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Bulletin of the National Research Centre (2024) 48:69
https://doi.org/10.1186/s42269-024-01224-z

Bulletin of the National Research Centre

RESEARCH

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Evaluation of smear layer and Ca/P ratio in intra-radicular dentine irrigated with different irrigants adjunct with diode laser

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Abstract

Background This study aims to determine the smear layer removal from the root canals using different irrigant solutions with diode laser.

Methods A set of sixty human upper incisor teeth were chosen. Access cavity preparation was done followed by canal instrumentation. Samples were divided into six groups (Group A: sodium hypochlorite NaOCl, Group B: NaOCl + diode laser, group C: NaOCl + EDTA ethylene diamine tetra-acetic acid solution, Group D: NaOCl + EDTA + diode laser, Group E: NaOCl + garlic extract, Group F: NaOCl + garlic extract + diode laser. Examining the smear layer removal at the coronal, middle and apical thirds of the root canals was done by using electron microscope analysis. Using an energy-dispersive X-ray, the calcium/phosphorus Ca/P ratio was measured.

Results Among the examined groups, there were not any significant differences regarding the smear layer removal. The elimination of smear layers at the middle and coronal thirds was greater in all examined groups than in the apical third. EDX analysis revealed no statistically significant variation between the tested groups.

Conclusions Using diode laser with sodium hypochlorite and ethylene diamine tetra-acetic acid irrigant solution was more effective for the elimination of smear layers. Garlic extract could be used as an alternative irrigant solution in debris removal.

Keywords Irrigant solution, Diode laser, Smear layer, Ca/P ratio

Background

Root canal irrigation is important procedure during root canal cleaning and shaping to remove microorganisms, their by-products and the smear layer. Some of the solutions such as phosphoric acid, citric acid, maleic acid, ethylenediaminetetraacetic acid (EDTA), and sodium hypochlorite (NaOCl) are used to remove the smear layer. (Kashikar et al 2023; Azizi Mazreah et al 2023).

In root canal therapy, a sodium hypochlorite solution is frequently utilized as an irrigant due to its antibacterial properties and capacity to dissolve organic tissues. However, excessive doses of NaOCl cause harm to the periapical tissues and are ineffective in eliminating the smear layer. On the other hand, it has been reported that conventional irrigation is ineffective for thoroughly cleaning the intricate architecture of the root canal system. Thus, research is being done to improve root canal therapy in routine endodontic treatment by using sophisticated irrigants and irrigating instruments. These days, the last irrigation pattern is chosen to dissolve the organic and inorganic smear layer residues using NaOCl and the

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chelating agent EDTA (Mukherjee et al. 2023; Nagendrababu et al 2018).

Furthermore, lasers have been used to eliminate the smear layer, including Nd: YAG, CO2 laser, Er: YAG, and diode. By using lasers, the irrigants temperature increased causing reduction in the irrigant surface tension (Nassar et al. 2022).

Recently, herbal extracts such as garlic (Allium sativum) has been introduced as an alternative to NaOCl. It has been proven that garlic has the ability to remove inorganic tissue and has an antibacterial effect, with minimum side effects. Nevertheless, few investigations have demonstrated its capacity to eradicate the root canal debris (Farahat et al. 2023).

The purpose of the present study was to investigate the outcome of different irrigants solutions on removal of smear layer from root canals with diode laser irradiation. The null hypothesis is that the irrigation activation with diode laser in conjunction with garlic extract could be used as an alternative irrigant solution in smear layer removal.

Methods

Samples preparation

Sixty human upper incisor teeth selected for the study were cleaned of any debris and kept in thymol solution 0.1% till use at 4∘C. The access cavity was prepared. The working length was adjusted 2 mm less than the length of the root by a #10 K-file for each root canal, cleaned & shaped to size # F3 with nickel-titanium Protaper instruments (Dentsply Maillefer, Ballaigues, Switzerland) rotary file system (Sherif and Dina 2023).

Samples grouping

The samples (n = 60) were distributed into 6 groups (n = 10):

Group A: NaOCl 5.25%
Group B: NaOCl 5.25% + diode laser
Group C: NaOCl 5.25% + EDTA 17%
Group D: NaOCl 5.25% + EDTA 17% + diode laser
Group E: NaOCl 5.25% + 10 mg/ml garlic solution
Group F: NaOCl 5.25% + 10 mg/ml garlic solution + diode laser

Garlic extract preparation (10 mg/ml)

The garlic extract was prepared at the Faculty of Agriculture, Cairo University, Giza, Egypt. A 100 g of cleaned and peeled garlic cloves had been immersed in 70% ethanol solution for 60 seconds. Afterward, the cloves were placed in a laminar airflow chamber for ethanol evaporation. A sterile mortar and pestle were used to obtain homogenized cloves, and then the mix was filtered through a double layer of fine mesh paper. The concentrated extract was diluted by distilled water to obtain the concentration of 10 mg/ml.

Irrigation protocol

A 27-gauge side ventilated irrigation needle (Vista Dental Products, Appli-Vac, USA) was placed 2 mm less than the working length. The irrigation of the prepared root canals was done using 2 ml solution of 5.25% NaOCl for 60 sec in all groups and then washed out with distilled water.

A 2 ml of EDTA 17% solution was used to irrigate the canals for 1 min in Groups C and D, and 5% garlic extract was used for canals irrigation for 1 min in groups E and F. Any remnants of irrigating solutions were removed after instrumentation from all canals by using for 1 min around 1ml of distilled water, and then drying of all canals was done with sterile paper points (Protaper, Dentsply, Switzerland).

A diode laser (QUANTA C-980nm, DNL0017-0108, 01/2008, Italy) with a power of 2 W, CW mode, was applied with total irradiation 30 s. A 320-μm fiberoptic tip was located inside the root canal (2 mm from the apex). The tip was withdrawn in a circular movement for 10 s in Groups B, D and F. This application was repeated 3 times.

Scanning eelectron microscope SEM analysis

SEM was used to investigate the smear layer in the samples along root canal and measuring smear layer thickness (μm). Grooves in longitudinal directions were created on the external surfaces of the root, buccally and lingually, using slow-speed, double-faced diamond disks. Root samples were then divided equally with a straight chisel into two halves in buccolingual direction. Then, each root half was embedded horizontally in acrylic resin block. Smear layer evaluation was done by using SEM analysis (Thermo, U LTRADRY FEI QUANTA 250 FEG, Czech Republic) under 6000× magnifications. The micrograph figures were then captured at the three thirds of root canal.

Energy-dispersive X-ray EDX analysis:

The change in dentin mineral content of the same samples was investigated by EDX (Inspect S50, Czech Republic) to assess the calcium and phosphorus content and determine the Ca/P ratio.

Results

The values of the mean and standard deviation were computed for every sample in every examined group. Shapiro-Wilk tests and Kolmogorov-Smirnov were used; a nonparametric distribution was shown for SEM data, while for EDX data, a parametric distribution was shown

Table 1 The mean values and the standard deviation (SD) of smear layer scores of each group at three thirds

Variables	Group NaOCl				Group NaOCl + EDTA				Group NaOCl + Garlic			
	Without laser A		With laser B		Without laser C		With laser D		Without laser E		With laser F	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Coronal	3.10	0.88	1.40	0.70	3.00	0.82	1.30	0.48	2.70	0.67	2.60	0.84
Middle	3.10	0.74	1.60	0.70	2.50	0.53	1.40	0.52	3.20	0.63	2.50	0.71
Apical	3.50	0.71	2.50	0.85	2.80	1.14	2.60	0.70	3.70	0.48	3.60	0.52
p-value	0.289ns	0.001*	0.423ns	0.001*	0.002 *	0.001*						

For ($p > 0.05$)—ns nonsignificant; for ($p < 0.05$)—significant*

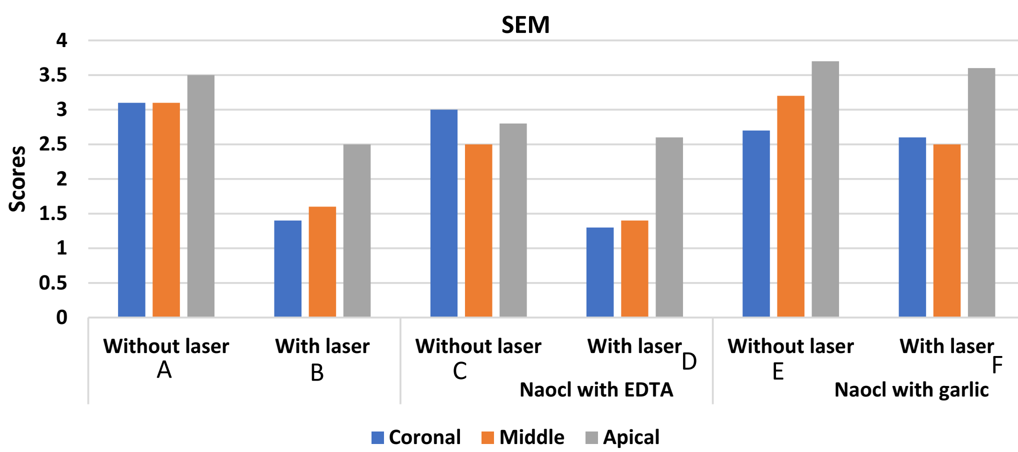

Fig. 1 Bar chart representing smear layer scores of each group at three thirds

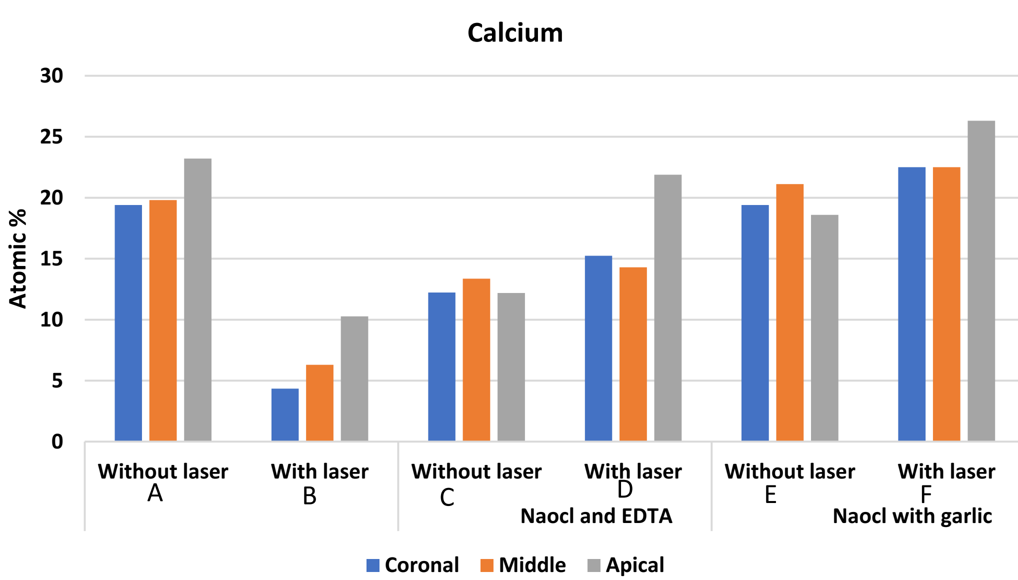

Fig. 2 Bar chart representing calcium (Ca) contents (weight %) of each group at three thirds

Table 1 and Fig. 1 show the findings of the smear layer scores for the three portions: coronal, middle and apical third of each examined group. Figures 2, 3, and 4 display the results of each group's Ca (weight %), P (weight %), and Ca/P ratio (weight ratio) at each third.

Comparison of smear layer scores between the six groups

There was no significant difference between the three parts of the root canals in Group A ($p = 0.289$).

In Group B, there was a statistically significant difference between all groups. There was a statistically significant difference between apical part and each of coronal ($p = 0.009$) and middle part ($p = 0.014$). In Group C, there was no statistically significant difference between all groups. ($p = 0.423$)

In Group D, there was a statistically significant difference across all test groups. However, there was no statistically significant difference between the coronal and middle parts. ($p = 0.564$).

Regarding Group E, the apical part differed from each of the coronal part ($p = 0.008$) and the middle part ($p = 0.025$) significantly.

In Group F, the difference was significant between the apical part and each of coronal and middle parts with, ($p = 0.015$ and $p = 0.009$), respectively, as displayed in Table 1 and Fig 1.

Energy-dispersive X-Ray results of calcium

For Group A, coronal part did not differ significantly from either of the middle ($p = 0.727$) or apical

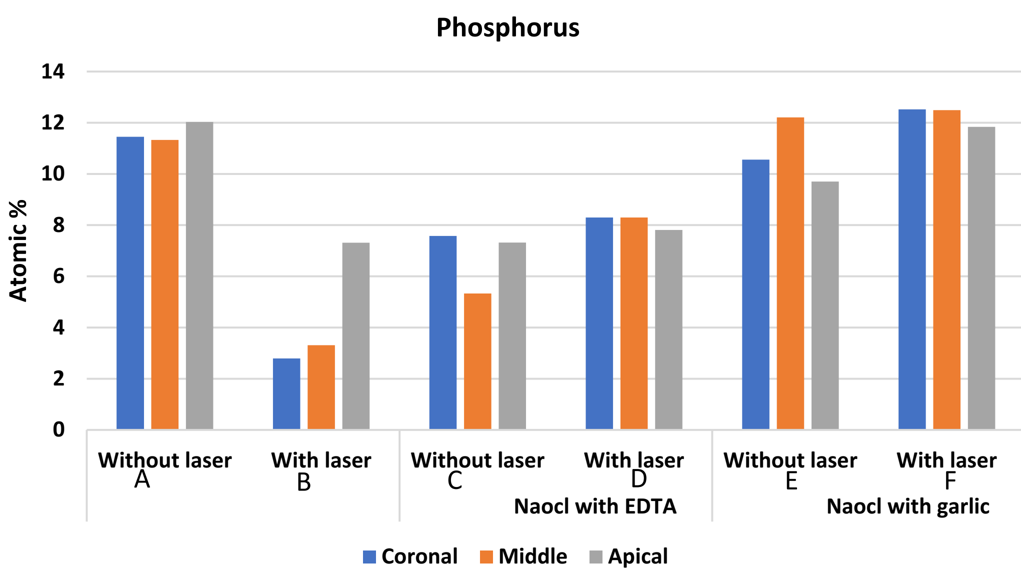

Fig. 3 Bar chart representing phosphorous (P) contents (weight %) of each group at three thirds

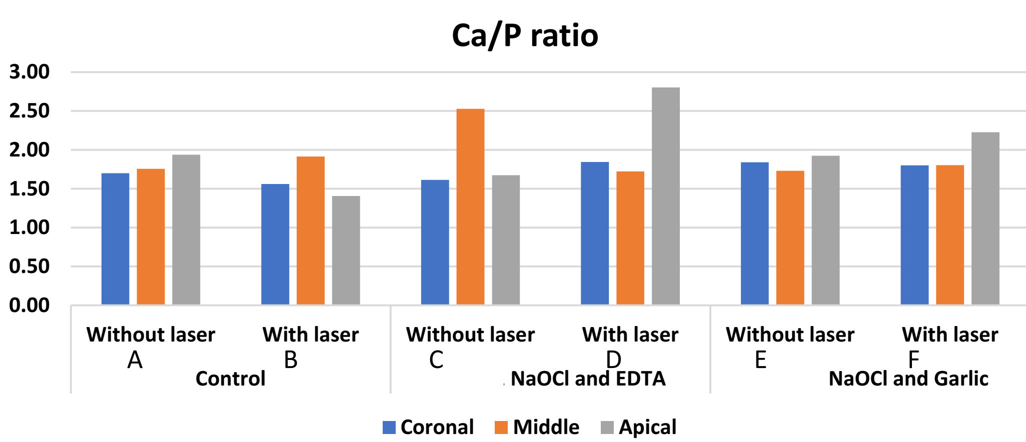

Fig. 4 Bar chart representing calcium /phosphorous (Ca/P) contents (weight ratio%) of each group at three thirds

(p = 0.067) parts. Same as for Group B, the coronal, middle, and apical parts did not differ significantly (p = 0.138).

A significant difference between the middle part and each of the coronal (p = 0.010) and apical parts (p = 0.029) was found in Group C.

In Group D, a statistical difference was seen between the coronal and both the middle (p<0.001) and apical parts (p = 0.008). A statistically significant difference (p = 0.019) between all the parts was found in Group E.

For Group F, there was a significant difference (p<0.001) between the apical and each of the coronal and middle parts (Fig 2)

EDX results of Phosphorus

Group A showed no significant difference between all the groups. (p = 0.676)

A significant difference was found between the Apical and each of the coronal (p = 0.001) and the middle parts (p = 0.027) in Group B.

In Group C, there was a significant difference between the middle part and each of the coronal (p = 0.006) and apical parts. (p = 0.020)

No significant difference was found (p = 0.368), across all parts in Group D.

Also, the coronal part did not differ significantly from any of the middle (p = 0.400) or apical (p = 0.162) parts in Group E.

In Group F, there was a significant difference between the apical and each of the coronal (p = 0.001) and the middle parts (p = 0.017). Fig 3

Energy-Dispersive X-Ray results of Ca/P

There was no significant difference (p = 0.264) in Group A between all the groups.

There was a significant difference in Group B between apical and each of coronal and middle parts where (p = 0.033) and (p = 0.034), respectively.

A significant difference was spotted between middle and each of coronal and apical parts where (p = 0.016) and (p = 0.016), respectively, in Group C.

Also, there was a significant difference between apical and each of coronal and middle parts where (p = 0.013) and (p=0.004), respectively, in Group D.

However, there was no significant difference between coronal and each of middle and apical parts where (p = 0.089) and (p = 0.320), respectively, in Group E.

A significant difference was seen between apical and each of coronal and middle parts where (p = 0.002) and (p < 0.001), respectively, in Group F (Figure 4)

Discussion

The smear layer elimination from the dentinal walls is a prerequisite for successful endodontic treatments. The debris formed during root canal instrumentation is composed of dentin chips, bacteria and their metabolites, and fragments of tissue. This layer covers the dentinal walls of root canals, protecting bacteria within the dentinal tubules. It also hinders the deep penetration of irrigating solutions and root canal sealants, and it stops obturation material from well adaptation to the root canal wall, which could disrupt the apical seal and lower the success rate of endodontic treatment (Sherif and Dina 2023).

Sodium hypochlorite (NaOCl) and EDTA were selected as the standard irrigation procedure in the current study because they are thought to be the most successful irrigation clinical protocols. Because the organic components of the smear layer are best removed by NaOCl, EDTA, a chelating agent, deals with the inorganic portion of the smear layer (İriboz et al. 2015).

Standard irrigation is unsuccessful in the apical third of the root canal, despite being a widely used technique. Because the apical portion of the root is smaller than the other thirds and so limits the movement and effect of the irrigating solutions, it is challenging to eliminate the remaining smear layer there. The irrigant exchange in this location is limited because the usual irrigation approach only distributes the solution slightly beyond the needle tip, failing to displace the air trapped in the apical section. (Qin H et 2023).

As a result, different irrigant materials and activation methods were developed, particularly to address the issue of vapor lock that develops at the apical section. In endodontic therapy, laser-induced agitation has been utilized to decrease bacterial number and modify the surface of the root canal. In order to increase the effectiveness of irrigating solutions at the apical region, irrigants' acoustic and hydrodynamic qualities have been investigated (Lagemann et al. 2014).

Several laser wavelengths have been studied. For practical disinfection and sterilizing treatments, the diode laser (DL) is effective and portable. A thin, flexible fiber that is easily able to fit into curved and narrow channels and reach inaccessible sections of the root canals can be used to do this. The effectiveness of combining root canal preparation with 980 nm laser irradiation to clean canal walls, open dentinal tubules, and decrease apical leakage was proven by Wang et al (2013).

The current study was designed using SEM pictures and EDX analysis of the Ca\P ratio in order to examine the efficacy of DL in eliminating the smear layer in root canals at all root thirds and their influence on the smear layer.

In this study, in all diode laser groups, elevated temperatures accelerate the pace of reaction of the irrigant, hence augmenting its capacity to dissolve organic wastes. This finding is in line with earlier studies that found laser treatment has demonstrated encouraging outcomes in root canal therapy, such as cleaning, disinfection, and the elimination of the smear layer following root canal wall instrumentation (Sherif and Dina 2023).

Moreover, previous research that used a systematic comparison of laser-based activation with different irrigant solutions showed that using laser activation may be useful in smear layer removal (Qin et al. 2023). These results can be attributed to the warming effect of laser radiation on the irrigating solutions that can improve the irrigants' activity by lowering its surface tension, thus facilitating the irrigant wetting to root canal walls (Lagemann et al. 2014). However, the difference in the results between the present study and previous researches would be explained by using different fiber diameters, mode of laser irradiation, exposure time and laser wavelengths (Strefezza et al. 2018).

The apical third of the root canals, in the current study, had a much higher smear layer removal rate than the center. This is because the apical region of the canal has a narrower canal diameter, which causes the laser tip to approximate the canal walls more closely. As a result, there was more noticeable smear layer melting and evaporation. This result was consistent with earlier studies (Paromita and Abiskrita 2018).

Sodium hypochlorite and EDTA irrigants groups showed significant reduction in Ca content compared to the garlic group. As concluded in previous researches, EDTA is used commonly as a chelating agent to remove the smear layer by binding to inorganic Ca content. This alters Ca/P ratio of dentin hydroxyapatite, which led to undesirable dentin de-calcification to 20– 30 µm depth in 5 min. Additionally, NaOCl is widely recommended because of its microbial and organic tissue-dissolving ability (Castagnola et al. 2024 and Balasubramanian et al. 2017).

On the contrary, using laser activation during root canal irrigation with garlic extract, led to a slight temperature rise and release of the allicin from garlic. This helps in necrotic and organic parts degradation of root canals, thus removing the smear layer occluding the dentinal tubules without affecting Ca in dentin (Minan et al. 2022 and Mohammadi et al. 2019).

In this present study, in every group, the cleanliness attained at the apical third was lower. The apical third showed the least quantity of dentinal tubules that had opened, making it challenging to clean using the clinical procedures employed. This finding is consistent with several research on the removal of smear layers, which show that the apical third is the hardest to clean (Ana et al. 2014) .

There was a significant increase in Ca content in all groups with laser irradiation (groups B, D and F). The heat produced by the diode laser could be the cause of this. Garlic releases calcium into the dentin hydroxyapatite when heated. This result was consistent with earlier research (Yahya et al. 2022; Baruwa et al. 2022).

The current study's findings are in favor of using the herbal extracts to remove smear layer from root canals; nevertheless, further clinical evaluation is required to ascertain whether or not herbal products can successfully replace chemical products during root canal therapy.

Additionally, the chemical, physical, and biological properties of garlic extract are yet unknown, more researches are required to confirm its usage as an irrigant solution.

Conclusions

Within the constraints of the current investigation, using diode laser with sodium hypochlorite & ethylene diamine tetra-acetic acid irrigant solution was more effective for the elimination of smear layers. Garlic extract could be used as an alternative irrigant solution in debris removal.

Abbreviations

NaOCL	Sodium hypochlorite
EDTA	Ethylene diamine tetra-acetic acid
Ca	Calcium
Ph	Phosphorus
EDEX	Energy-Dispersive X-Ray Analysis
Nd	YAG laser: Neodymium-doped yttrium aluminum garnet laser
CO_2 laser	Carbon-dioxide laser
Er:YAG laser	Erbium-doped yttrium aluminum garnet laser
SEM	Scanning electrode microscope
ICC	Inra-class correlation coefficient

Acknowledgements

The authors refer deep acknowledgment to all colleagues and professors in National Research Centre.

Author contributions

TS carried out the practical work, collected the data and was a major contributor in writing the manuscript. DS performed the statistical analysis, participated in writing the manuscript. EA participated in writing the manuscript and made final revision. HS carried out final writing and editing, plagiarism step and submission. All authors have read and approved the final manuscript before submission.

Availability of data and materials

Data are available under reasoning demand.

Declarations

Ethics approval and consent to participate

Faculty of Dentistry, Minia University, Minia, Egypt under Committee No: 89, Decision No: 637 in 4-10-2022.

Received: 26 March 2024 Accepted: 4 July 2024
Published online: 15 July 2024

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