

Effect of chelating agents on shear bond strength of EpoSeal Plus™ sealer to root canal dentin: *In vitro* study

Prajwal Shetty, Sandya Kini, Nidambur Vasudev Ballal, Nagaraja Upadhaya¹

Departments of Conservative Dentistry and Endodontics and ¹Dental Materials, Manipal College of Dental Sciences, Manipal Academy of Higher Education, Manipal, Karnataka, India

Abstract

Introduction: The aim of the study was to evaluate the effect of 17% ethylenediaminetetraacetic acid (EDTA) and 7% maleic acid (MA) on shear bond strength of EpoSeal Plus sealer to root canal dentin.

Materials and Methods: Twenty-one decoronated maxillary central incisors were longitudinally split into two halves, and each half of the root was further split into three sections, coronal, middle, and apical thirds, respectively, and embedded in dental stone block and abraded with silicon carbide paper to create smear layer. Random division of samples was done into Group 1 ($n = 12$): 0.9% of saline (5 mL/min), Group 2 ($n = 15$): 17% EDTA (5 mL/min), and Group 3 ($n = 15$): 7% MA (5 mL/min). After drying the specimens using paper points, polyethylene tubes were placed on the center of the root canal dentin, and EpoSeal Plus sealer was placed into the tubes, followed by which testing of shear bond strength using a universal testing machine was done. One-way ANOVA and Tukey's honestly significant difference *post hoc* test was done with a significance level ($P = 0.05$).

Results: The results demonstrated significantly more shear bond strength in MA than EDTA in the middle and apical thirds ($P < 0.05$), whereas no significant difference was observed in the coronal third ($P = 0.05$).

Conclusion: The shear bond strength of EpoSeal Plus was found to be the maximum at all thirds of root canal dentin when irrigated with 7% MA.

Keywords: Bond strength, ethylenediaminetetraacetic acid, EpoSeal Plus sealer, irrigation, maleic acid

Address for correspondence: Dr. Sandya Kini, Department of Conservative Dentistry and Endodontics, Manipal College of Dental Sciences, Manipal Academy of Higher Education, Karnataka, India.

E-mail: sandya.kini@manipal.edu

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INTRODUCTION

The ultimate goal of root canal therapy is to ensure complete debridement and three-dimensional seal of the root canal system. Achieving predictable bonding to root canal dentin has been a challenge in endodontics.^[1,2] Good bonding between the root canal sealer and dentin within the canal space is among the most important and

ideal properties because it can influence both leakage and strength of the root canal.^[3] Hence, good adhesion between the filling material and the root canal walls seems beneficial as it helps to eliminate any void that will help leaching of fluids into sealer/dentin interface. Although gutta-percha was used with numerous sealers such as calcium hydroxide-based sealers, glass ionomer-based

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sealers, and eugenol and noneugenol-based sealers, these sealers had disadvantages such as apical leakage and poor bond to the canal dentin.^[4,5]

Numerous *in vitro* studies have found that root canals obturated with resin-based sealers are more resistant to fracture and bacterial leakage than those obturated with the traditional sealers.^[6]

Schroeder introduced that the epoxy resin-based sealers in endodontics and modifications of the original formula are widely used nowadays.^[7] The epoxy resin-based sealers have enhanced physical properties such as longer setting time, better solubility, excellent flow, less polymerization shrinkage, and interfacial adaptation. These sealers are also linked to the formation of covalent bonds in collagen network between epoxy rings and exposed amino group.^[8] Along with superior physical properties, these sealers have also shown no or minimal cytotoxicity^[9-11] and superior marginal adaptation has been observed when compared to bioceramic sealers.^[12] Currently, AH Plus is a well-known resin sealer used in endodontics, which has numerous advantages such as radiopacity, dimensional stability, longer setting time, low solubility, and better flow rate. Owing to its properties, such as low solubility, small expansion, adhesion to dentin, and very good sealing ability, AH Plus sealer is considered as a “gold standard” sealer.^[13]

EpoSeal Plus (Prevest DenPro Limited, Jammu, India) is a recently introduced two-component resin root canal sealer. The base paste consists of epoxy oligomer resin, ethylene glycol salicylate, tricalcium phosphate, bismuth subcarbonate, and zirconium oxide. The catalyst paste consists of poly-aminobenzoate, calcium hydroxide, zirconium oxide, and yellow iron oxide pigment. It was claimed that epoxy resin-calcium phosphate base present in this sealer gives it high adhesive properties along with other advantages such as radiopacity. According to the manufacturer, the sealer is suitable for single-cone technique as well as thermoplasticized gutta-percha technique along with other condensation techniques. It can also be used with different core-filling materials and flows easily into the lateral canals. The sealer can also be easily removed in retreatment cases.^[14]

Irrigation plays an important role in endodontic treatment. Irrigants are used mainly during shaping and cleaning of the root canal system so as to facilitate the removal of microorganisms, tissue debris, and dentinal chips from the root canal space during and after instrumentation through a flushing mechanism. The efficiency of an irrigant in removing the smear layer directly influences the bond of

the sealer to root dentin as it may act as a barrier between the root canal wall and the filling material.^[15,16] After thorough irrigation, the sealer penetrates the dentinal tubules, which prevent the obturating material being used within the canal from being expelled.^[15] Increased sealer penetration has been suggested as a mechanism that entombs any remaining bacteria preventing residual microorganisms from repopulating the canal space.^[16] Therefore, for efficient removal of the smear layer and increase in bonding of sealers, chemical agents such as 17% ethylenediaminetetraacetic acid (EDTA), citric acid, mixture of a tetracycline isomer, an acid, and a detergent, and maleic acid (MA) have been used in the field of endodontics.^[17,18] *In vitro* studies have shown that 17% EDTA is a gold standard and widely used to remove the smear layer. 17% EDTA as a final irrigant has shown to increase the shear bond strength of AH Plus sealer to root canal dentin.^[19]

Final irrigation using 7% MA has proven to be more effective than 17% EDTA in smear layer removal, especially from the apical third of the root canal system, which is an important part for disinfection.^[20] It has also proven to be less cytotoxic when compared to 17% EDTA.^[19] 7% MA also produces increased surface roughness in the walls of the root canal as compared to 17% EDTA. This rough surface produced plays a crucial role in micromechanical bonding of resin-based sealers.^[21]

Till date, no studies have evaluated the effect of EDTA and MA irrigation on the shear bond strength of EpoSeal Plus sealer to root canal dentin. Hence, the aim of the present study was to evaluate the effect of 17% EDTA and 7% MA on the shear bond strength of EpoSeal Plus sealer to root canal dentin.

MATERIALS AND METHODS

Specimen preparation

After acquiring ethical clearance (IEC-719/2017) from the institutional ethical committee, 21 extracted single-rooted human central incisors were selected. The exclusion criteria included teeth with previous history of root canal treatment, curved roots, and open apices. The teeth were evaluated using dental operating microscope ($\times 10$) (ZEISS OPMI, Oberkochen, Germany) to ensure that they were free of any fracture and cracks.^[22] The soft-tissue debris on the surfaces of the roots was cleaned using scaling instruments, and the specimens were stored in 0.9% saline containing 0.2% sodium azide (Millipore Sigma, St. Louis, MO, USA) at 4°C until use. The acquired teeth samples were then decoronated at the cemento-enamel junction

to standardize the root length to 15 mm with high-speed diamond disc (Horico Dental, Berlin, Germany) using water as a coolant. The roots were then divided into a test group ($n = 15$) and a control group ($n = 6$). In the test group, each of the 15 roots was longitudinally split into two halves to obtain 30 halves. Sections of the same root were used for the comparative evaluation of shear bond strength for the two different test groups. The roots in the control group were also longitudinally split into 12 halves. Later, each half of the root in the test and control groups were further split into three sections, coronal third, middle third, and apical third with longitudinal dimension of 5 mm each. Each sample was then embedded in dental stone blocks. By abrading the surface of each sample using a 400-grit silicon carbide paper for 20 s, a smear layer was created over it. Specimens were then treated in the following manner.

Irrigation regimen

Group 1 (control): The coronal ($n = 12$), middle ($n = 12$), and apical thirds ($n = 12$) were irrigated with 5 mL of 2.5% sodium hypochlorite (NaOCl) for 1 min, followed by 0.9% saline (5 mL for 1 min). Later, each specimen was irrigated with 5 mL of distilled water for 1 min.

Group 2 (17% EDTA): The coronal ($n = 15$), middle ($n = 15$), and apical thirds ($n = 15$) were irrigated with 2.5% NaOCl (5 mL for 1 min), followed by 17% EDTA (5 mL for 1 min). Later, each specimen was irrigated with 5 mL of distilled water for 1 min.

Group 3 (7% MA): The coronal ($n = 15$), middle ($n = 15$), and apical thirds ($n = 15$) were irrigated with 2.5% NaOCl (5 mL for 1 min), followed by 7% MA (5 mL for 1 min). Later, each specimen was irrigated with 5 mL of distilled water for 1 min.

The irrigation for all the samples was carried out using disposable syringes with 30G side-ported stainless steel irrigation needles (Appli-Vac, Vista Dental, and Racine, WI, USA).

Bonding procedure

After treatment with the respective irrigation solutions, paper points (Dentsply Sirona Endodontics) were used to render the root canals dry. Polyethylene tubes of 1.8-mm internal diameter and 3-mm height were placed on the center of the surface at the coronal, middle, and apical thirds of the root canal dentin, respectively. EpoSeal Plus (Prevest DenPro) was mixed as per instructions given by the manufacturer; 1 μ L of base paste and 1 μ L of catalyst were dispensed using a micropipette in 1:1 ratio on to a mixing pad and mixed using an agate spatula for 15–20 s

till a creamy homogeneous consistency was obtained and placed into the polyethylene tubes. The samples were then stored at 37°C at 100% humidity for 7 days to help ensure complete set of the sealer.

Evaluation of shear bond strength

The specimens were then attached to a testing jig on the universal testing machine (INSTRON 3366, UK). The assembly was then used to test the shear failure making the use of a chisel blade which was kept parallel to the surface of the root canal dentin using a crosshead speed of 1.0 mm/min. Maximum load at debonding was noted and the shear bond strength was calculated in megapascal.

Statistical analysis

One-way ANOVA and Tukey's honestly significant difference *post hoc* test were used for statistical analysis of the obtained data. Data were analyzed using SPSS software (PASW Statistics 18; SPSS Inc., Chicago, IL, USA). A significance level of $P = 0.05$ was used for all statistical analyses.

Fractographic analysis

The fracture mode was analyzed with the help of a scanning electron microscope (JEOL, USA) at $\times 1000$ magnification to analyze for the mode of failure between dentin–sealer interfaces. The representative fractured specimens from each group were mounted using metallic stubs and were gold sputtered with the help of an ion sputter, after which it was examined using a scanning electron microscope. The modes of failure were classified as: (1) adhesive between the sealer material and the root dentin interfaces, (2) cohesive within the sealer material, and (3) mixed failure.^[23]

RESULTS

The mean and standard deviation values of the shear bond strength for the different groups are represented in Table 1. The results demonstrated no statistically significant difference in the shear bond strength of EpoSeal Plus sealer in the coronal third of the root dentin when treated with 17% EDTA and 7% MA ($P > 0.05$). However, in the middle and apical thirds of the root dentin, 7% MA showed better shear bond strength than the 17% EDTA which was statistically significant ($P < 0.05$). Samples treated with saline showed the least bond strength. While comparing the different thirds of the root dentin, it was found that the apical third showed the highest shear bond strength as compared to the coronal and middle thirds in both the 7% MA and 17% EDTA groups. The analysis of the specimens under scanning electron microscope for the failure mode analysis demonstrated that the bond

failure in the coronal, middle, and apical thirds of the root canal dentin to be mainly cohesive for all the tested groups [Figure 1].

DISCUSSION

The goal of endodontics is to achieve complete debridement and a three-dimensional seal that allows a foreseeable bonding. Although there are various methods to check bond strength such as shear, tensile, and push-out forces, shear bond strength of sealer to root canal

dentin is a proven feasible and reproducible method and therefore was used in the present study.^[3,23,24] It has been proven that shear bond strength gave comparable results with considerably low variation of the bond strength values of sealer to root canal dentin.^[3,23,25] The universal testing machine was used in the present study as it allows evaluating the shear strength of dental materials and adhesives in a controlled *in vitro* experiment. As the test progresses, the fixture provides the linear guidance that is needed to make sure that a pure shear force is applied to the assembly.^[3,23,25]

Table 1: Mean values of shear bond strength of EpoSeal plus sealer to the root canal dentin after treated with different irrigating solutions

Section level	Irrigant	Number of samples	Mean	SD	SE	95% CI
Coronal third	Saline	12	0.715	0.257	0.021	0.634-0.881
	EDTA	15	1.618	0.592	0.039	1.412-1.774
	Maleic acid	15	2.108	1.087	0.072	1.874-2.231
Middle third	Saline	12	0.629	0.022	0.001	0.611-0.654
	EDTA	15	3.472	1.308	0.087	2.897-3.772
	Maleic acid	15	4.705	1.649	0.109	3.863-4.992
Apical third	Saline	12	0.214	0.062	0.005	0.188-0.236
	EDTA	15	4.113	1.235	0.082	3.932-4.362
	Maleic acid	15	5.254	1.805	0.120	4.876-5.543

SE: Standard error, =SD: Standard deviation, EDTA: Ethylenediaminetetraacetic acid, CI: Confidence interval

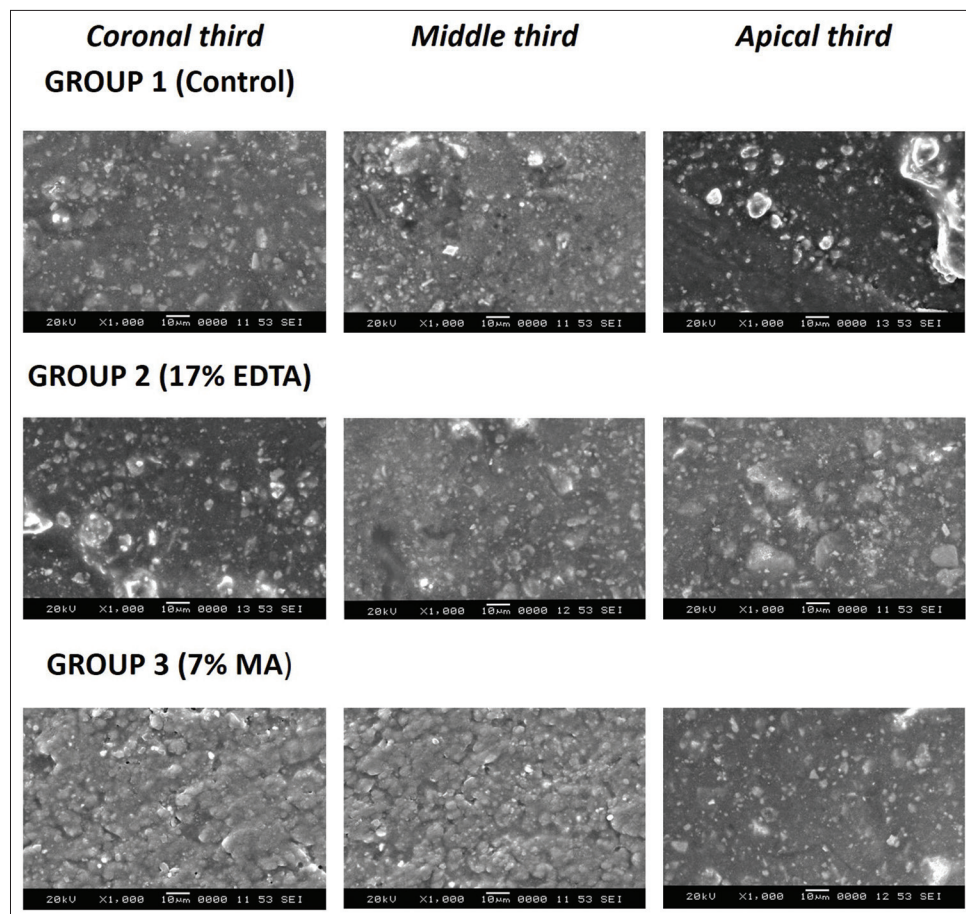


Figure 1: Scanning electron microscopic image showing cohesive mode of failure for the saline (control), ethylenediaminetetraacetic acid, and maleic acid groups at coronal, middle, and apical thirds of root canal dentin, respectively

Sections of the same tooth were used in the present study for the comparative analysis as different teeth may vary in their anatomy and degree of mineralization which could have an influence on the bond strength of the resin sealer.^[23]

Polyethylene tubes were used in this study to prevent the assembly from distortion.^[23,26] Silicone molds have also been used for the same purpose in other studies.^[3,27]

Smear layer is a layer of debris that coats the dentin and clogs the orifices of the dentinal tubules. It holds paramount importance in bonding of the sealer to the dentin surface. It is known to prevent the penetration of sealer into the dentinal tubules.^[15,28] Chelating agents aid in removal of smear layer from the dentinal tubules, which helps in better penetration of the sealer into the dentinal tubules, thus improving the bond strength.^[15]

The present study analyzed the effect of 7% MA and 17% EDTA on the shear bond strength of EpoSeal Plus to root canal dentin. The results of the current study showed that the group treated with 7% MA had a better bond strength than the 17% EDTA group at the apical third of the root dentin. This could be attributed to the fact that MA has better efficacy in smear layer removal at the apical third of the root canal system.^[20] 7% MA has also been proven to bring about an increase in the surface roughness as compared to 17% EDTA.^[21] Resin-based root canal sealers (adhesive) need the presence of surface roughness or surface irregularities for the purpose of micromechanically bonding to root canal dentin (adherend).^[29-31] As per results of this study, the 7% MA group also showed better bond strength than the 17% EDTA group at the middle third of the root canal. This could be attributed to the fact that 7% MA is also more effective than 17% EDTA in smear layer removal at the middle third of the root canal.^[32] In the coronal third of the root canal dentin, the shear bond strength of the MA and EDTA groups was found to be almost similar. This may be because smear layer removing capacity of both 7% MA and 17% EDTA was found to be similar in the coronal aspect of the root canal dentin.^[20]

Shear bond strength values in the apical third of the root canal system were found to be higher than in the coronal and middle thirds for both the test groups. As the density of tubules reduces from the cervical to apical dentin, and if bond strength was correlated to the amount of resin tag formation and depth of resin tags, the bond should decrease in the apical third as the density of the tubules decreases apically in accordance to Mallmann *et al.*^[33] However, this was not the case in the present study. Hence, the increase in bond strength in the apical third could be

related to the increased area of solid intertubular dentin present as compared to the tubule density.

7% MA used in the present study showed superior bond strength as compared to 17% EDTA at all the thirds of the root canal dentin. The reason here could be the fact that 17% EDTA causes complete demineralization of the exposed dentin wall which is not as appropriate for bonding when compared to the structure that is created by a final rinse of 7% MA.^[34] As 7% MA is a weaker acid, it can trigger a gradual mineral gradient in the exposed dentin instead of the entire surface demineralization that is caused with the use of phosphoric acid or EDTA. This is in accordance with studies by Ballal *et al.*^[23] and Shivanna.^[35]

Saline, which was used in the present study as a control group, showed the least shear bond strength values as compared to the 7% MA and 17% EDTA groups at all thirds of the root canal. This may be due to the fact that, as saline does not have any chelating properties the smear layer remained intact which could be responsible for the decreased bond strength values.^[26]

The current study did not use NaOCl for the final rinse in the irrigation protocol as it can hamper the process of polymerization in the resin-based sealer. Sodium hypochlorite breaks down to give sodium chloride and oxygen, which creates oxygen bubbles at the resin–dentin interface which may cause hindrance to infiltration of resin into tubules and intertubular dentin.^[36] However, the use of NaOCl is essential in routine clinical practice for its antimicrobial activity and tissue-dissolving property,^[29] and therefore, treatment of dentin surface with antioxidants has been suggested.^[37] However, in the present study, the aim was to draw a correlation between chelating agents and bond strength of the resin sealer, and hence, EDTA and MA were considered as the final irrigating solutions.^[23]

The disruption between sealer and dentin interfaces becomes less as the resistance to dislodgement of sealer from the root canal dentin increases and failure is most likely expected to be caused within the sealer material itself. In the present study, the mode of failure observed with 7% MA and 17% EDTA was found to be predominantly cohesive in all thirds of the root canal system. The reason could be due to the formation of a complete bond at the resin–dentin interface.^[38]

The main benefit of the study was to be able to assess the ideal chelating agent for use when considering EpoSeal Plus which has been established as 7% MA. The testing of

push-out bond strength could have been a more valuable entity in the above study.

CONCLUSION

The results of the study demonstrate that irrigation with 7% MA showed better shear bond strength of EpoSeal Plus sealer in the middle and apical thirds of the root canal dentin, whereas in the coronal third of the root dentin, shear bond strength of EpoSeal Plus sealer when treated with 17% EDTA and 7% MA was almost similar.

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Conflicts of interest

There are no conflicts of interest.

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