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Effectiveness of Continuous Irrigating Solution on the Microhardness of Root Dentin

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Abstract

Aim of the study: This *in vitro* study compared the effects of different irrigation solutions on the root canal dentin's microhardness, including 17% ethylenediaminetetraacetic acid (EDTA), 0.5% chitosan nanoparticles (CNP), and Triton.

Material and method: Forty mandibular premolars that had been extracted were chosen, and root length of 14 mm was determined by decorating the samples. The X4 (40/0.06) rotary file for ProTaper Next was used to instrument each sample. After instrumentation, based on an irrigation solution, the samples were randomly split into four groups (n = 10). Groups I (Triton), II (0.5% CNP), III (17% EDTA), and IV (distal water) are the samples. The samples were rinsed by Triton throughout instrumentation with Triton and final irrigation for 1.5 min by 1 ml of Triton, CNP, EDTA, and distal water were rinsed with final irrigation by 5 ml of 0.5% CNP, 17% EDTA, and distal water for 3 minutes, respectively. Following the longitudinal sectioning of the samples, a Vickers microhardness test was performed on them in the apical, middle, and coronal thirds. The mean of these three-thirds was used to determine the microhardness of each sample. ANOVA test was used for statistical evaluation.

Results: There was a significant difference ($p \leq 0.05$) between all evaluated irrigating solutions, Triton demonstrated the lowest mean of reducing the microhardness compared to the other irrigation solutions followed by CNP and EDTA which had the highest mean of decreasing the root dentin's microhardness.

Conclusions: Comparing this study's irrigation solution to the others, Triton had less of an impact on microhardness. EDTA demonstrated the highest effect on root dentin microhardness in contrast to alternative irrigation methods.

Keywords: irrigation solution, microhardness, Triton, ethylenediaminetetraacetic acid, chitosan nanoparticles

Introduction

To achieve complete disinfection, root canal preparation involves the use of irrigation solutions and endodontic equipment. An amorphous granular smear layer comprising both organic and inorganic materials is produced by the canal's mechanical instrumentation, covering the canal's wall and blocking the dentinal tubule apertures (Silva et al., 2013). Areas that have not been thoroughly cleaned may contain germs and debris that can lead to chronic illnesses and eventually lead to root canal therapy's failure. Moreover, successful endodontic therapy entails the removal of the entire smear layer of the root dentin. Smear layer thickness is based on several variables, such as the use of an irrigation solution during instrumentation,

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root canal dimension and structure, instrument configuration, and type and amount of irrigation solution (Violich & Chandler, 2010). Consequently, irrigation is considered a crucial component in shaping and cleaning root canals, as it aids in removing dentin debris and necrotic tissue from both mechanically and manually prepared areas.

The most common used irrigation solution for root canal therapy is sodium hypochlorite (NaOCl) at a concentration of 2.25–5.25%. However, when used alone, its effectiveness is restricted to removing the organic component of the smear layer. Therefore, as a final irrigant to remove the inorganic smear layer component, chelating agents like ethylenediaminetetraacetic acid (EDTA) are combined with NaOCl (Haapasalo et al., 2014; Rath et al., 2020). On the other hand, EDTA is an irrigation solution most commonly used for smear layers removal. In addition to cleaning, it also has calcium chelation properties enabling the dissolution of dentin up to 20–30 μm depth in approximately five minutes by reacting with the underlying ions of calcium found within the calcified dentinal tubules (Attur et al., 2016). EDTA performs a powerful demineralizing action which leads to softening of the dentin, denaturation of collagen fiber and an enlargement in the size of this dentinal tubules. As a result, the obturating material may not be able to adapt appropriately within root canal walls. Spanó et al. The latter authors also highlighted another disadvantage of EDTA: it is classified as a pollutant because of its non-natural source (Giudicianni et al. 2009). In order to reduce the damaging effects of irrigants into periapical tissues, scientists are seeking a safer alternative for EDTA. Another chelating agent that can be employed is Chitosan, a low-cost and abundant natural material derived by the deacetylation of chitin found naturally in crab and shrimp shells. Glucosamine chitosan based on a natural polymer, has desirable properties such as antibacterial activity, biocompatibility, and biodegradability which makes it possible to base effective adhesive formulations (Del Carpio et al., 2015; Zhou et al). With the use of (CNP) chitosan nanoparticles, which have a higher absorption and penetration into the dentinal tubules, Ratih et al. (2020) report an increase in the efficiency of root canal irrigation (chitosan). Brasseler, USA, has developed Triton, an irrigation solution that combines the benefits of EDTA, CHX, and NaOCl in one easy step. It has a lower concentration of

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NaOCl solution, a unique combination of surfactants, and mild chelating agents. A patent application is pending for this product. Triton is utilized for continuous irrigation rather than as the ultimate irrigant, which makes it operate differently than other advanced 2:1 solution or classic irrigants. When organic waste is exposed to a reduced concentration of NaOCl, Triton's non-NaOCl constituents aggressively dissolve it, lowering the amount of buffering action needed (Brasseler, 2020). Because Triton eliminates using sterile water rinses and multiple irrigation solutions, chairside time is reduced. To the knowledge of the author, no prior study has compared the influence of Triton irrigation solution on the microhardness of root dentin. Therefore, the purpose of our study was to examine and assess the effects of several irrigation solutions on the microhardness of the root canal dentin, comprising 17% EDTA, 0.5% CNP, and Triton. The objective of this in vitro investigation was to evaluate the impact of 17% EDTA, 0.5% chitosan nanoparticles, and Triton on the microhardness of root dentin.

Materials and methods

Sample selection and preparation

This study was approved by the ethical review board of Mustansiriyah University, College of Dentistry (MUOPR23). A total of 40 mandibular premolars were collected from patients aged 25 to 35 years, extracted for orthodontic purposes. The following criteria were used to choose the teeth by using a radiograph: all roots needed to be free of cavities, fissures, past endodontic therapy, internal or external desorption; calcified canals, a single, straight canal and fully developed apices are ideal for every tooth. Following their extraction, the teeth were cleaned to get rid of any remaining hard tissues or soft tissue particles. After being kept for 24 hours at 37°C in a 0.1% thymol solution, they were preserved in regular saline, which was changed every day to avoid dehydration (Arun et al., 2022). To obtain a root length of 14 mm, the teeth's crowns were decorated. Next, a #10 K file was used to confirm the working length (WL). The file was inserted into the canal until the tip reached the foramen. A mm was added to the WL's creation. The root that had an initial size of 20 was the only one chosen.

Sample grouping

Based on the type of irrigation solution used, the samples were divided into four groups (n

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= 10): Group II is 5.25% NaOCl+ 0.5% CNP; Group III is 5.25% NaOCl+ 17% EDTA; Group IV is distilled water (control group); Group I is Triton all-in-one irrigant.

CNP preparation and evaluation

After 100 milliliters of 1% (v/v) acetic acid was added to dissolve 0.5 g of CNP powder (EPRUI, China), the mixture was vigorously stirred for eight hours. The samples underwent 40 minutes of sonication. In an independent experiment, 10 milliliters of distilled water were subjected to a 40-minute sonication followed by an 8-hour continuous stirring period for STPP (0.1 g). After that, drop by drop, the STPP solution was added to the CS solution until the CS:STPP ratio was 2:1. A 50 ml syringe was used, and 15 drops were dripped per minute. After a 40-minute sonication, this mixture was mixed for an extra eight hours (Bangun et al., 2018). Dynamic light scattering and a NanoBrook 90Plus Particle Size Analyzer (Brookhaven Instruments, USA) were used to determine the size of CS-TPP. The effective diameters of the CS suspensions were 84.4 nm.

Irrigation protocol

The irrigation protocol was as follows:

Group I Triton (Brasseler, Savannah, USA): Irrigating canals with approximately 5 ml as needed during instrumentation was done in accordance with the manufacturer's instructions. Five milliliters of distilled water were used for washing after the last irrigation, which required using one milliliter of Triton for 1.5 minutes (Brasseler, 2020).

Group II 0.5% CNP (EPRUI, China): 1 ml of 5.25% NaOCl is added to each of the three instrument strokes to yield an approximate volume of 5 ml. The final irrigation step was as follows, according to Hussein et al. (2022): irrigate with five milliliters of 0.5% CNP for three minutes, rinse with five milliliters of distilled water, and dry with paper point #4.

Group III 17% EDTA (Cerkamed, Poland): During each of the instrument's three strokes, about 5 ml of 5.25% NaOCl irrigation was added. After the final irrigation, 5 milliliters of 17% EDTA were added and left for three minutes. After that, 5 milliliters of distilled water were added, and paper point #40 was used to complete drying (Hussein et al., 2022).

Group IV (distilled water): approximately 5 ml of the distilled water was absorbed into the canals between each three instrument strokes. Final irrigation with 5 mL of distilled water in 3 minutes.

Irrigation with the irrigation needle followed within 2 mm of WL into the canal (Attur et al., 2016).

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Root canal instrumentation

ProTaper Next rotary files (Dentsply Maillefer, Ballaigues, Switzerland) X4, (#40/0.06) were used to instrument all of the root canals. As instructed by the manufacturer, the files were driven to their full WL using an electric motor operating at 300 rpm and producing 4.0 Ncm of torque. After the samples were taken out of the mold, two parallel longitudinal grooves were made on each root's buccal and lingual sections using a high-speed diamond bur (Shi et al., 2022). A tiny cotton plug was used to seal the hole. Gutta-percha was then inserted into the canal to act as a depth gauge for the groove and stop bur intrusion, which could have contaminated the canals with debris related to sectioning (Caron et al., 2010). Using a chisel, the root was then cut in half lengthwise along the channels, as illustrated in Fig. (1).

Statistical analysis

The data analyses were performed using SPSS Version 26.0 (SPSS Inc., Chicago, IL, USA), and the level of significance was set at p=0.05. Shapiro wilk test was applied to determine the normal distribution. ANOVA test was used for comparison among the groups whereas Post-hoc Tukey was applied to assess the differences between each two groups.

Results

The Vickers microhardness values (mean, ± S.D., minimum, and maximum) for each irrigating regimen are shown in Table 1. An ANOVA test was conducted to determine if there was a significant difference between the groups. The results are displayed in Table 2, indicating a statistically significant difference ($p \leq 0.05$) in the mean hardness between the four groups. The post-hoc Tuckey test revealed a statistically significant variation in the average dentin hardness between all tested groups as shown in Table 3.

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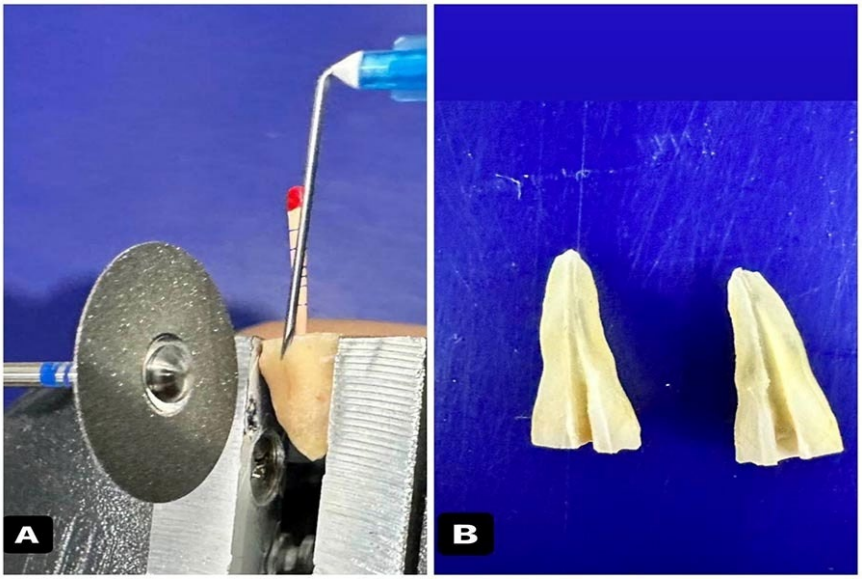

Figure 1. Splitting of the tooth, A: Parallel longitudinal grooves on the buccal and lingual aspects of each root were made by using a high-speed diamond bur under water-cooling. B: A chisel separates the root into two parts longitudinally along the grooves.

Table 1. The descriptive statistic for each experimental group's microhardness values (N/µm ²)

Groups	N	Mean	±SD	Min	Max
Triton I	10	63.2	3.162	61.4	69.9
CNP II	10	55.6	3.551	52.0	64.0
EDTA III	10	46.1	2.649	41.5	50.3
Distilled water IV	10	66.2	3.107	58.6	69.5

Table 2. One-way ANOVA test among all tested groups

	Sum of squares	df	Mean square	F	p-value
Between groups	2415.923	3	805.308	81.978	0.000
Within groups	353.643	36	9.823		
Total	2769.566	39			

Table 3. Employing the post-hoc Tuckey test, pairwise comparisons of the microhardness of the groups

Pairs	p-value	Significance
Group I-Group II	0.001	Significant
Group I- Group III	0.001	Significant
Group I- Group IV	0.040	Significant

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Group II- Group III	0.001	Significant
Group II-Group IV	0.001	Significant
Group III-Group IV	0.001	Significant

Discussion

According to the results of this study, all tested groups showed a significant difference with distilled water, which has a higher mean of microhardness than others.

The application of chelating agents decreased dentin microhardness, consistent with prior studies (Poggio et al., 2012; Nikhil et al., 2016; Quteifani et al., 2019; Tsenova et al., 2020), where the microhardness decreased as chelating agents were applied. A significant decrease in dentin hardness indicates that the irrigation treatment significantly altered the dentin structure.

With CNP and 17% EDTA, the Triton solution demonstrated the highest mean microhardness when compared to other irrigation methods. This may be due to differing contact times and concentrations of irrigant; the concentration of Naocl used in Triton is 4%, while the concentration used with other irrigation is 5.25%. Due to its proteolytic properties, NaOCl degrades collagen by breaking the fibers down into smaller peptide chains (Hülsmann, 2013), higher NaOCl concentrations (5%, for example) cause significant peripheral dentin matrix alterations (Aranda et al., 2013). This is clinically significant because higher concentrations of NaOCl (6%) penetrated the dentinal tubules up to 300 micrometers deeper than lower concentrations did (Zou et al., 2010). It is unknown how this affects the ultrastructure and characteristics of dentin. Moreover, NaOCl might gradually dissolve the collagen fibers that are encapsulated (Zou et al., 2010). This explanation was improved by a study done by Rath et al., which stated that the concentration and duration of NaOCl are the factors that may have an impact on the demineralization effect (Rath et al., 2020).

The composition of Triton, which contains only 2% citric acid and has a lower smear layer removal capacity as compared with other solutions, the concentrations of citric acid have been reported to vary between 1% and 50%. According to research done in 2008 by Reis et al., 10% citric acid was twice as effective as 1% citric acid. The fact that Triton removed less smear layer may have been caused by the lower concentration of citric acid (only 2%), but it was noted that using 6% was sufficient to remove the smear layer (Vallabhaneni et al., 2017). This

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indicates that citric acid is more potent at higher concentrations than at lower concentrations. According to a study that backed up this theory, the irrigants that removed more of the smear layer also changed the microhardness of the dentin. One explanation might be that the smear layer acts as a shield to keep irrigants away from dentin, enabling extremely little changes in the microhardness of the material (Dhawan et al., 2019).

EDTA demonstrated a significant reduction in microhardness in comparison to CNP, this is consistent with the findings of Berastegui et al. in 2017, who found that EDTA reduced dentin microhardness more than chitosan. However, the results of Antunes et al. 2020 showed that 15% EDTA and 0.2% chitosan did not significantly differ in dentin microhardness and that both materials had a similar effect on dentin microhardness when activated by endovac. This difference in results may be due to the usage of different material concentrations and activation of the irrigation materials by endovac.

It has been reported by Ari et al., 2005 and Poggio et al., 2012 that EDTA reduces dentin microhardness. Its chelating ability allows EDTA to lower dentinal microhardness effectively. EDTA binds to calcified components (particularly +2 ions) of dentin through chelating action; thus, in turn, it causes demineralization and softening of dentin. Panghi & G'Sell (1992) posit that the degree of mineral content and the amount of hydroxyapatite in the intratubular substance significantly impact the dentin structure's intrinsic hardness profile.

The exact mechanism of Chitosan's action is not fully understood. Because Chitosan polymer is hydrophilic, it is thought to favor close contact with root canal dentin and adsorbed to the walls of the root canal. Its cationic properties facilitate the ionic interaction between the chelating agents and are the calcium ions in dentin. (Zhang et al., 2010). Since 1% acetic acid is used to create a solution with chitosan, which is insoluble in water, the acid may increase the chelating effectiveness of the chitosan.

Conclusions

When comparing the effect of the irrigating solution on the microhardness of root dentin, triton had less deleterious effect on microhardness. While EDTA irrigation demonstrated the highest harmful effect on root dentin microhardness.

Supplementary Material

None.

Author Contributions

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Summer Hilu Hammdallah: data curation, writing-original draft preparation. Mohammed Qays Mahmoud Fahmi: Conceptualization, methodology, writing-review and editing. Haider Al-Waeli: validation, formal analysis, investigation, supervision.

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Data Availability Statement

Data are available from the authors upon reasonable request.

Conflict of interest

The authors reported that they have no conflicts of interest.

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