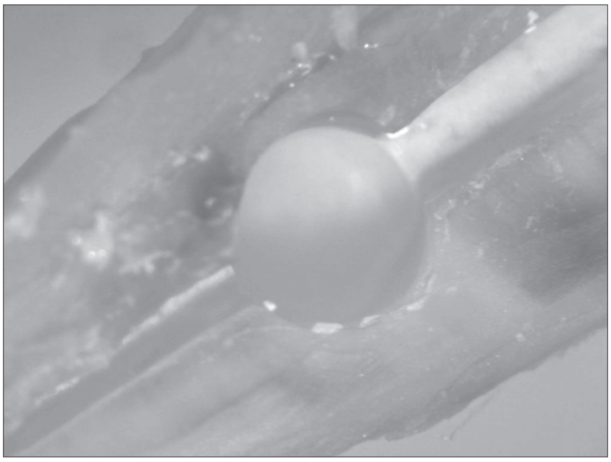

Figure 3a. Stereomicroscope image of residual medication after irrigation with XP-endo Finisher; grade 2; magnification 45×

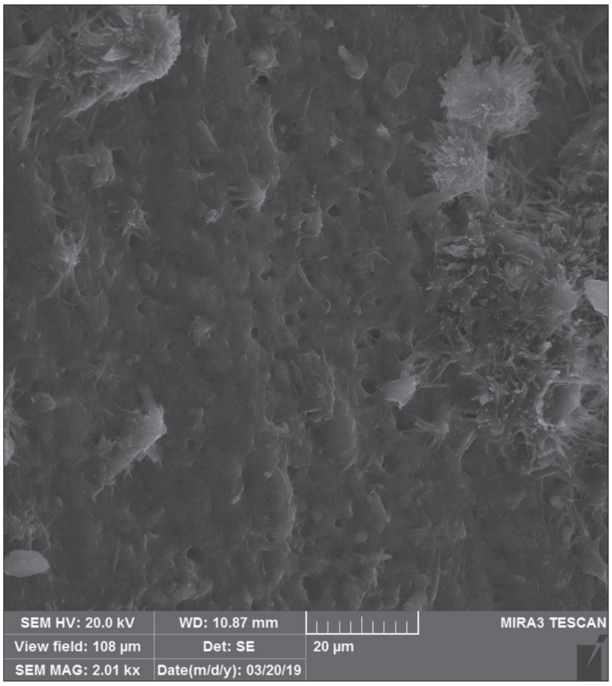

Figure 3b. Scanning electron micrograph of open dentinal tubules in the apical third, in places covered with $Ca(OH)_2$

For this research, teeth with a simple canal system were used to examine the effectiveness of irrigation no matter how complicated and complex the canal system was. The crowns of the teeth were not completely removed to provide room for irrigant storage. Despite all these extenuating circumstances for experimental purposes, no technique completely removed the medicament from the canal walls, which is also in agreement with other researchers [7, 24].

Passive ultrasonic irrigation has shown a high efficiency in removing the CH paste from internal root resorption, which is confirmed by the SEM findings where open dentinal tubules are observed. This is an effective way of removing CH from canal walls, other researchers point out [13, 14, 16, 18, 25]. Its effectiveness is reflected in the penetration of irrigants into all spaces, which is achieved by agitation of irrigants (acoustic waves and cavitation) [26]. With this technique it is important to constantly add new amounts of fresh irrigant (intermittent protocol), because the amount of residual CH is inversely proportional to the volume of used irrigant [24]. In our study, along with NaOCl, citric acid was also used, as it was found that only one irrigant was not sufficiently effective in removing the smear layer and/or calcium hydroxide [13]. This has been confirmed by previous research where PUI with both irrigants was very effective, with an average of 88% of the cleaned root canal walls [20]. Newer research underlines activating chelating irrigants (ultrasonically agitated) with PUI as very efficient [27, 28]. The combination of these two irrigants produces optimal results with the appropriate duration of irrigation, as well as the amount of irrigants themselves [8, 13, 18, 29]. Irrigation lasted 60 seconds per canal (3 × 20 seconds), as agreed by Gokturk et al. [14], although Phillips et al. [7] point out that even 30 seconds per canal is completely adequate time to remove CH.

The XP is presented as a very flexible instrument that has a large amplitude of motion in the A-phase, which is 100 times larger than a “regular” instrument of the same size, so it is recommended for inaccessible canals (FKG Dentaire). Research indicates that it is very effective in removing smear layers and dentin debris [13]. However, it did not fully meet expectations in this research. The results varied. In some samples, the drug was completely removed from internal root resorption, which is confirmed by the SEM findings, and this is in agreement with the finding of Keskin et al. [5], who state that XP is superior to other techniques but not to PUI. Its efficiency is the result of a change in shape with respect to body temperature, thus extending the cleaning effect by up to 6 mm in diameter. However, there are samples (22.2%) where internal resorption is filled with medication, even more than 50% of the resorptive lacuna. This finding is in agreement with the findings of Wigler et al. [9], who obtained similar results, and the efficiency of XP did not meet their expectations either. The aforementioned finding can be explained by the inappropriate amplitude of movement of the instrument, which does not allow the penetration of the instrument into all areas of internal resorption, or the time of 1 minute, recommended by the manufacturer, is insufficient in CH removal. Also, Bao et al. [15] point out that XP is much more efficient when used with intermittent irrigation protocol, thereby cumulatively increasing the effect of NaOCl. Continuous irrigation was used in this study. However, there is no statistically significant difference between XP and PUI systems, as found by other authors [5, 9, 14]. CSI has shown the worst results, which is in line with the findings of numerous researchers [11, 14, 15, 16]. The tip of the irrigation needle is positioned at least 1 mm shorter than the working length, to prevent extrusion of the irrigation via the foramen apical. In this way, large amounts of CH remain on the canal walls in the apical third. Despite the modification of the CSI, by introducing successive files to the MAF, our results clearly showed the limitations of mechanical instruments in irregular canal spaces.

Additional research is needed to test these and other techniques in a complex root canal system.

CONCLUSION

The null hypothesis was not confirmed. No irrigation system completely removed calcium hydroxide from artificial internal root resorption. Passive ultrasound irrigation has shown the highest efficiency in cleaning medication from internal root resorption.

For complete clinical success, multiple medicament removal systems need to be combined.

Conflict of interest: None declared.

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Ефикасност техника уклањања калцијум-хидроксида из симулираних интерних ресорпција корена – *in vitro* студија

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САЖЕТАК

Увод/Циљ Калцијум-хидроксид (КХ) јесте медикамент избора у ендодонтском лечењу интерних ресорпција.

Циљ овог рада је био да се провери ефикасност три различите технике уклањања КХ из симулираних интерних ресорпција корена.

Методе Двадесет девет једнокорених зуба је обрађено применом *NiTi* ротирајућих инструмената *BioRaCe* (40/0,04), уз иригацију. Коренови су потом засечени дијамантским диском на латералним странама и длетом подељени на две половине. Округлим дијамантским сврдлом су симетрично направљене стандардизоване интерне ресорпције на 6 *mm* од апекса. Коренске половине су затим састављене, а лентулом је унет медикамент КХ. Тестиране су три технике за уклањање КХ из интерних ресорпција: модификована конвенционална иригација шприцем (КИШ), пасивна ултразвучна иригација (ПУИ), *XP-endo Finisher* (*XP*). Ресорптивни кавитети и апексне трећине су посматрани под стереомикроскопом (×45) и бодовани системом од 1 до 5, а репрезентативни узорци су анализирани коришћењем *SEM*. Добијени резултати су статистички обрађени Краскал–Волисовим и Ман–Витнијевим *U*-тестом ($p < 0,05$).

Резултати Најефикаснији систем био је ПУИ, са 66,7% узорака оцењених оценом 1, а 33,3% оценом 2. Следећи је био *XP-endo Finisher*, а најмање ефикасан је био КИШ са 33,3% узорака са оценом 1 (ресорптивни дефект без медикамента). Статистички значајна разлика постоји између система ПУИ и КИШ ($p < 0,05$), док не постоји између система ПУИ и *XP*.

Закључак Ниједан систем није у потпуности уклонио медикамент из симулираних интерних ресорпција. Комбинацијом техника обезбеђује се бољи учинак у уклањању заосталог медикамента са зидова канала.

Кључне речи: интерна ресорпција корена; иригација; ултразвук; *XP-endo Finisher*