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Smear Layer Removal from the Apical Third Using the Er,Cr:YSGG Photon-induced Photoacoustic Streaming

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ABSTRACT

Introduction: The purpose was to study the influence of Erbium,Chromium:Yttrium Scandium Gallium Garnet (Er,Cr:YSGG) laser at short pulse duration on smear layer removal from apical root third. **Materials and Methods**: Twenty-four single-rooted mandibular premolars were used. The roots length was cut to 14 mm and instrumented using the Protaper Gold system to size 40/0.06. The samples were divided into four groups (*n*=6) as follows: (A) conventional irrigation with 5 mL of 17% Ethylenediaminetetraacetic acid (EDTA), (B) passive ultrasonic irrigation with 5mL of 17% EDTA, (C) Er,Cr:YSGG induced photoacoustic streaming (PIPS) with 5 mL of 17% EDTA and (D)Er,Cr: YSGG induced PIPS with 5 mL of 5.25% sodium hypochlorite (NaOCl). After final irrigation with normal saline and drying, the roots were painted with nail varnish externally, and 2%methylene blue dye was injected into the canal. The tooth split horizontally at the fourth millimetres. The picture was taken by a professional Digital SLR camera. The dye penetration was measured using analytical software (measure picture CAD-KAS Kessler Germany). Dunnett's test was used to compare control group against experimental groups. **Results:** After the statistical test, the highest mean percentage was presented in the laser group with 17% EDTA (Group C=85.9804) followed by the laser group with 5.25% NaOCl (Group D=69.7817) and ultrasonic group (Group B=34.5453) respectively and the lowest mean percentage was in the control group (Group A=20.7969). **Conclusion:** Based on this in vitro study, PIPS technique using Er,Cr:YSGG pulse laser (0.5 W,5 Hz, air and water off) at short pulse duration and 17% EDTA proved effectively in smear layer removal, while the PIPS proved an acceptable result when the laser is used alone assisted by NaOCl.

Keywords: Endodontics; Er,Cr:YSGG Laser; Irrigation; Photoacoustic Effect; Smear Layer; Sodium Hypochlorite

Introduction

One of the main requirements to reach successful root canal treatment is the cleaning and shaping of root canals. In addition to chemomechanical preparation of the root canal(s), hermetic three dimensional sealing of the root canal is considered a key factor in successful endodontic therapy [1]. Different factors/causes can contribute to unsuccessful endodontic treatment; *e.g.* the presence of bacteria in the root canal(s) and dentinal tubules as well as irregular and apical canals [2]. Therefore, chemical cleansing is required *via* irrigation alongside mechanical preparation. Numerous advancements have been introduced to endodontics; *e.g.* hand-/engine-driven instruments and altered irrigating solutions [3]. Plotino *et al.* [4] used endodontic files with different sizes/tapers and concluded that all basic root canal preparation techniques were accompanied by less smear layer and/or debris on canal walls in the coronal and middle thirds, without revealing any differences between them. Although in the apical third, debris and smear layer were always present, an apical file size of 25, compared to a size of 20, resulted in significantly cleaner canal walls [4]. Irrigation is a critical part of successful endodontic treatment as it performs numerous important chemical, mechanical and (micro)biological functions [3]. Seemingly, irrigation is the only method to reach distant areas of the root canal/walls which are not touched and/or cleaned *via* mechanical instrumentation [2]. An ideal chemical irrigant should have antibacterial effect and act as a tissue solvent/lubricant in addition to physical flush for debris exclusion [5]. A mixture of two or more irrigants is vital for safe and effective irrigation; because no single solution has desired effects [6]. Irrigation with sodium hypochlorite (NaOCl) is critical for achieving success in endodontic treatments, and several agitation techniques have been developed to improve the efficacy of this irrigation method [7]. Ethylenediaminetetra-acetic acid (EDTA) is the irrigant of choice for smear layer removal, assisting in the removal of inorganic constituents of smear layer and acting as an aide to irrigation [8]. EDTA decalcifies dentine to a distance of 20–30 µm in 5 min [9]. Using irrigation solutions, especially 17% EDTA + 5.25% NaOCl, has been effective in smear layer removal, although the mechanical pressure of saline was somewhat able to remove smear layer [10]. An ultrasonic unit for use in endodontics to debride root canals, designed by Martin *et al.*, has become commercially available [11].

Root canal irrigants agitation/activation by use of lasers is a new concept in endodontics. The mechanism of interaction between dental hard tissue (enamel, dentine) and Erbium lasers family is the explosive thermo-mechanical or water-mediated ablation; the procedure that occurs within wavelengths between 2.7 µm and 3 µm, and leads to the expulsion of mineral particles with the conservation of their mineral structure [12]. Montero-Miralles *et al.* [13] conducted a study to compare smear layer/debris removal with EDTA and Erbium, Chromium: Yttrium Scandium Gallium Garnet (Er,Cr:YSGG) laser and concluded that Er,Cr:YSGG laser displayed a better cleanliness in the middle third, with statistically significance differences compared to 17% EDTA [13]. The impact of laser in the removal of smear layer can be more efficient than other conventional approaches for agitation [14]. In 2020, Yilmaz *et al.* [15] considered the efficiency of varied irrigation methods on the penetration of sealers using confocal laser scanning microscopy and found higher penetration rates in all experimental groups compared to the control with maximum penetration depth of 652 µm [15]. The current study aimed to investigate the effect of Erbium, Chromium: Yttrium Scandium Gallium Garnet (Er,Cr:YSGG) laser at short pulse on the removal of smear layer in the radicular apical third using 24 single-rooted human mandibular premolars with (17% EDTA) and 5.25% NaOCl as disinfection solutions next to the shock wave generation for photon-induced photoacoustic streaming (PIPS) technique. The effects were then compared using ultrasonic technique.

Materials and Methods

Twenty-four straight/single-rooted mandibular premolar teeth with completely formed/matured canal, extracted for orthodontic or periodontics purposes, were selected. The teeth were clinically and radiographically examined to ensure that there were no root fractures, open apices and/or root resorption. Then, they were stored in a plastic container containing 0.1% thymol solution until performing the experiment.

The roots length was uniform to 14 mm from the anatomic apex. The exact apical foramen location and canals patency were recognized using a stainless steel K-file #10 (Dentsply Maillefer, Ballaigues, Switzerland) and the correct working length was determined *via* deducting 1mm from the length previously measured.

The canals were mechanically instrumented using Rotary ProTaper Gold NiTi endodontic files (Dentsply, Maillefer, Ballaigues, Switzerland) (SX, S1, S2, F1, F2, F3 and F4) to size F4 (size 40, 0.06 taper) and irrigated with 2.0 mL of 5.25% NaOCl (Chloraxid Extra, PPH Cerkamed, Stalwa Wola, Poland) at each instrument change. Next, ENDO1 (Guilin Woodpecker Medical Instrument Co., Guilin, Guangxi, China) was used with an ultrasonic activator tip ED98 (#25, 2% taper, 18.5 mm) (Guilin Woodpecker Medical Instrument Co., Guilin, Guangxi, China). Then, Er.Cr: YSGG pulse laser (Waterlase iPlus Biolase, CA, USA) with 2780 nm wavelength was irradiated at short pulse duration (60 µsec) delivered by MD/iplus Glass Tips (MZ6) with 600 µm in diameter, length of 6 mm and calibration factor of 1.00. The device settings were set as follows: Power=0.5 W for EDTA, 0.75 W for NaOCl group, pulse duration: 60 µsec and repetition rate: 5 Hz, with water and air turned off. We did pilot study on different laser powers (0.25 W,0.5 W,0.75 W,1 W,1.25 W) and conclude that the best power used with EDTA is 0.5 W and for NaOCl is 0.75 W. The specimens were arbitrarily divided into four groups:

G1: control group (n=6): The samples were irrigated with 5 mL of 17% EDTA (Disodium edetate, PPH Cerkamed, Stalwa Wola, Poland) for 1 min *via* side-vented irrigation needle positioned within 2 mm of the working length. Then, the canal was irrigated with 5 mL of distilled water and dried with paper point F4 (Diadent, Seoul, Korea).

Figure 1. Calibration; to change pixels into millimeters using numerical scale

G2: ultrasonic activation (n=6): The samples were irrigated with 5 mL of 17% EDTA for 1 min. The ultrasonic system Ultrasonic activator ENDO1 (Guilin Woodpecker Medical Instrument Co., Ltd. china) was used according to manufacturer's instructions. After the injection of solution into the root canal, the ultrasonic activator tip ED98 was fitted passively inside the canal 2 mm shorter than the working length, and then, the tip was moved in an up and down motion in a small range. After 20 sec, the action was stopped in order to clean the root canal. Each canal was irrigated for 3 times and each time took 20 sec. The final rinse was performed with 5 mL of distilled water and then, the canal was dried with F4 paper point (Diadent, Seoul, Korea).

G3: 17% EDTA + pulsed Er,Cr:YSGG laser 2780 nm (n=6): The samples were irrigated with 5 mL 17% EDTA and agitated with Er,Cr:YSGG pulsed laser 2780 nm for 1 min. The delivery was *via* MD/iplus Glass Tips (MZ6) (Biolase, San Clemente, CA, USA). The fiber tip was inserted into the canal orifice. Then, the final rinse with 5 mL of distilled water was performed and F4 paper point (Diadent) was used to dry the canal. Each fiber tip was used for only one canal, and discarded afterwards.

G4:- 5.25% NaOCl + pulsed Er,Cr:YSGG laser 2780 nm (n=6): Er,Cr:YSGG pulsed laser was used for 1 min with 5 mL of 5.25% NaOCl for PIPS technique (shock wave generation and disinfection).

The delivery was by MD/iplus Glass Tips (MZ6) (Biolase, San Clemente, CA, USA) with water and air turned off. The fiber tip was inserted into canal orifice. Then, the final rinse was performed with 5 mL of distilled water and the canal was dried using F4 paper point. Each fiber tip was used for only one canal, and discarded afterwards.

Permeability test experiment

The test was executed/performed to assess the dye penetration zone in the apical third of the root canal. Wax was initially used to seal the root apex. Then, two layers of nail paint were left to dry and cover the surface of root(s). Afterwards, 2% methylene blue dye (India) was introduced to the canal using hypodermal syringe with needle gauge #23, *via* placing the needle 2 mm inside the canal. Next, K-file #20 (Dentsply Maillefer, Ballaigues, Switzerland) was inserted and then withdrawn once to confirm that the dye had reached the apical third of the canal [16, 17]. The dye was left inside the canal for 20 min at room temperature (27 - 29°C). Subsequently, they were copiously washed under running tap water to clean the external root surface. The root canal(s) were constantly dried with absorbent paper cones.

Root sectioning for permeability test

The tooth was split at 4 mm from the root apex using a diamond disc immediately below the guiding line representing the apical third. Images were taken using Professional Digital SLR camera (Nikon D7100, Nikon Corporation, Thailand) for each sample under 40× magnification.

Radicular dentine permeability measurement and evaluation

The obtained images were opened with analytical software (Measure pictures version 1.0; CAD-KAS Kassler Computer software GbR, Markranstädt, Germany) for computing radicular dentine permeability. The dye penetration and the total root section areas were calculated. Then, the whole area of the root canal was subtracted from the earlier mentioned areas to acquire the actual zone of dye penetration and root section. Calibration was initially done *via* numerical scale in order to change pixels into millimeters (Figures 1 and 2). The Data on permeability of root canal dentine was stated as the percentage of dye penetrating area at the apical third of root canal using the following equation: Dye penetration percentage in root section=(Net dye penetration area/Net total root third area) 100×.

Statistical analysis

Dunnett's Test was used to compare the control group against experimental groups to see there is a difference. Values of $P>0.05$ were reflected statically non-significant while $P\leq0.05$ and $P<0.01$, 0.001 were reflected significantly different, highly significantly difference, respectively. The statistical analysis done with SPSS software (SPSS version 20; SPSS, Chicago, IL, USA).

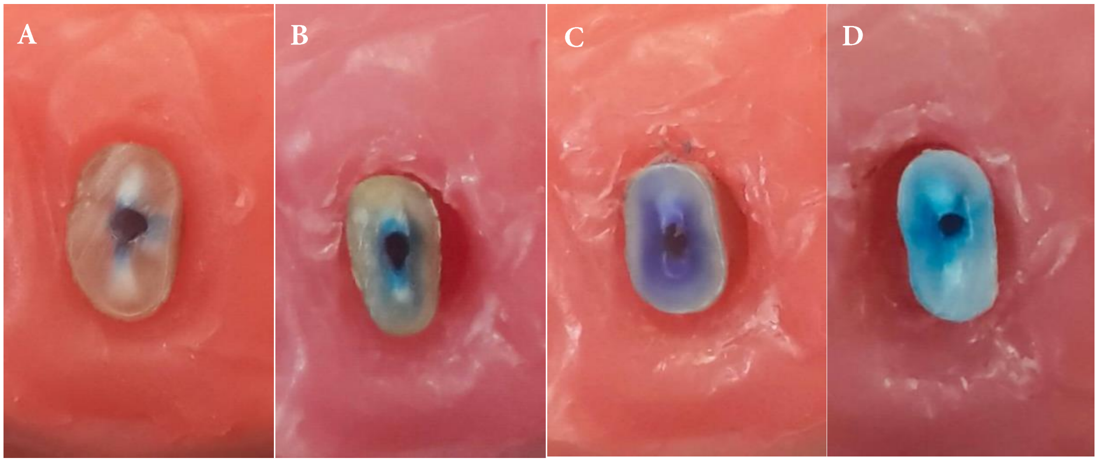

***Figure 2**. A*) Image after transversal cuts and penetration of dye solution for control group; *B*) 17% EDTA + ultrasonic group; *C*) 17% EDTA + pulsed Er,Cr.YSGG laser group; *D*) 5.25% NaOCl + pulsed Er,Cr.YSGG laser group

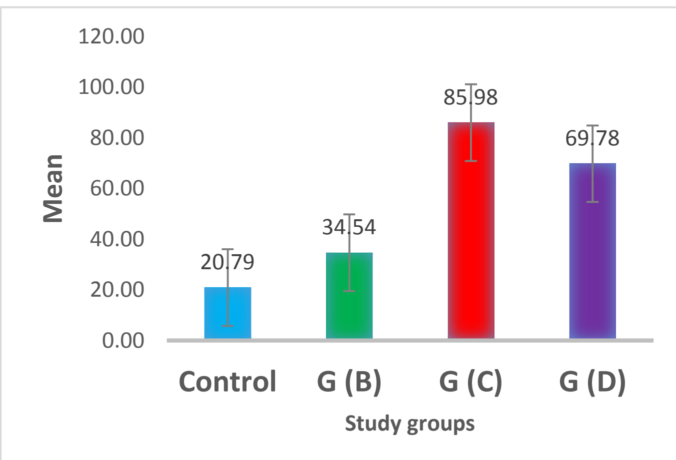

Figure 3. Bar chart showing the mean of dye penetration percentage of the study groups

Results

Table 1 displays the summary of descriptive statistics of permeability amongst groups, and demonstrates that the highest mean percentage was presented in the activated Erbium laser group with 17% EDTA (Group C), followed by the activated Erbium laser group with 5.25% NaOCl (Group D) and ultrasonic group (Group B). The lowest mean percentage was reported in the control group (Group A).

Dunnett's Test was used to compare the mean of control group against the mean of experimental groups in order to study the difference. Table 2 shows that the high mean difference was between the control group and the activated Erbium laser group with 17% EDTA (Group C), followed by the activated Erbium laser group with 5.25% NaOCl (Group D) and ultrasonic group (Group B).

Figure 3 exhibits the mean values of the dye penetration area in percentage amongst all four groups and shows that the percentage of dye penetration area increased in the ultrasonic group and became highest in 17% EDTA + Er:Cr.YSGG. Then there will some reduction in percentage when laser used alone.

Discussion

In this study, four different agitation procedures were examined in terms of their impact on the removal of smear layer in the apical third of root canals. The procedures/techniques consisted of the agitation of an irrigation solution using a conventional syringe; *i.e.* passive ultrasonic irrigation (PUI), Er,Cr:YSGG photon-induced photoacoustic streaming (PIPS) with 5 mL of 17% EDTA and Er,Cr:YSGG photon-induced photoacoustic streaming (PIPS) with 5 mL of 5.25 % NaOCl. Statistical analysis revealed that the high mean difference of dye penetration area was observed between the control and activated Erbium laser groups with 17% EDTA (Group C) (65.18348*) followed by Erbium laser group with 5.25% NaOCl (Group D) (48.98482*) and ultrasonic group (Group B) (13.74838*). In a previous study that used Er,Cr:YSGG laser with "Radial Firing Tip", the mean value of dye penetration percentage in apical root third was (59.118) [18]; however, in the present research, a better result (85.980) was obtained *via* using PIPS technique.

Based on the obtained results, the mean values of the percentage of dye penetration area in the three experimental groups were higher than that of the control group. The percentage of dye penetration in Er,Cr:YSGG laser induced photoacoustic streaming with 17% EDTA was significantly high, followed by Er,Cr:YSGG laser induced photoacoustic streaming with 5.25 %

NaOCl laser group that showed a distinct percentage of dye penetration, while the ultrasonic activated group resulted in less percentage of dye penetration than both Erbium groups. Using rotary instruments and chemical irrigation in existing instrumentation techniques cannot successfully remove the smear layer; a phenomenon which was seen in the control group (group A) where conventional method was used.

The Er,Cr:YSGG laser in the current study was used with 600 µm diameter Glass Tips (MZ6) by the means of subablative parameters (power 0.5 W with irrigant and 0.75 W without irrigant, 5 Hz) at short pulse (60 µsec) and showed the best effect in the removal of smear layer compared to conventional and ultrasonic methods. The main reason could be the photomechanical effect, which occurs after light energy is pulsed in a liquid [19], [20]. Once activation occurs in a liquid with minimum volume, the Er,Cr:YSGG wavelength absorption in water, with its peak power after the short pulse (60 µsec), may cause the photomechanical phenomenon. It was conclude that the resulted phenomenon was the reason for the elimination of smear layer in group 4, where laser irradiation was joined with NaOCl. The laser irradiation alone did not disturb the smear layer but could be used in PIPS for shock wave generation and disinfection. Moreover, a reflective "shockwave-like" impact was obtained after tips immersed in root canal filled with liquid. As a result, the effect of the diminutive volume can remove residual tissue/the smear layer and lower the bacterial content within the tubules and accessory canals [21-23]. In this study, the smear layer was removed by means of photomechanical flowing of liquids due to laser activation in the coronal access of the root, not by the usual thermal vaporization. This light energy phenomenon is referred to as photon-induced photoacoustic streaming (PIPS). The current study showed that the irradiation impact of Er,Cr:YSGG laser at subablative power settings was enhanced by the existence of EDTA, as it was seen in group 3. This leads to significantly better percentage of dye penetration area in comparison to all groups; because when the specimens were cleaned with EDTA, the delivery and transformation of the laser to thermal energy directly into the dentine matrix disclosed the existence of a low mineral constitute related to the effect of EDTA [24].

It is important that irrigants are brought into the direct contact with entire canal wall surfaces for their effective action, particularly in the apical portions of root canal due to the typically challenging complexity of root canal morphology [25]. In conventional treatment procedure, the irrigation syringe becomes more active when the tip is positioned closer to the working length; however, when in laser method, the tip is not positioned within the canal and is restricted to the coronal access beyond the orifice. Therefore, the laser method allows simple access and photomechanical effects can happen inside the root canal. This can help clean numerous canals shapes. In addition, standard laser applications need typical preparation at minimum size of 30, with the laser tip reaching the apical root third. Nonetheless, the PIPS tip can be placed in the root canal(s)' coronal reservoir and does not need deep penetration to the canal end. As a result, the laser permits minimally invasive cleaning of the canal system [26].

Table 1. Descriptive statistics of Permeability amongst groups

Test Groups (N)	Mean	95% Confidence interval for mean		Minimum	Maximum
		Lower bound	Upper bound		
A (Control) (*n*=6)	20.79	19.58	22.01	19.10	22.00
B (*n*=6)	34.54	34.06	35.02	34.06	35.26
C (*n*=6)	85.98	83.15	88.80	82.43	89.76
D (*n*=6)	69.78	68.35	71.20	67.86	71.95
Total (24)	52.77	41.45	64.09	19.10	89.76

A) Conventional irrigation with 5 mL of 17 % EDTA, B) Passive ultrasonic irrigation with 5 mL of 17% EDTA, C) Er.Cr:YSGG induced photoacoustic streaming with 5 mL of 17% EDTA, D) Er.Cr:YSGG induced photoacoustic streaming with 5 mL 5.25% NaOCl

Table 2. Multiple Comparison between control and experimental groups (Dependent Variable: Test Groups; Dunnett t (2-sided) [a]

(I) Group	(J) Group	Mean difference (I-J) (Sd)	Significant difference
B	Control A	13.74838*(0.94205)	0.000
C	Control A	65.18348* (0.94205)	0.000
D	Control A	48.98482* (0.94205)	0.000

*A) Conventional irrigation with 5 mL of 17% EDTA, B) Passive ultrasonic irrigation with 5 mL of 17% EDTA, C) Er.Cr:YSGG induced photoacoustic streaming with 5 mL of 17% EDTA, D) Er.Cr:YSGG induced photoacoustic streaming with 5 mL 5.25% NaOCl. (*The mean difference is significant at 0.05); a. Dunnett t-tests treat one group as control, and compare all the other groups against it*

Conclusions

It is concluded that using Er,Cr:YSGG pulsed laser (0.5 W, 5 Hz, air and water off) at short pulse duration (60 µsec) and 17% EDTA using PIPS technique for the activation of an irrigant in endodontic treatment proved effective for the removal of smear layer, whereas PIPS showed an acceptable result when it was used alone assisted by NaOCl.

Conflict of Interest: 'None declared'.

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