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ORIGINAL ARTICLE / ОРИГИНАЛНИ РАД

The influence of the final irrigation protocol on the efficiency of root canal cleaning

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SUMMARY

Introduction/Objective Irrigation has an important role in root canal cleaning and its efficiency depends on the type of irrigants, the amount, the technique and the irrigation protocol.
The aim of this work was to estimate the efficiency of cleaning of the canal walls by using scanning electron microscope analysis after the instrumentation by rotary NiTi instruments with the use of three different irrigation solutions and two final irrigation protocols.

Methods Sixty extracted human incisors were divided into two groups after the rotary instrumentation with the iRace instruments. In both groups, the same amount (1.5 ml) of three solutions (2% sodium hypochlorite solution, 2% chlorhexidine solution, and 10% citric acid solution) and total final irrigation time (90 seconds) was the same. The final irrigation in the first group was accomplished using the technique of continuous irrigation and in the second group it was done using the intermittent protocol. The roots were cut longitudinally and analyzed by thirds (coronal, middle, and apical) on a scanning electron microscope (JSM 6460LV, JEOL Ltd., Tokyo, Japan) with 1,000× magnification.

Results The most efficient cleaning of the root canal walls in both groups was seen after the use of citric acid with the intermittent protocol of the final irrigation (90.7% clean walls), while the least efficient was the final irrigation by chlorhexidine with continuous irrigation (80.3%). The most efficient cleaning of the canal walls in both groups was observed in the coronal third and the largest amount of the smear layer in the apical third.

Conclusion The most efficient cleaning of the canal was achieved by the use of citric acid and the intermittent protocol of the final irrigation. In all tested solutions, the intermittent protocol of irrigation was more efficient than continuous irrigation.

Keywords: final irrigation protocol; irrigants; smear layer removal

INTRODUCTION

The success of endodontic treatment significantly depends on the possibility of complete elimination of microorganisms from the root canal, and prevention of reinfection of periapical tissue. Microcomputer tomographic studies have shown that a large part of the surface of the main canal remains untouched by instruments, and in the case of the presence of isthmuses, ramifications and lateral canals, this percentage ranges 30–50% indicating the extreme importance of irrigation in the cleaning and disinfection of the root canal system [1, 2].

Preparation of the root canal manually and particularly by rotating Ni-Ti instruments, leads to the formation of dentine debris and a smear layer, which are most often accumulated in the uninstrumented parts of the root canal system [3]. The smear layer prevents adequate adherence of a sealer to the walls of the root canal and can be a potential area for the growth of numerous bacteria, but also prevent antibacterial agents from reaching the residual bacteria in the dentinal tubules [4, 5]. Mechanical instrumentation eliminates the largest number of bacteria, but maximum reduction of the number of microorganisms organized into biofilms demands an irrigant with good antibacterial effect and adequate irrigation techniques [6, 7, 8].

Irrigation of the canal whose efficiency depends on the type of irrigant, quantity, technique and the protocol of irrigation, is of crucial importance for the efficient cleaning of the complex root canal system [9, 10, 11]. Optimal irrigation today involves the use of two or more solutions and the application of appropriate protocols in order to increase its efficiency [6].

The most commonly used solution for irrigation in endodontics is NaOCl due to its strong antibacterial and exceptional soluble effect, despite the toxicity for periapical tissues [11, 12]. Chlorhexidine is also used because of the extraordinary and prolonged antibacterial effect and the absence of cytotoxicity [12, 13]. Chelating agents, EDTA (tetrasodium ethylenediaminetetraacetic acid), and citric acid effectively dissolve inorganic substances and thus significantly contribute to the removal of the smear layer [14, 15]. The precondition for the success of the endodontic treatment is clean dentinal walls of the root canal without the

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presence of a smear layer and debris to allow best sealing and adhesion of the sealer [16, 17].

Contemporary irrigation also involves different activation protocols in order to improve the efficiency of the irrigant. Studies have confirmed that passive ultrasonic irrigation (PUI) is more effective than the conventional one [15, 18, 19, 20], and De Moor et al. [18] found that PUI in three cycles is equally effective in debris removal as well as laser-activated NaOCl solution. Leoni et al. [20] found that the XP Endo Finisher is as effective as PUI, and they also showed that activated irrigation is significantly more efficient in cleaning the root canal than conventional irrigation.

The objective of this study was to evaluate the efficiency of cleaning root canal walls after instrumentation by rotary Ni-Ti instruments and application of three different irrigation solutions and two final irrigation protocols using scanning electron microscope (SEM) analysis.

The hypothesis of this study was that the final three-step irrigation (intermittent protocol) provides more efficient cleaning of the root canal system than the conventional irrigation protocol.

METHODS

The study was conducted on 60 extracted human incisors, which were stored up to experiments in a 0.01% solution of NaOCl at a temperature of 4°C. The crowns of the teeth were cut off so that each root sample was 15 mm long.

After the formation of the access cavity, the initial penetration of the root canal was established by K-file #10. The working length was determined to be 1 mm shorter than the apical foramen, i.e. 14 mm. At the top of each root, a pink wax ball was placed in order to prevent the irrigation solution leaking during the instrumentation. The instrumentation of all canals was carried out by one researcher. After adjusting the working length by a hand instrument and before starting the instrumentation, the canal was irrigated with 2 ml of 1% solution of NaOCl.

Mechanical preparation of all canals was performed by NiTi rotating instruments iRace (FKG Dentaire SA, La Chaux-de-Fonds, Switzerland) using three instruments: R1 #15/06, R2 #25/04, and R3 #30/04. After each instrument, the canals were irrigated with 2 ml of 1% NaOCl solution with 2 ml plastic syringes and gauge 27 needles. After each use of an instrument, irrigation was carried out in the manner described so that the total amount of the irrigant used during preparation for each sample was 8 ml of 1% NaOCl solution.

After the instrumentation of the canals, samples were randomly selected in two groups of 30 teeth, where the final irrigation was carried out in group 1 by a continuous protocol, while in group 2 an intermittent final irrigation protocol was used. In both groups, three solutions were used in the same amount (1.5 ml each) and total final irrigation time (90 seconds): 2% solution of sodium hypochlorite (Chloraxid, 2%, Cerkamed, Stalowa Wola, Poland); 2% solution of chlorhexidine (Glucohex, 2%, Cerkamed), and 10% citric acid solution was obtained by diluting 40% citric acid solution (citric acid, 40%, Cerkamed).

Group 1 – in the first group, the final irrigation was performed by the continuous flushing protocol in the amount of 1.5 ml of irrigant for the duration of 90 seconds. Ten teeth were irrigated with 1.5 ml of 2% solution of sodium hypochlorite. The amount of 1.5 ml of 2% chlorhexidine solution was used for each of the following 10 teeth, and the last 10 teeth from this group were irrigated with 1.5 ml of 10% citric acid solution.

Group 2 – in the second group, the final irrigation was carried out according to an intermittent flushing protocol of 3 × 0.5 ml of irrigant for a period of 3 × 30 seconds. Each subgroup of 10 teeth was irrigated with following solutions: 3 × 0.5 ml 2% solution of sodium hypochlorite for 3 × 30 sec, 3 × 0.5 ml 2% chlorhexidine solution for 3 × 30 sec, and 3 × 0.5 ml of 10% citric acid solution for 3 × 30 sec.

The roots were longitudinally cut with a diamond disc (so that the root canal remains intact) separated with sharp spatula into two halves. The halves obtained in this way were prepared for SEM analysis (JSM 6460LV, JEOL, Tokyo, Japan). A total of 120 samples were dried and filled with gold and scanned by an electron microscope. For each sample, five standardized microphotographs were made for coronal, middle, and apical thirds at magnification of 1,000×. SEM microphotographs were independently analyzed and appraised by two researchers. In the event of disagreement, the ratings were reconsidered until a consensus was reached.

The criteria set by Hülsmann et al. [21] were used to qualitatively estimate the residual smear layer, according to the cleaning efficiency:

Score 1 – the root canal wall is without a smear layer, all dentinal tubules are open;

Score 2 – a small quantity of a residual smear layer and most of the dentinal tubules are open;

Score 3 – a homogeneous smear layer covers the walls, a few dentinal tubules open;

Score 4 – the entire wall of the root canal is covered with a smear layer, there are no open tubules;

Score 5 – a non-homogeneous smear layer covers the entire surface of the root canal.

The scoring implies that grades 1 and 2 represent a clear root canal wall, and the wall with a smear layer includes grades 3, 4, and 5.

The obtained result was statistically processed in IBM SPSS Statistics, Version 20.0 (IBM Corp., Armonk, NY, USA) using the descriptive statistics method and the χ^2 test.

The study was approved by the Ethics Commission of the School of Dental Medicine, University of Belgrade (36/6).

RESULTS

The results of the SEM analysis are shown in Tables 1 and 2 and Figures 1–4.

In the group with a continuous final irrigation protocol when NaOCl was used as the irrigant, the lowest average value of the assessment of the smear layer presence was observed in the coronal third (1.6), then in the middle (1.7), while the weakest cleaning was recorded in the apical

Table 1. Mean value of the assessment of the residual smear layer on root canal walls by thirds

	Groups	Solution for irrigation	Third of root canal	Smear layer rating					
				n	χ	SD	Med	Min.	Max.
Final irrigation	Group 1 continuous protocol	NaOCl	coronal	100	1.60	0.67	1.5	1	3
			middle	100	1.70	0.73	2	1	4
			apical	100	2.14	1.16	2	1	5
		Chlorhexidine	coronal	100	1.62	0.66	2	1	3
			middle	100	1.76	0.79	2	1	4
			apical	100	2.26	1.21	2	1	5
		Citric acid	coronal	100	1.5	0.58	1	1	3
			middle	100	1.64	0.69	2	1	3
			apical	100	2.04	1.08	2	1	5
	Group 2 intermittent protocol	NaOCl	coronal	100	1.54	0.61	1	1	3
			middle	100	1.66	0.62	2	1	3
			apical	100	2.06	0.99	2	1	4
		Chlorhexidine	coronal	100	1.62	0.66	2	1	3
			middle	100	1.66	0.65	2	1	3
			apical	100	2.11	1.03	2	1	4
		Citric acid	coronal	100	1.52	0.61	1	1	3
			middle	100	1.52	0.61	1	1	3
			apical	100	1.76	0.71	2	1	3

n – number of teeth; χ – mean value; SD – standard deviation

Table 2. Assessment of the cleaning efficiency of root canal walls regarding the final irrigation solution and applied irrigation protocol

Final irrigation protocol				Continuous irrigation		Intermittent irrigation	
Assessment of the presence of the smear layer				Clean walls Score 1, 2	Smear layer present Score 3, 4, and 5	Clean walls Score 1, 2	Smear layer present Score 3, 4, and 5
NaOCl	SEM analysis	Coronal third	n	90	10	94	6
			%	90	10	94	6
		Middle third	n	88	12	92	8
			%	88	12	92	8
		Apical third	n	71	29	72	28
			%	71	29	72	28
			n	249	51	258	42
			%	83%	17%	86%	14%
Chlorhexidine	SEM analysis	Coronal third	n	90	10	90	10
			%	90	10	90	10
		Middle third	n	86	14	90	10
			%	86	14	90	10
		Apical third	n	65	35	69	31
			%	65	35	69	31
			n	241	59	249	51
			%	80.3%	19.7%	83%	17%
Citric acid	SEM analysis	Coronal third	n	96	4	94	6
			%	96	4	94	6
		Middle third	n	88	12	94	6
			%	88	12	94	6
		Apical third	n	72	28	84	16
			%	72	28	84	16
			n	256	44	272	28
			%	85.3%	14.7%	90.7%	9.3%

SEM – scanning electron microscope

third (2.14) (Table 1). Slightly higher mean values of the evaluation of the smear layer was observed after the application of chlorhexidine, mostly in the apical (2.26), then in the middle (1.76), and the coronal third (1.62). The most effective cleansing was observed in the group with citric acid [in the coronal third (1.5), in the middle one (1.64), and the least effective cleaning was noted in the apical third (2.04)] (Table 1).

In the group with an intermittent protocol of final irrigation, the mean values of the presence of the residual smear layer were slightly lower in regard to the first group. After using NaOCl, the lowest mean was in the coronal third

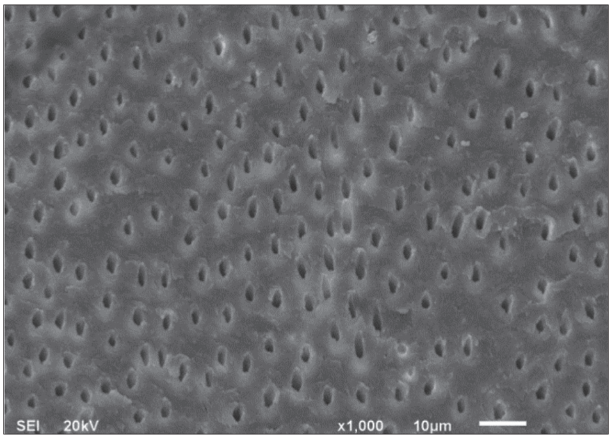

Figure 1. Representative microphotography of the coronal third (citric acid, intermittent protocol) (score 1) – scanning electron microscope, magnification ×1,000

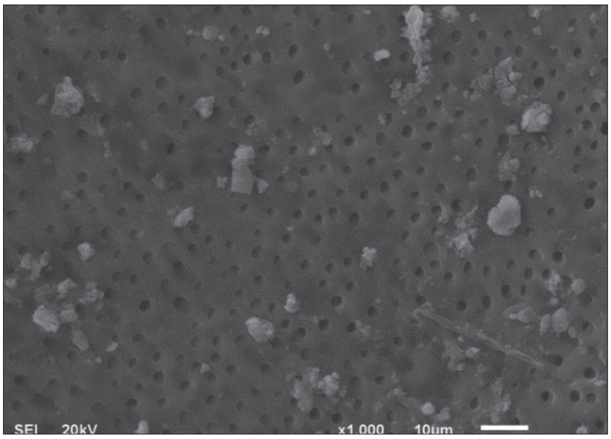

Figure 2. Representative microphotography of the middle third (NaOCl, intermittent protocol) (score 2) – scanning electron microscope, magnification ×1,000

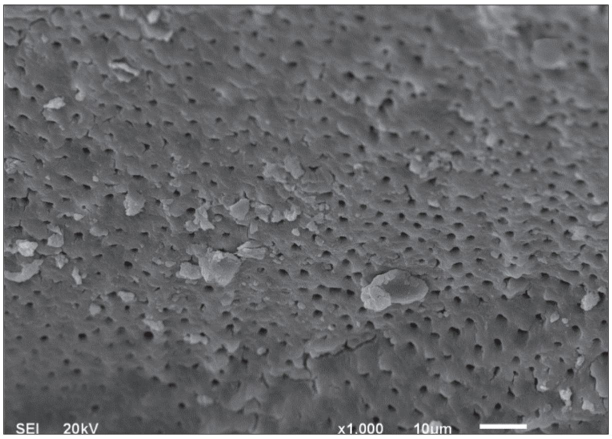

Figure 3. Representative microphotography of the apical third (citric acid, intermittent protocol) (score 2) – scanning electron microscope, magnification ×1,000

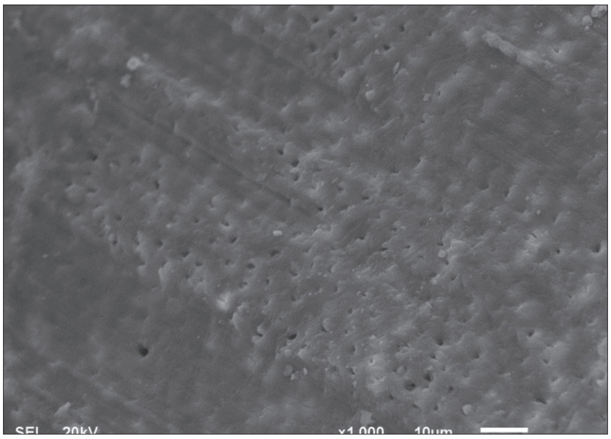

Figure 4. Representative microphotography of the apical third (chlorhexidine, intermittent protocol) (score 3) – scanning electron microscope, magnification ×1,000

(1.54), followed by the middle one (1.66), and the highest value was in the apical third (2.06). When chlorhexidine was used as a final irrigant, the highest mean value was observed in the apical (2.11), slightly lower value was in the middle (1.66), and the lowest mean value was on the walls of the coronal third (1.62). The smallest amount of the residual smear layer was observed in the group with citric acid, the same average value was in the coronal and the middle third (1.52), and the weakest cleaning was in the apical third (1.64) (Table 1).

The analysis of the cleaning efficiency of root canal walls showed that each irrigant was more efficient with the protocol of intermittent final irrigation, with no statistically significant difference. After using NaOCl, 83% of clean walls in group 1 were detected, while 86% of clean walls were recorded in the second group. Less efficient cleaning was observed after the application of chlorhexidine, 80.3% of clean walls with continuous protocol, 83% of clean walls with the intermittent protocol. The most effective cleaning was observed after the final irrigation with citric acid using the intermittent irrigation protocol (90.7%), and slightly weaker in the group with continuous irrigation (85.3%) (Table 2).

The most effective cleaning of the root canal walls in both groups was observed after the application of citric acid with the intermittent final irrigation protocol (90.7% clean walls), while the final irrigation with chlorhexidine with continuous irrigation (80.3%) was the least effective.

The most efficient cleaning of root canal walls in the first group was observed in the coronal third (92%), followed by the middle third (87.3%), while on the walls of the apical third there was the largest amount of residual smear layer (69.3%) (Table 2).

By analyzing the effectiveness of wall cleaning in the second group, the largest amount of smear layer was observed on the walls of the apical third of the root (75% clean walls) (Figures 3 and 4), followed by the middle third (92%), while most of the dentinal tubules were open in the coronal thirds (92.7%) (Table 2).

DISCUSSION

Root canal instrumentation produces a smear layer on all instrumented surfaces of the root canal walls, while the uninstrumented areas of the canal system (isthmuses, lateral canals, anastomoses between the canals, etc.) are usually occluded by debris. Although there are studies showing that the presence of the smear layer does not affect the outcome of endodontic treatment [16], most studies confirm that its presence prevents penetration of intracanal medications into the dentinal tubules and reduces the adhesion, so it is necessary to remove it before definitive obturation [1, 4, 17].

Earlier investigations used light microscopy to identify the smear layer on the canal walls, but today SEM analysis is the standard in the field of quantitative and qualitative estimation of the presence of the smear layer due to high resolution and high magnification [22–25].

One of the tasks of irrigation is to clean dentinal walls by removing the smear layer and debris and to reduce the number of microorganisms, i.e. to improve the adhesion of the sealer and thus minimize microleakage [6, 17]. The efficiency of irrigation depends on a number of factors, and above all on the type, quantity, concentration, time of exposure of the walls to the effect of irrigant and irrigation techniques [3, 7, 11, 12, 20, 22].

The complete instrumentation of the canal in this study was performed by one operator, on simple single root teeth, and all canals were instrumented in the same way with the same quantity of irrigant and the same total duration of irrigation, but with two different final irrigation protocols (continuous and intermittent irrigation) with three different irrigants.

The results of this study show that the mechanical instrumentation with rotating Ni-Ti files followed by extensive irrigation ensures efficient cleaning of the canal walls with a small amount of smear layer present on the walls.

Since no statistical significance was found, the hypothesis of this study is rejected, yet slightly better cleaning of the canal walls in all three thirds was observed after the intermittent final irrigation protocol in three steps in comparison with the convectional continuous irrigation. This is in accordance with the findings of other authors who have showed that increasing the number of irrigation cycles increases the cleaning capacity as the amount of fresh solution is restored, while in the case of continuous irrigation, the saturation of the solution occurs faster [7, 11, 25, 26,]. Živković et al. [25] have determined that the protocol of the final irrigation in three cycles improves the efficacy of removing the smear layer in the apex segment of the root canal, and Macedo et al. [26] showed that the irrigation protocol in three cycles of fresh NaOCl solution increases its cumulative effect and thus the efficacy of cleaning root canal walls.

Such good results can be explained by the fact that instrumented canals were straight and simple, and adequate diameters of apical preparation (30/04) ensures that the tip of the irrigation needle will reach almost the working length of the instrumentation and in this way effectively clean the walls of the root canal. It also explains very good results for chlorhexidine, which, unlike NaOCl and citric acid, does not have the ability to dissolve tissues, but it is used because of a wide antibacterial spectrum (including *Enterococcus faecalis*) and prolonged antimicrobial effect [8, 12, 13, 23].

Citric acid showed the best cleaning effects (in both groups). This chelating agent is equally effective in removing the smear layer as well as the EDTA according to the findings of Lenarda et al. [14]. This mineralolithic perfectly dissolves inorganic material and significantly affects the removal of the smear layer from root canal, although it does not have antibacterial properties [9, 15].

The worst cleaning of dentinal walls in both groups is observed in the apical third of the root canal, then in the middle, while the smallest amount of the smear layer is noticed in the coronal third of both groups, which is in compliance with the results of other studies confirming that the smear layer from the canal walls is more easily removed from the coronal and middle third [9, 10, 21, 26]. The cleansing problem is particularly emphasized in the region of the apical third due to anatomical specificity (isthmuses, ramification, additional canals), and due to the small diameter of the apical preparation, which makes the debridement of the canal more difficult [3, 5, 7, 25].

So far, research has shown that none of the irrigation protocols or tested solutions are able to completely clean root canal walls by removing the smear layer, and nowadays some kind of activation of the irrigation solution during the irrigation process is recommended [6].

Currently, passive ultrasonic irrigation (PUI) has an important role in the activation of irrigants, and its activity is based on cavitation and acoustic streaming of solutions during irrigation. Numerous studies have shown that PUI increases the effect of irrigation by removing more organic tissue, planktonic forms of bacteria, and debris from canal walls. [15, 18, 19, 20, 26].

Laser-activated irrigation is also very effective, but De Moor et al. [18] have found that PUI in three cycles is equally effective in the removal of debris as well as the laser-activated NaOCl solution.

Research has shown that XP-endo Finisher, which is used for the final debridement of the root canal, due to its specific design and extreme flexibility (it changes shape during instrumentation), can reach the inaccessible parts of the canal system [7, 20, 24].

Kato et al. [27] examined Easy Clean (Easy Dental Equipment, Belo Horizonte, MG, Brazil), new mechanical irrigant agitating device, powered by the reciprocating or continuous rotation, and indicated that Easy Clean in reciprocating motion is more efficient in cleaning the apical third of the curved canals compared to the PUI. Duque et al. [28] compared the effectiveness of Easy Clean in continuous and reciprocating motion, PUI, Endoactivator systems (Dentsply Maillefer, Ballaigues, Switzerland), and convectional irrigation for debris removal from the root canal and isthmus, and found that Easy Clean used in continuous rotation provides better cleaning of the canal and isthmus. They also concluded that protocol of three irrigating solution activations for 20 seconds ensures better cleaning.

CONCLUSION

The most efficient solution for final irrigation after root canal preparation with rotary iRaCe instruments, in this study was 10% citric acid, while the least effective one was chlorhexidine.

Under the conditions and limitations of this research, it can be concluded that root canal instrumentation by rotary instruments followed by the final irrigation was efficient in smear layer removal from root canal walls. An intermittent irrigation protocol in three steps showed slightly more efficient cleaning of root canal walls compared to continuous irrigation.

Conflict of interest: None declared.

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Утицај протокола финалне иригације на ефикасност чишћења канала корена

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САЖЕТАК
Увод/Циљ Иригација има важну улогу у чишћењу канала корена, а њена ефикасност зависи од врсте ириганса, количине, односно технике и протокола иригације.
Циљ овог рада био је да се анализом СЕМ процени ефикасност чишћења зидова канала после инструментације ротирајућим *NiTi* инструментима уз примену три различита раствора за иригацију и два протокола финалне иригације.
Методе Шездесет екстрахованих хуманих секутића је после машинске инструментације *iRaCe* инструментима подељено у две групе. У обе групе су коришћена по три раствора – 2% раствор натријум-хипохлорита, 2% раствор хлорхексидина и 10% раствор лимунске киселине, у истој количини (1,5 *ml*) и укупном времену финалне иригације (90 секунди). Финална иригација у првој групи је реализована техником континуиране, а у другој техником интермитентне иригације. Коренови су пресечени уздужно и анализирани по трећинама (крунична, средња и апикална) на скенирајућем електронском микроскопу (*JSM* 6460*LV JEOL*, Токио, Јапан) на увеличању од 1000×.
Резултати Најефикасније чишћење зидова канала корена у обе групе уочено је после примене лимунске киселине уз интермитентни протокол финалне иригације (90,7% чистих зидова), док је најмање ефикасна била финална иригација хлорхексидином уз континуирану иригацију (80,3%). Најефикасније чишћење зидова канала и у првој и у другој групи уочено је у круничној трећини, а највише размазног слоја у апикалној трећини.
Закључак Најефикасније чишћење канала остварено је применом лимунске киселине и интермитентног протокола финалне иригације. Код свих тестираних раствора интермитентни протокол иригације је био нешто ефикаснији од протокола континуиране иригације.

Кључне речи: протокол иригације; финална иригација; размазни слој