EGYPTIAN DENTAL JOURNAL
www.eda-egypt.org
VOL. 69, 2447:2454, JULY, 2023
PRINT ISSN 0070-9484 • ONLINE ISSN 2090-2360
CONSERVATIVE DENTISTRY AND ENDODONTICS
Submit Date : 11-05-2023 • Accept Date : 03-06-2023 • Available online: 10-07-2023 • DOI : 10.21608/EDJ.2023.210355.2546

# SEM EVALUATION OF SEALER PENETRATION INTO DENTINAL TUBULES WITH AND WITHOUT ULTRASONIC IRRIGATION ACTIVATION (AN-IN VITRO STUDY)

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## *ABSTRACT*

**Aim:** The study was conducted to evaluate the effect of ultrasonic irrigation activation on the depth of penetration of bioceramic sealer (CeraSeal) and Epoxy resin sealer (Adseal) into the dentinal tubules with cold lateral compaction obturation technique.

**Materials and Methods:** 56 extracted upper central incisors were divided randomly into four experimental groups (each group, n = 14) based on sealer type and irrigation method. Obturation of teeth was performed using lateral compaction technique. Scanning electron microscope (SEM) was used to evaluate sealer penetration at 1600x magnification.

**Results:** CeraSeal has significantly higher tubular penetration with ultrasonic activation in all sections. There was significant difference between all sections among each group as coronal was significantly the highest while apical was significantly the lowest.

**Conclusion:** According to the limitations of the present study, irrigation activation is an essential step in canal preparation for better smear layer removal that gives deeper sealer penetration inside the dentinal tubules leading to a higher sealing ability of obturation. Bioceramic sealers have higher sealing ability than Epoxy resin sealers.

**KEYWORDS:** Ultrasonic activation, Dentinal tubules penetration, Cold lateral compaction, Bioceramic sealers, Epoxy resin-based sealer.

## INTRODUCTION

Eradication of intraradicular bacteria is the key for root canal treatment success.[1,2] Many authors detected that bacteria and its toxins can penetrate the dentinal tubules approximately 300-500 micrometers ($\mu$m).[3,4] Various methods of irrigation enhance the efficiency of irrigating solution for bacterial elimination from the canal space and walls. Apical third of the root is the most difficult area to be disinfected due to the presence of complexities (recesses and irregularities, lateral canals, ramifications and apical deltas).[5]

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The apical area cleanliness is affected by master file size which affect depth of needle insertion and irrigation protocol that depending on the nature of irrigants and the dynamics of irrigation flow to reach the anatomically complex regions.[6] Irrigation with syringe and needle results a cleaner apical third with large apical preparation size than smaller one, while passive ultrasonic irrigation technique results in similar apical third cleanliness in large and small size apical preparation.[7] Sodium hypochlorite (NaOCl) is the most commonly used irrigating solution for its antimicrobial action and efficiency in soft tissue dissolution .[8] Passive ultrasonic activation increases the potential of sodium hypochlorite to penetrate inside the dentinal tubules for debris and soft tissue elimination.[9,10]

Formation of smear layer is a phenomenon occurs due to instrumentation of dentine, where it consists of bacteria and debris that occlude the dentinal tubules preventing the irrigants and sealers from penetration inside them, the smear layer must be discarded to allow the penetration of irrigant to clean the dentinal tubule followed by the sealer to prevent leakage and reinvasion by the surviving bacteria.[11,12] Solution used for smear layer removal are sodium hypochlorite for organic matter elimination and ethylenediaminetetraacetic acid (EDTA), citric acid, phosphoric acid, and maleic acid (MA) are used for inorganic matter removal.[13, 14]

The smear layer removal allows the sealer penetration into the dentinal tubules which have direct relation with the penetration depth of irrigants where the sealer will fill the space created by the irrigant. Sealer penetration into the dentin walls form a micromechanical locking, chemical bonding, physical barrier, retention improvement, and entombs the residual survival bacteria.[15-17]

The application of lateral condensation technique recommends a thin film layer sealer to increase the ability of tubular penetration. Pseudoplastic behavior is one of the endodontic sealers characteristics that appears during compaction where the shear rate increased resulting in viscosity reduction and flow increase of the sealer that enhance the tubular penetration.[18]

The study was conducted to evaluate the effect of ultrasonic irrigation activation on the depth of penetration of bioceramic sealer (CeraSeal) and Epoxy resin sealer (Adseal) into the dentinal tubules with cold lateral compaction obturation technique.

## MATERIALS AND METHODS:

### Sample size calculation:

Sample size of dentinal tubules penetration was calculated depending on a previous study.[19] According to statistical calculations, the accepted sample size was 14 samples per group minimally, when each subject group response was distributed normally with standard deviation 25.28, the mean difference estimation was 28, when the power was 80% & type I error probability was 0.05.

TABLE (1) Materials tested and their compositions according to manufacturer's data sheets

| Materials | Manufacturer | Composition | Batch |
|---|---|---|---|
| CeraSeal | *Meta Biomed, Chungju, Korea* | Calcium silicates, zirconium oxide and thickening agent | CS18020501 |
| Adseal | *Meta Biomed, Chungju, Korea* | Base: 25%–50% bisphenol A 10%–25% zirconium dioxide NS calcium tungstate NS iron oxide<br>Catalyst: 2.5%–10% N, n-dibenzyl-5-oxanonandiamin-1,9 2.5%–10% amantadine | ADS1608271 |

**Specimen preparation:**

The study was approved by research Ethics committee at faculty of dentistry, (RECO6U/18-2021) and conducted following the principles of declaration of Helsinki. Fifty-six freshly extracted upper central incisors were used in the present study. The teeth were collected from oral surgery department which were extracted due to periodontal diseases.

All teeth were investigated for cracks and resorption under magnification of 2.5X using surgical microscope (Karl Kaps, SOM 62, Germany). The cracked and resorbed teeth were excluded and replaced by other incisors free of cracks and resorption. Radiographic examination was performed from proximal view to ensure the integrity of apical foramen and the presence of single canal. All teeth were decapitated to total length of 15 mm. Patency of the canals were checked using K-file #15 and working length (WL) of 14 mm was established.

Root canal preparation was performed using ProTaper Next system (Dentsply Sirona, York, PA, USA) till size X4 (#40/0.06). Irrigation was done using 2.5 ml of (NaOCl) 2.6% before the insertion of each file used for canal preparation.

A random distribution of specimens was performed into four experimental groups according to type of sealer and irrigation method (each group, n = 14) as follows:

***Group 1:*** CeraSeal and final irrigation using syringe and needle only.

***Group 2:*** CeraSeal and final irrigation using syringe and needle followed by ultrasonic activation (IRR20-21, Satelec, Acteon, France) for 30 sec.

***Group 3:*** Adseal and final irrigation using syringe and needle only.

***Group 4:*** Adseal and final irrigation using syringe and needle followed by ultrasonic activation (IRR20-21, Satelec, Acteon, France) for 30 sec.

Irrigation using 5 ml EDTA 17% (META BIOMED Co., Chungbuk, Korea) and a final flush with 5 mL NaCl 0.9%. All specimens were incubated at 37°C and 100% humidity for 24 hours before obturation to simulate body temperature.

All teeth were dried using sterile paper points. Preparation of sealers was performed following the manufacturer's instructions. A master cone (size 40/0.04 taper) (META BIOMED Co., Chungbuk, Korea) was inserted then WL and Tug-back were checked. The master cone was inserted inside the canal after coating it with sealer till reaching the full WL then lateral compaction technique was done using appropriate size of finger spreader and auxiliary cones of the same size of the spreader.

Confirmation for the quality of obturation was performed using two radiographs (1 labial and 1 proximal). Orifices of all specimens were filled using Scotchbond universal adhesive (3M ESPE Deutschland Neuss, Germany) and resin composite (3M Filtek Z250 XT, Universal, St Paul, USA). All groups were kept in an incubator (100% humidity and 37 °C) for one week to allow complete setting of the sealers.[20]

After finishing the incubation, the specimens were eliminated from the vials and submerged into resin (Technovit 4071, Hereaus Kulzer, Hanau, Germany) horizontally. The mesial and buccal aspects of the tooth were marked then the root was cut into 1 mm thickness slices using a diamond saw under coolant (Leitz, Wetzlar, Germany) at 2 to 3, 5 to 6 and 8 to 9 mm from the root tip. 17% EDTA was used for rinsing the specimens for 15 seconds followed by distilled water rinsing for the same time to remove any smear layer, then stored in a closed container.

Evaluation of sealer penetration was performed using SEM (Quanta FEG 250, Netherland). Each root section was divided into four quadrants (mesial, distal labial, palatal,), and images were captured for each quadrant at 1600x magnification. The sealers

penetrating the dentinal tubules were identified by EDX analysis as the sealers used in the study contain a radiopacifier (zirconium oxide). Each quadrant was evaluated for the highest penetration depth of sealer then statistical analysis was done.

## RESULTS:

### Statistical analysis:

All data were presented as mean &standard deviation. Data were presented in table 2 & graph 5. Statistical analysis was performed with SPSS 16® (Statistical Package for Scientific Studies, IBM Co., New York, USA), Graph pad prism & windows excel.

Shapiro-Wilk test and Kolmogorov-Smirnov test for normality were applied on the given data which revealed that the significance level (P-value) was insignificant as P-value > 0.05 which reject the alternative hypothesis, and the concluded data arises from normal distribution (parametric data) resembling normal Bell curve.

Independent t-test was used to compare between 2 different groups, while One Way ANOVA test was used to compare between 4 groups and 3 sections, followed by Tukey's Post Hoc test for multiple comparisons.

One Way ANOVA test was used to compare different groups, which showed significant difference between them in all sections as $P < 0.05$, followed by Tukey`s Post Hoc test for multiple comparisons which showed significant difference in means with different capital superscript letters as $P < 0.05$, while showed insignificant difference in means with the same capital superscript letters as $P > 0.05$.

### Comparison between different groups

***Coronal section:*** Ceraseal Lateral compaction not activated group (Figure 1) and Ceraseal Lateral compaction Activated group (Figure 2) were significantly the highest, Adseal Lateral compaction not Activated group (Figure 3) was significantly the lowest, while there was insignificant difference between Adseal Lateral compaction Activated group (Figure 4) and all other groups.

***Middle, apical sections and overall:*** Ceraseal Lateral compaction Activated was significantly the highest, while Ceraseal Lateral compaction Not activated group, Adseal Lateral compaction Activated group and Adseal Lateral compaction Not Activated group were significantly the lowest with insignificant difference between them.

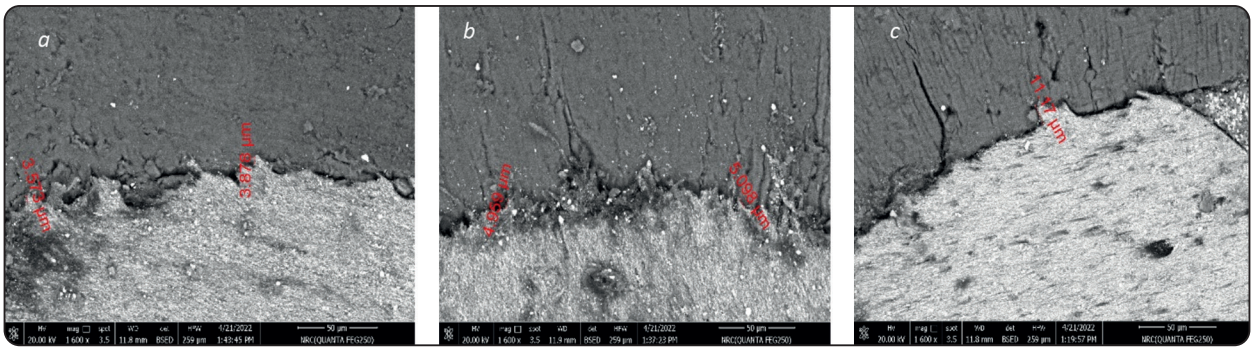


Fig. (1) SEM 1600x magnification, showing the dentinal tubules penetration of CeraSeal/ CLC/ Not Activated ($\mu$m) a) Apical section b) Middle section. c) Coronal section

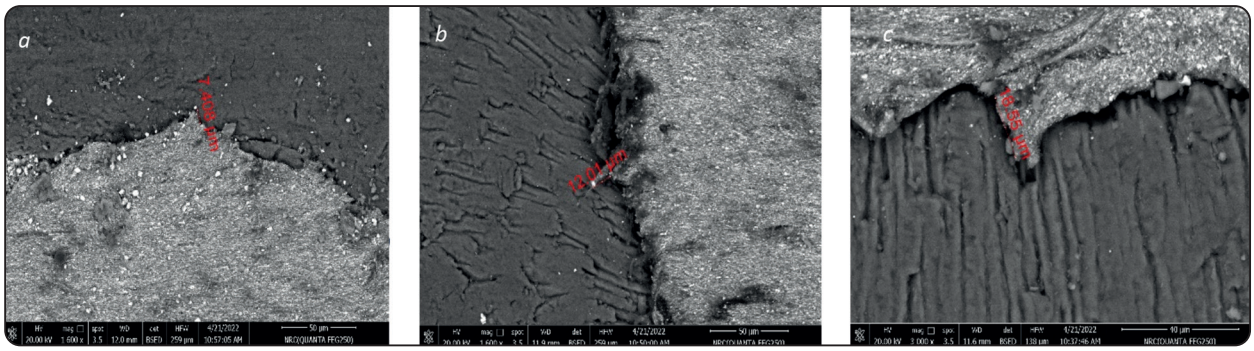


Fig. (2) SEM 1600x magnification, showing the dentinal tubules penetration of CeraSeal/ CLC/ Activated ($\mu$m)Apical section. b) Middle section. c) Coronal section

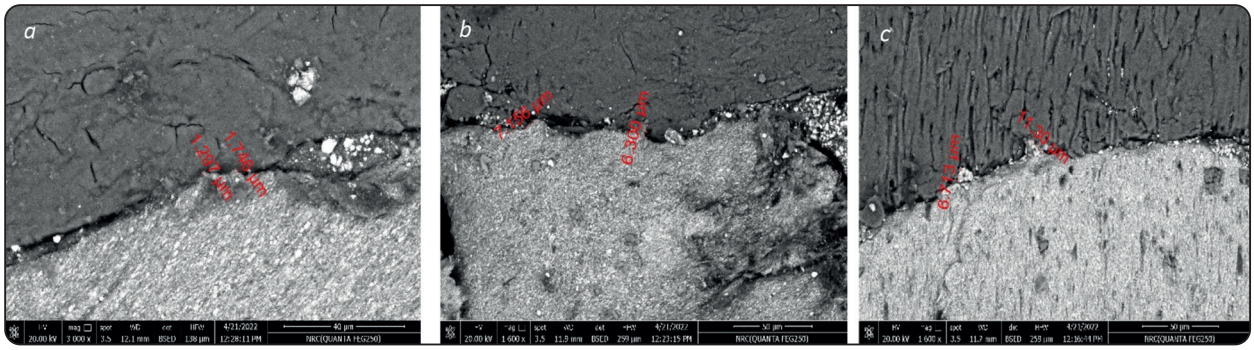


Fig. (3) SEM 1600x magnification, showing the dentinal tubules penetration of Adseal/ CLC/ Not Activated ($\mu$m) a) Apical section. b) Middle section. c) Coronal section

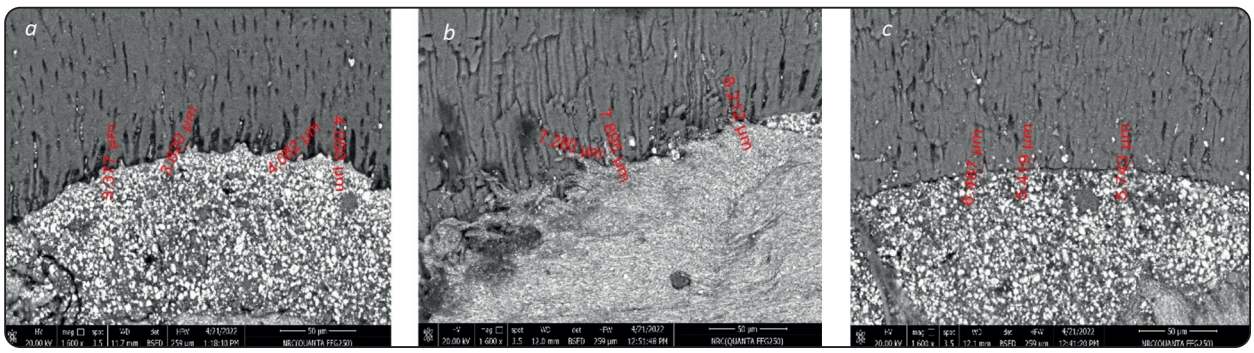


Fig. (4) SEM 1600x magnification, showing the dentinal tubules penetration of Adseal/ CLC/ Activated ($\mu$m) a) Apical section b) Middle section. c) Coronal section

**Comparison between different sections:**

***In Ceraseal Lateral compaction not activated group and in Adseal Lateral compaction Not Activated group:*** there was significant difference between all sections as coronal was significantly the highest while apical was significantly the lowest.

***In Ceraseal Lateral compaction Activated group:*** coronal section was significantly the highest, apical section was significantly the lowest, while middle section revealed insignificant difference with other sections.

***In Adseal Lateral compaction Activated group:*** coronal and middle sections were significantly the highest with insignificant difference between them, while apical was significantly the lowest.

TABLE (2) Mean and standard deviation dentinal tubular penetration in apical, middle coronal and sections in Adseal, Lateral compaction, Not activated ($\mu$m) Adseal, Lateral compaction, Activated ($\mu$m) Adseal, Lateral compaction, Not activated ($\mu$m) Adseal, Lateral compaction, Activated ($\mu$m) groups:

| N=14 | Ceraseal, Lateral compaction, Not activated ($\mu$m) | | Ceraseal, Lateral compaction, Activated ($\mu$m) | | Adseal, Lateral compaction, Not Activated ($\mu$m) | | Adseal, Lateral compaction, Activated ($\mu$m) | | P value |
|---|---|---|---|---|---|---|---|---|---|
| **Groups** | M | SD | M | SD | M | SD | M | SD | |
| **Coronal** | 12.45 $^{Aa}$ | 1.48 | 12.90 $^{Aa}$ | 5.13 | 8.56 $^{Ba}$ | 2.96 | 9.95 $^{ABa}$ | 2.25 | 0.002* |
| **Middle** | 6.96 $^{Ab}$ | 1.94 | 12.66 $^{Bab}$ | 1.38 | 6.97 $^{Ab}$ | 0.57 | 7.97 $^{Aa}$ | 0.57 | <0.0001* |
| **Apical** | 3.03 $^{Ac}$ | 0.64 | 9.65 $^{Bb}$ | 2.44 | 2.88 $^{Ac}$ | 1.57 | 3.79 $^{Ab}$ | 0.32 | <0.0001* |
| **Overall** | 7.48 $^{A}$ | 1.35 | 11.74 $^{B}$ | 2.98 | 6.60 $^{A}$ | 1.47 | 6.77 $^{A}$ | 1.28 | <0.0001* |
| **P value** | <0.0001* | | 0.02* | | <0.0001* | | <0.0001* | | |

***Means with different superscript letters were significantly different (Capital per raw/small per column) as P <0.05***
***Means with the same superscript letters were insignificantly different (Capital per raw/small per column) as P >0.05***

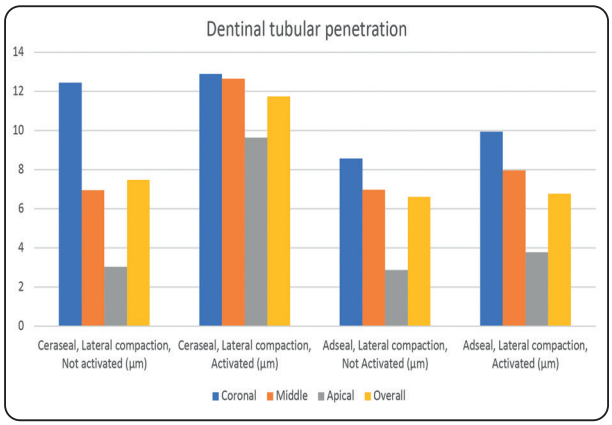


Fig. (5) Bar chart showing dentinal tubular penetration in apical, middle and coronal sections in Adseal, Lateral compaction, Not activated ($\mu$m) Adseal, Lateral compaction, Activated ($\mu$m) Adseal, Lateral compaction, Not activated ($\mu$m) Adseal, Lateral compaction, Activated ($\mu$m) groups

## DISCUSSION

According to the previous studies examining the sealer penetration in dentinal tubules, it was confirmed that the smear layer has a great effect on sealers penetration. Also, the extent of sealer penetration depth has not been adequately investigated. The presence of different types of root canal sealers nowadays with various formulas and components need to be investigated for its capability of tubular penetration.[12]

To increase the obturation sealing quality, activation for the irrigating solutions by ultrasonics has been introduced, showing better results. Ultrasonic activation of irrigation increases the elimination of smear layer leads to dentinal tubules clearance for a better sealer penetration into areas of canal complexities. The specific ultrasonic tips used for activation produce a high-frequency vibrations that promotes acoustic streaming, cavitation and raising of irrigation temperature.[21]

Sealers have an important role in root canal sealing. Prevention of secondary infection is depending on the capability of obturating material to form a hermetic seal with the canal wall to avoid the bacteria from invading the periapical area.[22] Sealer penetration is a parameter that used to determine sealer–dentin interaction[23].

In all groups, it was found that using of 17% EDTA irrigation, leads to removal the smear layer preliminarily or thinning of smear layer thickness. While in group 2 and 4 the ultrasonic activation of Irrigation increases the amount of smear layer

removed. The exerted pressure generated during the lateral compaction of gutta-percha, resulted in entrance of the sealer even into the partially opened dentinal tubules. [24]

The Adseal sealer used in the present study is epoxy resin-based sealer where it has good physical and sealing properties, but the main disadvantage is its hydrophobic nature. While CeraSeal has better physiochemical properties in addition to its hydrophilic nature and antibacterial effect. [25]

The groups obturated with CeraSeal sealer either with or without irrigation activation (group 1 & 2) have significantly the highest scores in tubular penetration results in coronal section, while for the apical and middle thirds, group 2 was the only significant group with highest tubular penetration, on the other hand the other groups have no significant difference between them.

Sealer adaptation to root canal is influenced by many factors as surface energy, cleanliness, surface tension, and the sealer's wettability. [26] the hydrophilicity, wettability and flow of bioceramic sealers is greater than the epoxy resin sealers and this may be the cause for the better penetration of CeraSeal in coronal sections due to the large diameter of dentinal tubules at this section of the teeth.

The comparison between different sections results showed significance between the coronal and apical sections where the sealer penetration was higher in the coronal sections in all examined groups. The explanation of the difference between coronal and apical thirds may be referred to the anatomy complexity of the apical region, the decrease of dentinal tubules number, the diminished diameter of dentinal tubules, and presence of higher sclerotic dentin in the dentinal tubules apically. [27]

Examination of smear layer by SEM in previous studies showed the presence of thicker smear layer at the apical third than the coronal one even with the application of different irrigation activation techniques. This could be the reason for the increased sealers penetration depth in the coronal dentinal tubules. [9]

Among the explanations of sealer penetration difference between different sections is the presence of compressed air in dentin surface during the insertion of gutta percha inside the canal leading to reduction of contact between sealer and dentin as the sealer moves in opposite direction. [27]

## CONCLUSION

According to the limitations of the present study, irrigation activation is an essential step in canal preparation for better smear layer removal that gives deeper sealer penetration inside the dentinal tubules leading to a higher sealing ability of obturation. Bioceramic sealers have higher sealing ability than Epoxy resin sealers.

### Data availability

Datasets related to this article will be available upon request to the corresponding auhor.

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