The Effectiveness of Endodontic Irrigating Solutions on Smear Layer Removal from Radicular Dentin

A scanning electron microscopic study

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The aim of our scanning electron microscopic study was to evaluate the cleaning efficiency of irrigating solutions used in endodontic treatment regarding smear layer removal from the root canal dentin walls. Ethylenediaminotetraacetic acid (EDTA)17%, citric acid(CA)10% and chlorohexidinegluconate (CHX)2% solutions were tested as final irrigants after endodontic treatment. The study was conducted on extracted teeth, divided in four groups according to the irrigation protocol used. The specimens were analyzed by scanning electron microscopy and the amount of smear layer present at apical, middle and coronal level was recorded, based on a scoring system. Data were statistically analyzed using Kruskal-Wallis and Friedman test and the level of significance was set at $p<0.05$. At the coronal and middle thirds we recorded no statistically significant difference between EDTA and CA regarding the smear layer removing capacity. Final irrigation with 10% CA proved to be more efficient than 17% EDTA in smear layer removal at apical level of the root canal, with $p<0.05$ ($p=0.036$), which is an important area for disinfection in endodontic treatment.

Keywords:chelating agents, smear layer, scanning electron microscope, radicular dentin

The long term success of endodontic treatment is based on proper instrumentation, disinfection and three-dimensional filling of the entire root canal system. During this process, a special attention is given to complete removal of organic and inorganic debris formed on the radicular dentin surface as a result of endodontic instrumentation, known as smear layer [1,2]. Most of the irrigating protocols proposed contain sodium hypochlorite (NaOCl) associated with a chelating agent, but the best sequence of solutions is still a reason of debate and controversy in the scientific literature [3].The combination of NaOCl (table 1) with a chelating agent as EDTA or CA has been proposed as an excellent irrigation method, which effectively removes debris and smear-layer [4].

EDTA is an aminopolycarboxylic acid and a colourless water soluble solid, which has the ability to capture metal ions such as Ca^{2+} by binding to its two amines and four carboxylates (fig. 1, table 2). It is an artificial aminoacid with a *p*H 7, biocompatible and with a reduced antibacterial effect, based on the inhibiting effect on bacterial growth by chelating with metallic ions used in their metabolisms [5,6].

At concentrations of 15-17% eliminates calcium from the dentin, leaving an organic matrix. EDTA solutions with the addition of a surfactant loosen up calcifications at the top or inside the root canal, allowing the complete instrumentation of obliterated canals.Citric acidis a week organic tribasic acid that has been suggested as a chelating

Table1

CHARACTERISTICS OF SODIUM HYPOCHLORITE

Chemical formula	NaOCl
Molar Mass	74.44 g/mol
Appearance	Greenish-yellow solid (pentahydrate)
Density	1.11 g/cm^3

Table2

CHARACTERISTICS OF EDTA

Chemical formula	$C_{10}H_{16}N_2O_3$
Molar Mass	292.24 g/mol
Appearance	Colourless crystals
Density	860 g/mL (at 20^{0}C)

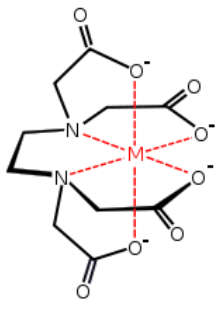

Fig.1. Chemical structure of EDTA

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Chemical formula	$C_6H_8O_7$
Molar Mass	192.124 g/mol
Appearance	Crystalline white solid
Density	1.66 g/cm^3

Table3
CHARACTERISTICS OF CITRIC ACID

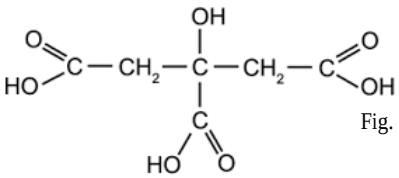

Fig. 2 Chemical structure of citric acid

agent with good capacity of smear layer remova (fig.2, table 3) [6].

It was used at concentrations from 1-50%, but the most common is 10% solution in contact with radicular dentin for 2-3 minutes. Another widely used irrigating solution is CHX2%; it belongs to the polybisguanide family, consisting of two symmetric 4-chlorophenyl rings and two bisguanide groups connected by a central hexamethylene chain. It is a strong base and is most stable in the forms of salts [7].

The aim of this SEM in vitro study was to evaluate the cleaning efficiency of EDTA and CA as final irrigating solutions in smear layer removal from root canal dentin walls during different endodontic treatment protocols.

Experimental part

Materials and methods

Ethical approval was taken before starting this study in which we used forty human freshly extracted teeth. The selection was based on the relative dimension, similar morphology and absence of root curvatures, in order to standardize the endodontic preparation. All teeth were stored in saline solution for 1-2 days and instrumented according to the step-back technique using K-files (Dentsply-Maillefer, Ballaigues, Switzerland) up to # 40, with NaOCl 5.25% irrigation after each file, followed by 5 mL of saline solution. The teeth were divided in 4 study groups according to the final irrigating protocol used (Group I – NaOCl 5.25% + saline solution + EDTA 17%, Group II - NaOCl 5.25% + saline solution + citric acid 10%, Group III - NaOCl 5.25% + saline solution + CHX 2% and Group IV –control, irrigation with saline solution). In each group the samples were irrigated for 1 min with 5 mL of each solution which were freshly prepared and standardized. For the SEM evaluation, longitudinal grooves were made on the buccaland oral surfaces of the roots using a diamond disk at low speed, without penetrating the canal. The roots were split in half with a sharp blade and were coded according to the protocol used. The specimens were dehydrated with increasing concentrations of ethyl alcohol, mounted on coded stubs and sputter-coated with 300 Angstrom gold layer. The specimens were examined using a SEM (Cam scan MV 2300, Oxford Instrument, UK)at x1000 and x750 magnification at the coronal, middle and apical thirds, based on a graded scale from 1-3(1 – complete smear layer removal, open dentin tubules, 2 –moderate smear layer, partially opened tubules, 3- dentin surface completely covered with smear layer) in order to assess the quality of smear layer removal. Photomicrographs of the examined areas were taken and evaluated by two independent observers in a double-blind manner. Statistical analysis was carried out with the Statistical Package for Social Sciences (SPSS)version 16.0 for Windows. Non parametric data of smear layer scores were presented as a percentage distribution and the mean ranks were calculated for each root section. The Kruskal-Wallis test was used to compare between final irrigation solutions at each section between study groups and Friedman test was used to compare between root canal thirds at each group. The level of significance was set at $p<0.05$.

Results and discussions

According to Kruskal –Wallis test there were no statistically significant differences between EDTA and CA except for the apical thirds of the root canal, where CA proved to be more efficient in smear layer removing ability with $p<0.05$ ($p=0.037$).The photomicrographs obtained after SEM evaluation are presented in figure 3 and 4. The mean scores of smear layer removal for the control and study groups are listed in table 4 that shows the ability of CA and EDTA to remove debris efficiently in the coronal and middle thirds of the root canal.

In the apical part, CA proved to have better cleaning properties compared to EDTA and CHX. The Friedman test had shown statistically significant differences within EDTA and CA groups regarding the degree of smear layer removal from canal sections, with a $p<0.05$($p=0.008$ and $p=0.006$ respectively). We recorded also a significant erosion of

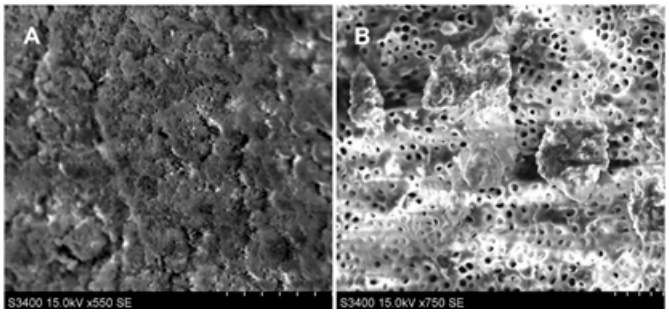

Fig. 3.A. Photomicrographs from the control group irrigated with saline solution, score 3. B. Specimen from groupIII final irrigation with CHX, score2

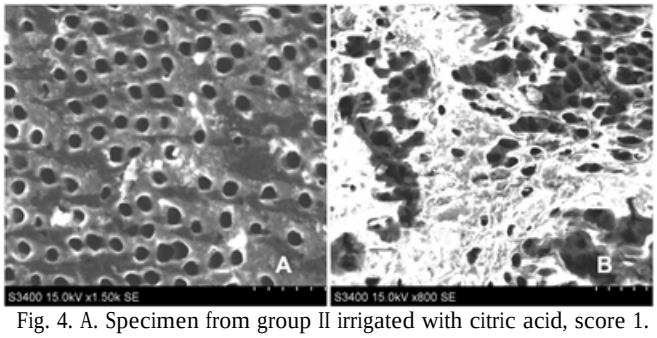

Fig. 4. A. Specimen from group II irrigated with citric acid, score 1. B. Photomicrograph from group I irrigated with EDTA, score 1 and evidence of dentine dissolution

Study group/ Root area	Coronal third Mean (+/-SD)	Middle third Mean (+/-SD)	Apical third Mean (+/-SD)	p-value
Control (saline)	3.0 +/- 0.0	3.0 +/- 0.0	3.0 +/- 0.0	P=1 (p>0.05)
EDTA	1.2 +/- 0.72	1.3 +/- 0.58	2.5 +/- 0.43	P=0.008 (p<0.01)
CA	1.1 +/- 0.81	1.2 +/- 0.78	1.75 +/- 0.20	P=0.006 (p<0.01)
Chlorhexidine	3.0 +/- 0.0	3.0 +/- 0.0	3.0 +/-0.0	P=1 (p>0.05)

Table 4
MEAN SCORES OF SMEAR LAYER REMOVAL FROM THE RADICULAR DENTIN WALL

dentinal tubules in samples irrigated with EDTA, disclosing an important decalcifying effect of this solution.

Root canal instrumentation produces a layer of organic and inorganic material called smear layer that may contain bacteria and their by-products. In endodontic treatment it has many disadvantages as it prevents the penetration of endodontic medicaments into dentinal tubules and disrupts the seal between the dentin walls and root filling, conditions that may lead to treatment failure [2, 7].Cleaning of endodontic system is extremely important and NaOCl 5.25% solution is considered to be the gold standard in endodontic irrigating protocols due to its very good tissue dissolving effect; at the same time, it has been suggested to degrade micromechanical characteristics of dentin. Furthermore, it is toxic to apical tissue and has no effect on the inorganic component of the smear- layer. NaOCl 5.25%solution promotes the formation of smear layer during instrumentation and the use of a chelating agent facilitates the smear layer removal [8]. Another important endodontic irrigant is CHX, a cationic polybisguanide which can be used due to the antimicrobial activity and its unique property called substantivity. Until now, using chelating agents was done mostly by irrigation with EDTA, which has been tested in different concentrations and for different time periods [9-11]. The disadvantages noticed were lack efficiency in the apical area and dentinal erosion in the middle and coronal part of the root. Previous studies had shown that there is no significant difference between CA and EDTA regarding the capacity of smear layer removal, but both disclosed a limited antibacterial effect [12-14]. Spano et al [15] examined smear layer removal with different solutions and found that EDTA and CA had comparable effectiveness. Our results showed that 5.25% NaOCl and 2% CHX did not promote an adequate cleaning of radicular dentin, with a great amount of smear layer present. Previous studies had demonstrated that CHX 2% solution could be an effective endodontic irrigant; it could maintain the canal free of microorganisms but few studied had shown its cleaning capacity. The present study was carried out on extracted teeth and therefore the results do not allow a definite conclusion regarding the effects of chelating solutions used in situ. The presence of blood and tissue fragments, together with other variables may influence the action of these chemical agents under use in the root canal system. Therefore, more long term clinical studies are necessary in order to confirm these results and to evaluate their relevance to dental endodontic practice.

Conclusions

The apical part of root canal imposes a special attention during irrigation as the borderline between safety and effectiveness is particularly important in this area. Final irrigation of the root canal with 10% CA is more efficient than 17% EDTA in smear layer removal at apical level, which represents the most important area for disinfection. The chelating agents used, especially EDTA, exhibited an important decalcifying effect, therefore the risk of dentin erosion should be taken into consideration.

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