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Original research

EVALUATION OF PENETRATION DEPTH OF SODIUM HYPOCHLORITE INTO DENTINAL TUBULES AFTER PASSIVE ULTRASONIC IRRIGATION COMPARED TO ER;YAG LASER ACTIVATION. AN IN-VITRO STUDY

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ABSTRACT

Objectives: The aim of this study was to compare penetration depth of sodium hypochlorite into dentinal tubules after passive ultrasonic agitation and ER; YAG activation.

Materials and methods: Twenty-four single rooted human mature mandibular premolars were decoronated and accessed. After locating the apex and determining the working length, preparation of root canal was done up to #35 file using Mtwo system and with 5.25%NaOCl irrigation. Teeth were then sealed apically with wax and submerged in a crystal violet dye for 48 hours to stain dentin. NaOCl Irrigation was activated with either Ultrasonic or ER;YAG laser. Specimens were sectioned longitudinally and depth of bleached zone was evaluated under a stereomicroscope 40X.

Results: Penetration depth was significantly higher in overall root canal in ultrasonic group than ER;YAG laser group (P=.000). In ER;YAG Laser group, the highest penetration depth was in the coronal third followed by middle and apical, with significant difference between apical third and both middle and coronal thirds (P=.009, .003 respectively), and no significant difference between middle and coronal thirds (P=.083).Highest penetration depth was seen in the middle third, followed by coronal and apical,withno significant difference in penetration depth between the three sections of the root canal activated with Ultrasonic (P=.664).

Conclusion: ultrasonic activation can lead to more NaOCl penetration into dentinal tubules than activation with ER;YAG.

Key words: Sodium Hypochlorite, Ultrasonics, ER;YAG Laser

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INTRODUCTION

Successful endodontic treatment is dependent on the proper cleaning and shaping of canals to eliminate the remnants of vital and necrotic tissues, microorganisms and their toxins, and accumulated dentinal debris resulting from instrumentation. Mechanical instrumentation alone is insufficient to disinfect the whole root canal system.[1] A study conducted by Peters*et al.*[2] showed that 35% of the root canal remained untouched after instrumentation. Thus, chemical irrigation has an important complementary role in disinfecting rootcanals, which allows the penetration to dentinal tubules, isthmus, lateral canals, and apical ramifications.[3,4]

Sodium hypochlorite has been introduced to endodontics as the main and most common irrigant due to its antibacterial effect, the ability to dissolve organic tissues and pulpal residuals, and the penetration of lateral canals, which is attributed to its low viscosity.[5–7]

Traditional irrigation with a syringe and needle has limited penetration into lateral canals.[8]Activating endodntic irrigants has deemed to increase the disinfecting properties of irrigants through the enhancement of its chemical and physical actions.[9]

Ultrasound was first introduced to Endodontics in 1957.[10]Passive Ultrasonic Irrigation (PUI)can disinfect root canal through the improvement of irrigant contact to the root canal walls, and its cavitation and acoustic streaming effects.[11] A review of the literature described the antibacterial effect of the PUI system, and its efficacy in better removal of the smear layer, facilitating cleaning the isthmus from curved root canals.[12]The rapid movement of this device enhances the shear stress of tissue remnants and biofilm.[13]

More Recently, Laser activated irrigant (LAI) has gained attention in root canal irrigation.[14]LAI has the ability to enhance root canal debridement by its photoacoustic wave shocks in the irrigating which results in vaporization of the irrigant, creating vapor bubbles that can expand and explode with cavitation effect. Expansions cause high pressures which leads to the rupture of bubbles.[15]

The literature yielded conflicting results regarding the most effective way in enhancing the penetration of NaOCl into dentinal tubules. While some studiesreported that laser activation is better than ultrasonic devices[14,16], others stated opposite results[17,18]. Thus, the aim of this study was to compare PUI and ER;YAG (LAI) in penetration depth of NaOCl into dentinal tubules.

MATERIALS AND METHODS

This study was carried out at the department of Operative Dentistry, Faculty of Dentistry, Hama University, and an approval of the Scientific Research Committee of Hama University with the ID: 307 has been obtained on 14/2/2018 before the initiation of the study.

Sample size calculation was done using G*power program v.3.1 (Heinrich-Hein-UniversitatDüsseldorf, Germany; http://www.gpower.hhu.de/), and 24 freshly extracted human mature single-rooted mandibular premolars were determined as the total specimens. Teeth were extracted due to orthodontic reasons.

Tooth selection criteria were as follows: single canal, no external or root internal resorption, lack of tooth caries, cracks or developmental anomalies under 20X magnification, and no previously endodontically treated canals, with canal curvature of no more than 5° according to Schneider.[19]

Teeth were debrided using CK6 hand instrument (Zeffiro-Lascod, Florence, Italy) to remove all tissue debris attached to the root surface after extraction. Then, teeth were stored in a plastic container with 0.5% chloramine T for 1 week to sterilize the specimens, before they were moved to another plastic container with 0.9% saline and kept in a refrigerator at 4°C until used.

Sample preparation

The crown of teeth were shortened using a diamond disk (Edenta, Switzerland) to standardize the length at 19 mm using a digital caliper.

Conventional access cavity was done using a 2mm round bur, and the roof of the pulp chamber

was removed with Endo-Z bur (Dentsply, Switzerland). The pulp of each tooth was extirpated with barbed broaches (VDW, Germany). The working length was measured after locating the apex with a 15# K-file until the tip of the file was observed from the apical foramen, then subtracting 1mm from the canal length.

Root canal preparation

Canals were prepared with Mtwo system (VDW; Germany) following the basic instrument sequence until #35 along the entire working length using a gentle in and out motions. Preparation was done according to manufacturer's instruction regarding the speed and torque. 2 ml of 5.25% NaOCl was irrigated after each instrument and a final rinse with 5ml NaCl was done before drying specimens with paper points.

The root surface of each tooth was covered with two layers of nail varnish, and the apical foramen was sealed in order to prevent the dye from leakage outside the canals.

A preliminary study was conducted in order to determine the most appropriate periodfor the type of sectioning (cross sectional vs. longitudinal), and the pigmentation process in which teeth were submerged in the crystal violet dye for 12 or 24 or 48 hours, and the final period was found to be the appropriate one to stain the hole root canal.

Specimens were submerged in a plastic container with crystal violet dye for 48 hours in 37°C temperature. Teeth were then washed with running water to flush away the dye.

Study groups and irrigation protocols

Specimens were randomly distributed into two groups based on the activation method as follows:

Group A (n=12): activation using ultrasonic device (Varios 350, NSK, Japan).

Group B (n=12): activated using ER:YAG laser (Kavo Key Laser III 1243, Germany).

In each group, 2 ml of 5.25% NaOCl was introduced for 60 secondsusing a 30-Gauge open ended needle 1 mm shorter of the apical foramen.

In group A: 2 ml of 5.25% NaOCl was introduced for 60 secondsusing a 30-Gauge open ended needle 1 mm shorter of the apical foramen. Ultrasonic activation was performed by inserting a stainless steel #22 U file into E11 Ultrasonic tip (Varios, NSK, Japan) at E4 power to agitate irrigant for 60s and 1 mm above the apical foramen. Root canal was irrigated then with 2 ml of 5.25% NaOCl for another 60s and a final rinse was accomplished with saline for 60s. A final rinse with saline solution was applied for 60 seconds.

In group B: 2 ml of 5.25% NaOCl was introduced for 60 seconds using a 30-Gauge open ended needle 1 mm shorter of the apical foramen. ER:YAG laser was used for activation with a special fiber head designed for endodontic usage (100 mill joules, 2 watts, frequency: 20 Hz) for 60s. Root canal was irrigated then with 2 ml of 5.25% NaOCl for another 60s and a final rinse was accomplished with saline for 60s. A final rinse with saline solution was applied for 60 second.

Teeth sectioning and measuring the depth of NaOCl penetration

Teeth were sectioned longitudinally after creating a groove of 1 mm depth on the mesial and distal surface of the root under an endodontic microscope with 30x magnification. Then, each tooth was put on a polyvinyl siloxane model in order to separate the two sections with a chisel and a mallet.[20]

Two parallel lines, 3 mm apart, were drawn on the surface of each section to calibrate the image. Each section was captured with a digital camera (Samsung NX500, Samsung; USA) under a stereomicroscope (Meiji; Japan) at 40X. The depth of sodium hypochlorite penetration was measured using Microdicom Program by evaluating the whitened areas inside the dentinal tubules in each third (coronal, middle, apical) of the tooth, where the dye was removed due to the oxidization effect of sodium hypochlorite. (Fig1 and 2).

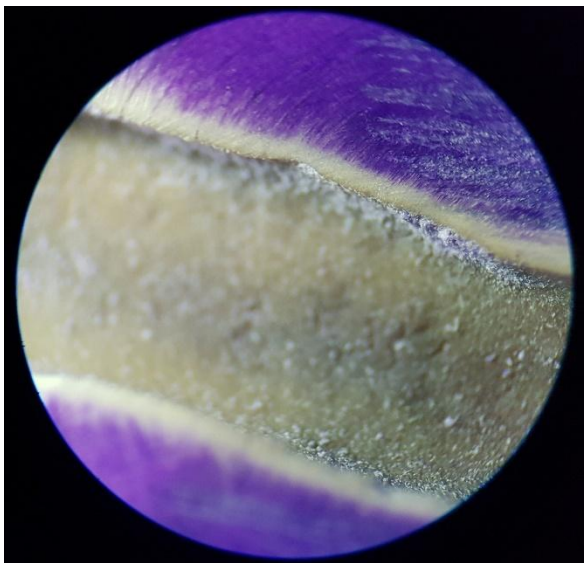

Figure 1 Shows the whiten bleached area that indicates penetration depth of NaOCl in PUI

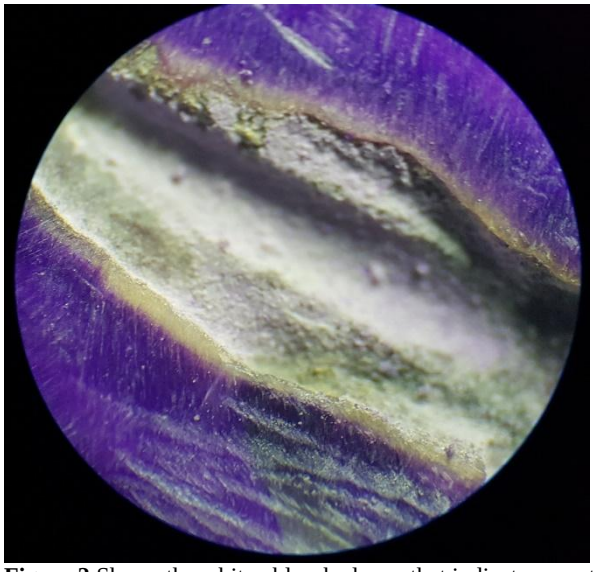

Figure 2 Shows the whiten bleached area that indicates penetration depth of NaOCl in LAI

Statistical Analysis

Normality of distribution was checked with Kolmogorov-Smirnov test. Data showed normal distribution. Thus, independent T test was used to determine if there was a statistically significant difference in NaOCl penetration depth betweengroupA and group B in each third (coronal, middle, third), and ANOVA test to determine if there was a statistically significant difference in the depth of penetration between the three sectionsof the root surfacewithin each group

Data were analyzed using SPSS V.23 (IBM; CORP., ARMONK, USA). The level of the P value was set 5%, and the level of confidence was set at 95%.

RESULTS

Descriptive results including minimum, maximum, mean, and standard deviation of NaOCl penetration depth in each third of the root surface and overall root canal surface areshown in (Table 1). Group A activated with ultrasonic showed more penetration depth in the three thirds of the root canal compared to groupB activated with ER:YAG laser.

Table1. Shows descriptive results of the Independent T test regarding the penetration depth of NaOCl between PUI amd LAI within each level of the root anal (coronal, middle, and apical third) in (mm)

Levels	Activation type	min	max	mean±SD	P Value
Apical third	**PUI**	0.17	0.62	0.45±0.13	.002*
	LAI	0.1	0.62	0.25±0.14	
Middle third	**PUI**	0.25	0.86	0.62±0.18	.003*
	LAI	0.26	0.65	0.41±0.12	
Coronal third	**PUI**	0.32	1.03	0.59±0.24	.076
	LAI	0.28	0.8	0.44±0.16	
Overall	**PUI**	0.17	1.03	0.55±0.20	.000*
	LAI	0.1	0.8	0.37±0.16	

*significant difference
SD- Standard Deviation

Independent T test showed significantly higher penetration depth in the apical, middle third, and overall root canal length in Ultrasonic group compared to ER:YAG laser group (P= 0.002, 0.003, 0.000 respectively). No significant difference in penetration depth in the coronal third between the two groups (P=0.076) (Table 1).

Descriptive results showed higher penetration depth in the middle third, followed by coronal, and apical third in group A. However,in group B, highest penetration depth was seen in coronal third followed by middle and apical thirds. (Table 2)

Table2. Shows descriptive results of the ANOVA test regarding the penetration depth of NaOCl between each level of the root anal (coronal, middle, and apical third) in PUI and LAI groups (mm)

groups	Levels	min	max	mean±SD	P Value
	Apical third	0.17	0.62	0.45±0.13	
PUI	**Middle third**	0.25	0.86	0.62±0.18	.0006*
	Coronal third	0.32	1.03	0.59±0.24	
	Apical third	0.1	0.62	0.25±0.14	
LAI	**Middle third**	0.26	0.65	0.41±0.12	.083
	Coronal third	0.28	0.8	0.44 ±0.16	

*significant difference
SD- Standard Deviation

ANOVA test showed no significant difference in penetration depth of NaOCl between the three thirds of the root canal within group A (activation with Ultrasonic) (P=0.083). However, significant difference in penetration depth between the three thirds of the root canal within group B (activation with ER:YAG Laser) (P=0.006) (Table 2).

LSD Post hoc test for pairwise comparison in group B (activation with ER:YAG Laser) showed significant difference between apical third and both middle and coronal thirds (P= 0.009, 0.003 respectively). No significant difference was observed between middle and coronal thirds (P=0.0664) (Table 3).

Table 3. LSD pairwise comparison between the three Levels of root canal in LAI group in (mm)

group	Levels		Difference in means	SE	P Value
LAI	**Apical third**	**Coronal third**	-0.18	0.06	.003*
		Middle third	-0.16	0.06	.009*
	Middle third	**Coronal third**	-0.02	0.06	.664

*significant difference
SE -Standard Error

DISCUSSION

Bacteria of severely infected root canal can invade smear layer along the root canal into dentinal tubules in lateral canals and as a result be responsible of treatment failure.[21] Smear layer can interfere with the penetration of irrigants and antimicrobial agents into dentinal tubules.[22] Nair *et al.*[23] showed residual infection in mesial root canals of mandibular molars after instrumentation, irrigation with NaOCl alone, and obturation. Therefore, enhancement of irrigation by devices is important to allow irrigants to disinfect these inaccessible areas.[22]

It has been declared that the presence of smear layer can impede the penetration of NaOCl into dentinal tubules.[24]

Olivi.[25] explained the effect of Er;YAG laser on dentinal debris and smear layer removal, besides its ability to enhance the action of NaOCl.This enhancement has been related to the shockwaves of lasers that can be absorbed through NaOCl creating vapor bubbles that can expand and implode reducing smear layer.[26]

On the other hand, Hazar *et al.*[27] showed high efficacy of ultrasonic agitation in triple antibiotic paste removal from the apical portion of the root canal, which was related to the high velocity of irrigant flow, while Uzunoglu *et al.*[28] showed that some ultrasonic dependent devices can have good effect in removing CaOH from root canal. In addition, ultrasonic can enhance irrigation through disinfection and smear layer removal through bubbles generated by cavitation and acoustic streaming effects.[22] Therefore, this study aimed to compare activation of Er;YAG laser, and Ultrasonic in penetration depth of NaOCl.

Crystal violet dye was chosen in this study due to its high ability in pigment dentine so it can be easily seen under a stereomicroscope, and since NaOCl is an antioxidant it can whiten the purple color of the dye revealing the normal color of dentine.

This study assessed the penetration depth after sectioning teeth longitudinally with a chisel and a mallet, in order to maintain the inside portion of the root canal intact and preserveit from damage that could be attributed due to diamond disc sectioning.

Studies showed that bacteria can penetrate dentinal walls and lateral canals into different depths.[29,30] In 62% of cases, bacteria can invade dentinal tubules and reach the surface of cementum.[31] This bacteria can be responsible of endodontic treatment failure.[32]

With regards to activation with Er;YAG laser, this study showed highest penetration depth in the coronal third, followed by middle and apical third. Significant difference was seen between apical third and both middle and coronal thirds. This was consistent with Rajakumaran *et al*.[33] and Ghorbanzadeh *et al.*[16] study that showed highest penetration depth with laser activated irrigant in coronal third followed by middle and apical thirds. Highest penetration depth in coronal third could be attributed to thelarge and densely packed dentinal tubules seen in the coronal thirdof the root canal followed by middle third, while narrower tubules are located more in the apical thirds, and as a result can limit the ability of irrigant penetration.[34] Both groups demonstrated minimum penetration depth in the apical third of the root canal, this was in agreement with Ghorbanzade *et al.*[16], Macias *et al.*[35], and Vandrangi.[36]

However, interesting findings was observed in Ultrasonic activation, where the highest penetration depth was in middle third, followed by coronal and apical with no significant differences between the three thirds. This finding was in agreement with Vandrangi.[36]Macias *et al.* [35] also showed highest penetration of Chinese ink in the middle third of the root canal when ultrasonic was used.In contrast, Baz *et al.*[37] showed lower penetration depth in middle third than coronal. However, different irrigation protocols were used with up and down motions in activation, while in this study the PUI was set at one position.

This study showed that penetration depth of ultrasonic was significantly higher compared to Er;YAG laser in overall root canal length. Therefore, within the confines of this study, the use of expensive laser devices is not necessary. Moor *et al.*[17] revealed that laser devices could be replaced with ultrasonic devices in order to remove smear layer, on condition that activation of ultrasonic should be for 60 seconds in total, which was similar time in the present study. Although Deleu and Meire [18] revealed better smear layer removal in LAI than PUI, no significant difference was observed between the two groups.

This study was not consistent with Ghorbanzade *et al.*[16] study which revealed that laser activation is better than ultrasonic with regards to penetration depth. However, this could be attributed to the different type of laser activation (Nd; YAG) used in their studies, and less activation times.Schlichting and Widbiller.[14] showed better penetration when PIPS was used compared to ultrasonic, but this difference could be related to the irrigation regimen used in their study, where NaOCl was preheated in conjunction to the use of EDTA as a final irrigation protocol.

CONCLUSIONS

Passive ultrasonic agitation can be an effective irrigant activation method through enhancing the penetration depth of NaOCl into dentinal tubules more than ER;YAG laser.

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CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

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