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Root Canal Cleanliness Following Mechanical Retrieval of Gutta Percha and Bioceramic Sealer with and without Laser Irradiation: An In-vitro Study

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ABSTRACT

Introduction: Lasers are a very promising technology that might be employed in endodontic retreatment cases to improve root canal cleanliness after mechanical gutta percha removal. ***Aim of the study:*** To evaluate root canal cleanliness after mechanical removal of gutta percha with and without Er-Yag laser irradiation in terms of gutta percha, sealer remains, and smear layer. ***Materials and methods****:* Thirty-six single-rooted teeth were divided into three groups according to the retreatment approach. Group I: mechanical gutta percha and sealer removal. Group II: mechanical gutta percha and sealer removal, followed by Er-YAG laser and saline irrigation. Group III is the same as Group II, except with sodium hypochlorite (NaOCl) irrigation. Root canal cleanliness was evaluated using a stereomicroscope, whereas the smear layer was evaluated using a scanning electron microscope. ***Results:*** Group I attained the largest remnant percentage, followed by Group II, while Group III had the lowest mean. There was a highly significant difference in the mean remnant percentage between the three groups. Regarding the open dentinal tubules area, there were significant disparities between group III and the other two groups, with no significant difference in the mean percentage of open dentinal tubules between group I and group II. ***Conclusion****:* The application of Er-YAG with NaOCl following mechanical endodontic retreatment showed the least filling material remnants after using the rotary retreatment files. Moreover, it had minimal effect on the patency of dentinal tubules. Finally, none of the used techniques were able to completely clean the canals from remnants.

Keywords: Retreatment; Laser; Bioceramic sealers; Er-YAG; Smear layer.

INTRODUCTION

Root canal treatment enjoys a very high success rate, exceeding 90%. Failing endodontic cases require non-surgical retreatment before resolving to surgical intervention or extraction. Retrieval of the root canal obturating material requires proper manipulation and armamentarium.[1] Good and McCammon[2], in their literature review,

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listed different mechanical, chemical, and thermal tools, materials, and methods for retrieval of root canal fillings and concluded that none of them was able to totally remove all the contents of the root canal. In order to re-instrument, re-disinfect, and re-fill the endodontic space, which are important prerequisites for endodontic success, the filling material must be completely removed from the root canal. Recent studies have evaluated the efficacy of various instrumentation systems and irrigation techniques in order to determine the best chemo-mechanical retreatment strategy. Moreover, the degree of removal is influenced by the type of sealer and the morphology of the root canals, as well as the instrumentation and irrigation methods.

The assessment of the retreatment of bioceramic cement used for root canal filling has received attention recently because of the increased demand for the use of bioceramic sealers. Because of their interaction with phosphates in tissue fluids that generate hydroxyapatite precipitates, these hydraulic types of cement are exceptional. They exhibit a stronger binding to root dentin than resin sealers.

Laser technology is used in root canal retreatment cases to enhance the effectiveness of the usual methods by improving cleaning capacity and aiding in the removal of debris and smear layer within the canals, thereby resulting in an improvement in canal cleaning. The term "laser-activated irrigation" describes the use of lasers to activate irrigant solutions. It has been demonstrated that laser-activated irrigation using erbium lasers is efficient in removing debris and the apical smear layer.[3]

MATERIALS AND METHODS

Sample Size Calculation: A power analysis was designed to have adequate power to apply a statistical test of the null hypothesis that there is no difference between using laser and conventional methods to remove gutta percha and bioceramic sealer. By adopting an alpha level of (0.05), a beta of (0.2) i.e. power=80% and an effect size (f) of (0.554) calculated based on the results of a previous study[4]; the predicted sample size (n) was found to be a total of (36) samples (i.e. 12 samples per group). Sample size calculation was performed using G*Power version 3.1.9.7.

Sample selection: Thirty-six human-extracted single-rooted mandibular premolars were selected and obtained from the Misr International University teeth bank. The teeth were assessed radiographically to exclude the presence of calcifications, pulp stones, fractures, cracks, and internal or external root resorption and to confirm the existence of a single straight canal and full root development.

Samples preparation: Endodontic coronal access cavities were prepared in all teeth, followed by obtaining patency by inserting k-file size 15 in the canal until the tip of the file coincided with the major apical foramen. Working length was determined by measuring the file length at this position and subtracting 1 mm from this length. All canals were instrumented and prepared using Mpro rotary files up to size #25/.06. Master apical file size #30 was reached by further enlargement using manual K-files sizes #30. Irrigation by 2.6% sodium hypochlorite and patency with manual k-file size #10 was carried out between each rotary or manual file.

Irrigation protocol: The irrigation used during preparation was 2.6% sodium hypochlorite (NaOCl), then flushed with 2 ml of saline. To remove the smear layer, the canals were irrigated with 3 ml of 17% EDTA solution, followed by a final flush with saline. Finally, the canals were dried using paper points size #35 between each irrigant to prevent different irrigants interaction.

Root canal obturation: The technique used for obturation was the lateral condensation technique. Obturation in all canals was done using gutta-percha master cone size #30 with taper 0.04 and bioceramic sealer. Following dryness of all canals by paper points, with the use of a plastic injection tip, the bioceramic sealer was injected into the canal. After completing the condensation of all auxiliary cones, the teeth were then radiographed to assess the quality of the obturation. The samples were stored at 37°C in a 100% humidity environment for two weeks for the complete setting of the sealer.

Sample classification and retrieval of root canal filling material: The samples were divided randomly into three groups (l, II, and III) regarding the retreatment method. A randomization list was made for the groups using an Excel sheet generated by specialized software and concealed to the principal investigator.

In group I (n=12), mechanical retrieval of gutta percha and sealer was done using rotary files (protaper retreatment kit). Mechanical retrieval started with the D1 file on the coronal third of the canal #30/.09 with 16 mm, then the D2 file was used to remove the gutta percha from the middle third #25/.08 with 18 mm working part. Finally, D3 was used to reach full working length #20/.07 with a 22 mm working part. Irrigation of the canals was done using two ml of 2.6% NaOCL. The retreatment procedure was considered complete when the rotary file reached the full working length and no remaining filling or debris on the rotating file was visible to the naked eye. After being used in four canals, each rotary file was discarded in accordance with the manufacturer's instructions. In group II (n=12) mechanical retrieval of gutta percha and sealer was done using rotary files (same as group l), then followed by the application of Er-YAG laser and saline irrigation as follows:

Laser irradiation was done using an Er:YAG laser (2,940 nm, Fidelis AT; Fotona, Ljubljana, Slovenia) at 0.3 W, 15 Hz, and 20 mJ per pulse in the very short pulse mode (VSP. Laser irradiation was applied for 20 seconds for each sample, and a stopwatch was used to calculate the time. Irradiation was delivered with a straight X-pulse tip 14-mm-long optic fiber plain tip[5,6] (**Figure 1**).

Figure (1): Photograph showing the used tip (X pulse tip).

The laser was activated after the tip of the optic fiber was placed 3 mm from the working length. Then, the tip was withdrawn gently from the apical to the coronal region with up and down motion without touching the walls.[4] In group III (n=12), mechanical retrieval of gutta percha and sealer was done using rotary files (same as group l), then the canals were flooded with irrigant (NaOCl 2.6%) then activated by Er-YAG laser (same as group II).

Methods of evaluation:

Evaluation of filling material remnants and smear layer:

All the samples were grooved longitudinally at the mesial and distal surfaces using stainless steel discs and sectioned mesiodistally using Isomet linear precision saw. Before reaching the canal lumen, the roots were split into buccal and lingual halves using a chisel, so that debris from cutting would not reach the canal lumen. One half of each split sample was subjected to stereomicroscope examination while the other half was examined under scanning electron microscope for examination of smear layer.

(i) Stereomicroscopic evaluation:

One half of each split sample was subjected to stereo microscopic examination to assess the canal cleanliness 10X magnification. Gutta percha and sealer remnants were identified and measured using ImageJ software, an image analysis software.[7] The Stereo photographs were processed using photographic editing software (Adobe Photoshop 7.0, Adobe Systems Inc., San Jose, California, USA), then the stained area was calculated as % of the total teeth area using ImageJ software (version 1.53a National Institutes of Health, USA).

After that, the areas with remnants (stained in white or orange color) were automatically detected and highlighted with blue color, and then separated from the rest of the image. The stained area in each third was automatically measured in mm2, and then calculated as % of the total third area using the following equation:

$$Remnants\ \% = \frac{Sum.\ of\ blue\ stained\ area\ (mm^2)}{Total\ third\ area\ (mm^2)} x100$$

(ii) Evaluation of smear layer:

The second half of each sample was examined by a scanning electron microscope) (SEM) (Thermo Fisher company) and attached to the EDX unit to assess the surface topography. Surface topography, which encompasses both surface roughness and profile form, is determined by examining variations in contrast between neighboring pixels as the focus position changes during axial scanning. Samples were prepared for SEM examination by critical point dryness then they were fixed on aluminum stubs with standard diameter using a carbon double sticky tape. No sputter coating was needed as we used environmental SEM in back scuttered mode. SEM examination of each sample was operated at an accelerating voltage of 30 kV. The examination of all groups was done at x1500 magnification for each third individually.[8] Representative images of different samples were selected.

The image analysis for SEM Images was processed using Image J software (version 1.53a National Institutes of Health, USA). The entire image area was automatically measured in μm^2, and then the total area of opened dentinal tubules was calculated as % of the total image area using the equation:

$$Open\ dentinal\ tubules\ \% = \frac{Total\ area\ of\ opened\ dentinal\ tubules\ (\mu m2)}{Total\ image\ area\ (\mu m2)} x100$$

Statistical analysis:

Statistical analysis for results was

performed by applying the Kruskal-Wallis test followed by the Mann-Whitney test for

multiple comparisons between different groups and thirds, $P \leq 0.05$ was considered statistically significant (95% significance level), and $p \leq 0.001$ was considered as highly statistically significant (99% significance level). The Shapiro Wilk test was used for testing the normality of data; statistical evaluation was performed using the SPSS statistical package (version 25, IBM Co. USA).

RESULTS

(I)Assessment of the percentage of residual filling material in the root canal:

1. Distribution of remnants along the root canal zones (coronal, middle& apical) among the three tested experimental groups: (Table 1)

Table (1): Mean ±SD and intra-group comparison of remnant percentage in different thirds in the three major groups.

	Coronal	Middle	Apical	P-value*
Group l	86.99±9.33[a]	87±9.36[a]	63.58±29.16[b]	0.963[NS]
Group ll	74.92±25.19[a]	67.27±25.04[a]	39.55±25.79[b]	0.041[S]
Group lll	48.75±24.45[a]	53.67±24.61[a]	51.42±22.91[a]	0.006[S]

- Overall P-value (Kruskal-Wallis test).*
- significant different at P ≤ 0.05.
- NS= Non-significant P < 0.05.

Group I (Mechanical Only): The mean remnant percentage was (86.99±9.33 %) in the coronal third, (87±9.36 %) in the middle third, and (63.58±29.16 %) in the apical third. There was no statistically significant difference in the mean of remnant percentage between the coronal and middle thirds, while there was a significant difference between the apical third and the other two- thirds.

Group II (Mechanical + ER +Saline): The mean remanent percentage was (74.92±25.19 %) in the coronal third, (67.27±25.04 %) in the middle third, and (39.55±25.79 %) in the apical third. There was no statistically significant difference in the mean of remnant percentage between the coronal and middle thirds, while there was a significant difference between the apical third and the other two-thirds.

Group III (Mechanical + ER+ NaOCl): The mean remanent percentage was (48.75±24.45 %) in the coronal third, (53.67±24.61%) in the middle third, and (51.42±22.91%) in the apical third. The difference between the three thirds was not statistically significant, and according to the Kruskal-Wallis test, the overall P-value for comparison between the three-thirds was not statistically significant (P-value = 0.963).

2. Overall data: (Table 2, and Figure 2)

comparison was statistically highly significant (P-value < 0.001).

(II)Assessment of the percentage of opened dentinal tubules area: (Table 3)

Table (2): Mean ±SD and comparison of the total remnant percentage in different thirds in the three major groups.

	Group l	Group ll	Group lll	P-value*
Total remnants %	81.96±9.09[a]	64.74±17.58[b]	31.46±15.2[c]	< 0.001[HS]

- Overall P-value (Kruskal-Wallis test).*
- significant different at P ≤ 0.05.
- HS= Highly significant at P ≤ 0.001

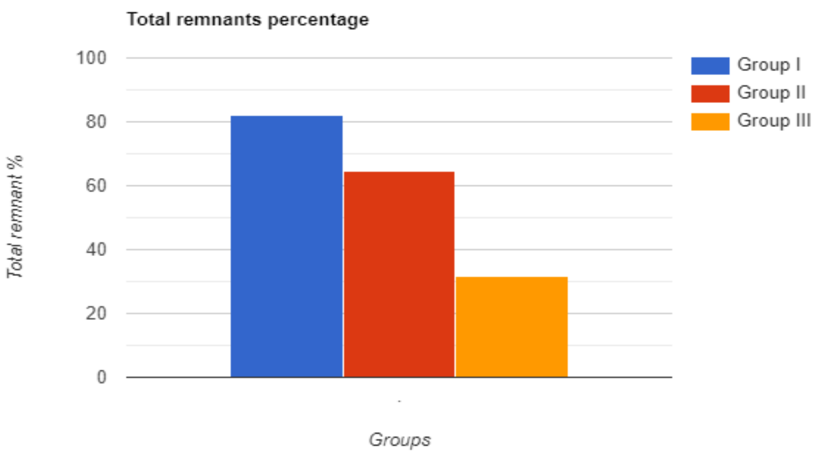

Figure (2): A bar chart representing the mean of total remnant percentage in the three major groups.

The highest mean of remnant percentage was achieved in the Mechanical Only group (81.96±9.09%) **(Figure 3)** then Mechanical + ER+Saline 64.74±17.58% **(Figure 4)** while the lowest mean was achieved in the Mechanical + ER+ NaOCl (31.46±15.2%) **(Figure 5)**. There was a significant difference in the mean remnant percentage between the three groups, and the overall P-value from Kruskal-Wallis test for inter group

Figure (3): Stereomicroscopic image analysis for the remaining filling material in a sample from group I (Mechanical only) (magnification 10x).

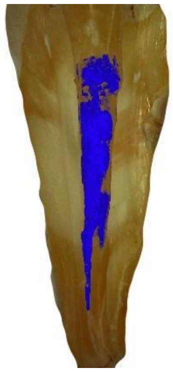

Figure (4): Stereomicroscopic image analysis for the remaining filling material in a sample from group II (Mechanical+ER+saline) (magnification 10x).

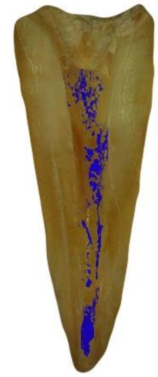

Figure (5): Stereomicroscopic image analysis for the remaining filling material in a sample from group III (Mechanical +ER+NaOCl) (magnification 10x).

For all studied regions (coronal, middle, and apical thirds), the Mechanical+ ER+ NaOCl (Group III) group achieved the largest area of opened dentinal tubules (599µm), followed by the Mechanical+ ER+ Saline group (group II) (290µm), while the Mechanical only group (group I) achieved the smallest area (2.6µm).

Group II (Mechanical + ER + Saline group): The mean of open dentinal tubules area among the three thirds of the canal showed the largest area of open dentinal tubules in the coronal third (159µm). On the other hand, the smallest area of open dentinal tubules was observed in the apical third (53.4µm), while the middle third was intermediate (77.7µm).

Table (3): Mean of open dentinal tubules area in different thirds in the three major groups.

Group	Sample total area (µm)	opened dentinal tubules in Coronal third (µm)	opened dentinal tubules in Middle third (µm)	opened dentinal tubules in Apical third (µm)	Total area of opened dentinal tubules (µm)
Group l	34036.416	0	0	2.6	2.6
Group ll	34036.416	159	77.7	53.4	290
Group lll	34036.416	226	230	143	599

Group I (Mechanical group): The area of open dentinal tubules among the coronal and middle thirds was (Zero) while the mean of open dentinal tubules area in the apical third showed a slight increase (2.6µm).

Group III (Mechanical + ER + NaOCl group): The mean of open dentinal tubules area among the three thirds of the canal showed the largest area of open dentinal tubules in the middle third (230µm). On the other hand, the smallest area of open dentinal tubules was observed in the apical third (143µm), while the coronal third was intermediate (226µm).

Percentage of open dentinal tubules area among the three experimental groups: (Table 4, and Figure 6)

Table (4): Comparison of percentage of open dentinal tubules area among the three major groups.

	Group l	Group ll	Group lll
Total area of open dentinal tubules %	0.007%	0.85%	1.7%

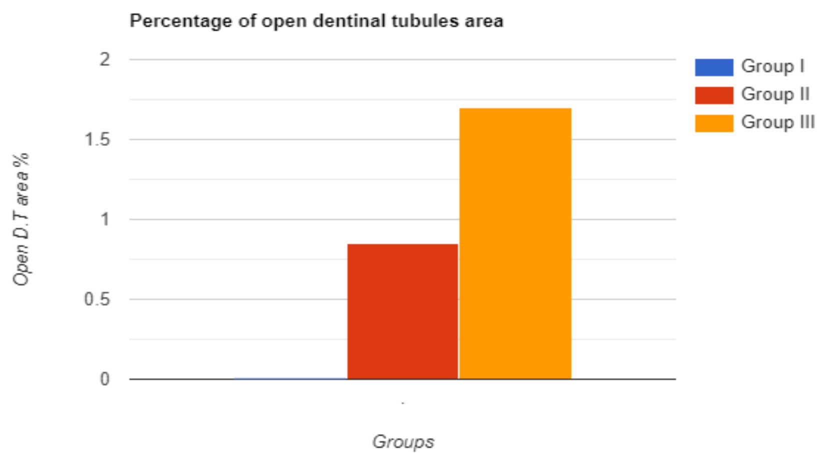

Figure (6): A bar chart representing the percentage of open dentinal tubules area in the three different groups.

The results of the inter-group comparison in the opened dentinal tubules percentage between the three groups showed that the least amount of open dentinal tubules area was seen among samples of group I (0.007%). On contrary the largest percentage was observed among samples of group III (1.7%), while the percentage of group II was intermediate (0.85%).

DISCUSSION

After root canal retreatment, previous filling materials should be eliminated as much as possible, and the WL and patency should be established to ensure periapical healing and good prognosis.[9] Because bioceramic root canal sealer generates hydroxyapatite tags inside dentinal tubules and adheres strongly to root canal walls, it was selected for this investigation.[10] Protaper retreatment files, a popular type of rotary retreatment file, were employed in this investigation. Protaper retreatment files include an active tip that facilitates initial penetration with small apical pressure, making it easier to debride the root canal. The obturation material was removed using three flexible files (D1, D2, and D3).[11] After the majority of the filling material was removed from all samples by mechanical retrieval, samples from groups II and III were subjected to Er-YAG irradiation to remove any remaining root canal remnants and for further disinfecting and cleaning. Thermal effects are primarily responsible for the improved elimination of residues when lasers are used in retreatment processes. The Er-YAG laser exhibits water-mediated photomechanical interaction based on photothermal and photoablation processes. The dentinal surface is likely affected by the photoablation mechanism facilitating the filling remnants separation from the canal walls and irrigation-induced removal.[12] Irradiation with an Er:YAG laser (2,940 nm) at 0.3 W, 15 Hz, and 20 mJ per pulse in the very short pulse mode (VSP)[4] promotes perfect rectangular pulses with variable pulse width produced by the Fotona Variable Square Pulse (VSP) technology, a novel, patented, and unique laser power supply system that regulates the energy and temporal duration of laser pulses. Precision, safety, and convenience are guaranteed with this special technology. Because Fotona's square-shaped laser pulses do not experience the gradual rise and even longer fall in pulse power typical of less sophisticated laser technology platforms, they offer great safety.[13] Laser irradiation was applied for 20 seconds for each sample, producing a negligible temperature rise. Giving a great accessibility, irradiation was delivered with a straight X-pulse tip 14-mm-long optic fiber plain tip 0.6 taper. The laser was activated after the tip of the optic fiber was placed 3 mm from the working length. Then, the tip was withdrawn gently from the apical to the coronal region with up and down motion without touching the walls to avoid laser focus on a single location. The path of the radiation was approximately aligned with the root canal axis in order to prevent dentin ablation during root canal therapy.[14]

In the current investigation, the application of lasers resulted in a considerable reduction in the amount of filling material remaining in the three-thirds of the canal after retreatment with Pro- taper retreatment rotary instruments. Among the studied approaches, a much higher percentage of removal of filling materials was found with the Er:YAG laser than with merely retreating with rotary files. The Er:YAG laser uses water-mediated photomechanical interaction based on photothermal and photoablation mechanisms.[15] Although the photothermal effect may have resulted in the carbonization of filling material,[14] the photoablation mechanism most likely affected the dentinal surface,[16] allowing the filling remnants to detach from the canal walls and be removed via irrigation. These findings were corroborated by the findings of Suk et al.[17] who used Er-YAG (20 mj) with NaOCl irrigant to remove the root canal remnants after retreatment with Pro-taper retreatment files. Moreover, Abduljalil and Kalender's,[18] in their investigation, used the Er,Cr:YSGG laser at various energy levels to remove root canal filling materials and increase root cleanliness; the results showed significant additional removal of root canal filling material. On the other hand, Turkel et al.[19] found that employing laser (PIPS) as a supplemental cleaning strategy compared to standard syringe irrigation is not different in terms of the amount of remaining filling material apically. This may be related to the positioning of the PIPS endodontic fiber tip in the coronal region of the root, which may be too far away to initiate fluid flow toward the apical section of the canal, reducing its debridement efficiency.

Scanning electron microscopy findings in this study showed the efficacy of the Er-YAG laser in opening dentinal tubules in comparison with only using the retreatment files, but with very small areas of opened dentinal tubules. Nasher et al.[20] found positive results for the debridement effect of laser treatment, these results were matching with several studies.[21-23] The authors concluded that lasers have been demonstrated to effectively and safely remove the smear layer and to disinfect the canals based on several researches.[24-26] These previous studies showed large areas of opened dentinal tubules in the laser group when compared to only using files. The difference between their studies and the present investigation may be attributed to using a different laser protocol (Er,Cr:YSGG 2W, 20 Hz, 50 µs) and diode 940 nm (2 W, 50% duty cycle). Moreover, the usage of the resin sealer to obturate the canals facilitated the cleaning process as the high bonding ability of bioceramic sealer used in our study is well known. On the other hand, Ozkan et al.[5] in their study found that using lasers as a supplementary cleaning approach had no effect on the remaining filling material removal after mechanical retrieval, although they used almost the same laser settings where the irrigant was activated by Er-YAG 2940 nm, a 14-mm-long 300-micrometer quartz laser tip delivered a beam at 0.3 W, 15 Hz, and 20 mJ. The only difference was regarding the used mode; in our study, we depended on very short pulse mode (VSP), which promotes perfect rectangular pulses with variable pulse width produced by the Fotona Variable Square Pulse.

In clinical settings, operators are expected to use multiple retreatment systems. To remove root canal filling and allow irrigation to reach the root canal space for cleaning and disinfection, additional methods like manual H-files, ultrasonic irrigation activation and laser activation are necessary. Future studies should incorporate these additional strategies to effectively remove root canal filling.

CONCLUSION

The application of Er-YAG with NaOCl following mechanical endodontic retreatment showed the least filling material remnants. However, this protocol had minimal effect on the patency of dentinal tubules. Finally, none of the used techniques were able to completely clean the canals from remnants.

CONFLICT OF INTEREST

No conflict of interest.

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