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Scanning Electron Microscopic Evaluation of Marginal Adaptation of Three Endodontic Sealers: An Ex-Vivo Study

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Authors' contributions

This work was carried out in collaboration among all authors. Author NP designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. Author VM Guided during designing and conducting the study. Author VV managed the analyses of the study & literature search. Author JV literature search and reviewing of the manuscript. Authors SJ and MP Reviewing of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Introduction: To avoid the entry of micro-organism or their products, sealer should completely adapt to the root canal wall so that no gaps will be present. AH plus is the most routinely used sealer. EndoRez is methacrylate resin-based self as well as light cured sealer. Endosequence BC RCS is the recently introduced bioceramic based sealer.
Aim: To evaluate the marginal adaptation of Endosequence BS RCS, EndoRez and AH plus as a root canal sealer to root dentin under electron scanning microscope.

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Methods: Total 75 freshly extracted permanent maxillary central incisors with single canal were included in this study. All the teeth were decoronated till twelve millimeter and access cavity was prepared. All the teeth were prepared with rotary protaper till# F3 with intermediate copious irrigation with 3% sodium hypochlorite and 17%EDTA.Teeth were randomly divided into three groups according to sealers and were obturated- Endosequence BC RCS, Ah Plus and EndoRez. Access cavity was restored with glass ionomer type II in all the groups. After seven days, teeth were vertically sectioned and evaluated under electron scanning microscope for marginal adaptation.
Results: Marginal gaps were present in all groups. Maximum number of gaps were present in Ah plus group. Endosequence BC RCS group showed least number of gaps.
Discussion: Marginal adaptation of sealer depends upon the properties like flow, viscosity, presence or absence of smear layer etc. Better adaptation of Endosequence BC RCS to root dentin is due to formation of mineral infiltration zone which results in formation of calcific tags in dentine.
Conclusion: Within the limitation of present study, newly introduced Endosequence BC RCS showed better marginal adaptability to root dentine. Further studies are required to evaluate and corelate these findings with other properties of these sealers.

Keywords: Sealer; AH plus; EndoRez; Endosequence BC RCS; marginal adaptability; electron scanning microscopic.

1. INTRODUCTION

Gutta percha and endodontic sealer are two main constituents of the root canal filling. Endodontic sealer acts as a bridge between the gutta percha and dentinal wall and creates a fluid tight seal by plugging the master gutta percha cone and accessory cones and thus blocking the accessory canals as well [1-2].

Ideally sealer helps inert gutta percha to get holded in the pulp canal space. Ideal properties of sealer are- ease of handling, non-toxic, biocompatible, no shrinkage on setting, effective working time, hydrophilic, antibacterial, ease of retreatment etc [3-5].

Oozing of bacteria and bacterial by-products is the major cause for the failure of root canal treatment. Limkangwalmongkol S et al suggested possible ways through which leakage into the root canal could occur: (a) through the apical foramen through the gaps or voids present between the root filling material and the root canal wall; (b)via the apical foramen through the gaps present between the core material [6].

To avoid the entry of micro-organism or their products, sealer should completely adapt to the root canal wall so that no minute gaps will be present coronally and apically coronal as well as apical plugging off the minute gaps and deeper penetration of sealer into the dentinal tubules which prevents leakage thus preventing the reinfection as entry of micro-organisms and their byproducts is blocked [7-8].

A better penetration, adaptation and adhesion of sealer will have increased tooth surface which is sealed and have antimicrobial action by blocking micro-organisms present inside the dentinal tubules [9-11].

Adaptation and penetration of the sealer depends upon particle size, smear layer removal, dentinal permeability - number and diameter of dentinal tubules, root canal dimension and physical and chemical properties of the sealer [12-14].

Sealers have been revolutionized from traditional zinc-oxide eugenol to recently introduced bioceramic based sealers and depending upon the contents, they are grouped as - Zinc oxide eugenol containing sealer, Iodoform containing sealer, Calcium hydroxide containing sealer, Resin containing sealer, Polyacrylic acid containing sealer, Silicone based sealer, MTA based sealer, Calcium-silicate-Phosphate containing bioceramic sealers, Calcium-phosphate containing sealers.

AH plus is most commonly used sealer in dentistry. It is epoxy resin based and available as two tube system-tube A and tube B. Epoxy resin was invented by P. Castan, a Swiss chemist of de Trey (Zurich, Switzerland) in 1938 [12,15].

Tube A contains Epoxy resins, Calcium tungstate, Zirconium oxide, Silica, Iron oxide pigments whereas contents of Component B are Amines, Calcium tungstate, Zirconium oxide, Silica ,Silicone oil [16].

It has high bond strength to dentine, adequate radiopaque, flow, dimensional stability, low solubility and high resistance [12].

EndoREZ is a hydrophilic, second generation of methacrylate resin based sealers. It is chemical as well as self-cured sealer and its composition is zinc oxide, barium sulfate, resins, and pigments in a matrix of urethane dimethacrylate. Gutta percha as well as resin coated gutta percha can be used with this sealer but latter has the intention of creating a monoblock. It is a hydrophilic sealer [17].

EndoSequence BC root canal sealer is a ready to use, recently developed bioceramic based sealer. It is manufactured by Brasseler USA, Savannah, GA, USA and contains Zirconium oxide, calcium silicates, calcium phosphate, calcium hydroxide, filler, and thickening agents [18]. It is hydrophilic in nature and has high alkaline pH which makes it antimicrobial [19-21] . It has a very fine particle size (less than 2 μ) that enables it to be delivered with a 0.012 capillary tip [22].

There are many methods to evaluate various properties of the sealer. Microscopic examination is the best method for assessing the sealer adaptation to the root canal walls. Stereomicroscopy, scanning electron microscopy, transmission electron microscopy, and confocal laser scanning microscopy (CLSM) etc. are the various types of microscope used for the evaluation but the images produced by Scanning electron microscopy (SEM)are of high resolution and can be used for studying the surface interference between the sealer and dentinal tubules [23].

The benefit of using SEM for evaluation is that the evaluation at the submicron level is achievable at required magnification and a final evaluation can be done by preserving microphotographs [24].

Thus to evaluate the hypothesis that no gaps or voids are present between sealers- AH Plus, Endosequence BC RCS and EndoRez and root canal walls under scanning electron microscope this study was undertaken.

2. MATERIALS AND METHODS

The present study was conducted in the Department of Pedodontics and Preventive Dentistry, Rural Dental College, Pravara Institute of Medical Science (Deemed to be University) Loni. Total seventy -five freshly extracted human permanent maxillary single canaled, single rooted teeth which were indicated for extraction were used for this study.

Permanent maxillary single rooted and single canaled teeth which were free of any developmental defects- anatomical and morphological defects, free of root caries, absence of root surface caries and resorption- external or internal were included in the study.

Root surface debris were cleaned with ultra-sonic scaler and then teeth were placed in 3% sodium hypochlorite solution for two hours for disinfecting them and then autoclaved. After autoclaving teeth were stored in saline till further study. All the teeth were decoronated twelve millimeter (mm) from the apex with a diamond disc using a water coolant and access cavity was made with a round diamond bur # 2(Mani, Inc. Pvt. Ltd) A K-file #10 (Dentsply Meillefer, OK, USA) was introduced into the root canal to the length until its tip was visible at the tooth apex. In order to attain the working length during the root canal preparation, this length was reduced by 1 mm. After preparing glide path with hand instrumentation K-files # 15,20 (Dentsply, Meillefer, OK, USA) and Ethylenediamine tetra acetic acid (EDTA) gel (Premier Dental R C Prep), teeth were then prepared using crown-down technique with Rotary Pro taper files (Dentsply, Meillefer USA) to F3 till the working length of eleven millimeter. During filing copious irrigation with 3% sodium hypochlorite and 17% EDTA (Vista Dental Products) solutions was carried out alternatively. Final irrigation with normal saline was done and canals were dried with sterile absorbent paper point. Based on the sealers, these prepared samples were divided randomly into three groups which were of 25 teeth each.

Group I—Bioceramic Sealer [EndoSequence BC (Brasseler, Savannah, GA, USA)] A syringe tip was used to place the premixed bioceramic sealer up to 2/3rd of the root canal. After dipping the gutta-percha cone in the sealer, it was inserted in the root canal with slow up and down motion until it reaches the full working length. The cone was later seared off at the level of the orifices. Canlas were not completely dried.

Group II—Resin-based Sealer [EndoREZ (Ultradent Products. Inc)] Prior to the placement of sealer, the canal has to be moist and not

dried. A skin syringe with a mixing tip at the back is used to express the EndoREZ sealer. Gutta-percha point smeared with sealer was inserted up to the working length. The gutta-percha cone was then seared off at the orifice level.

Group III - Ah Plus sealer- Equal amount of Paste A and Paste B were mixed on mixing pad and then introduced in the canals. Gutta-percha point smeared with sealer was inserted up to the working length. The gutta-percha cone was then seared off at the orifice level.

Access cavity of all teeth were sealed with glass ionomer cement [restorative type (Type 2) (GC Gold Label-GC Fuji Japan]. The samples were stored at 100 % humidity and at 37° C in incubator for one week to allow setting of the sealers. After one week, teeth were sectioned vertically and observed under SEM for evaluation of the marginal gap at root dentin and sealer interface at apical third at 1000x.

3. RESULTS AND DISCUSSION

3.1. Results

The scanning electron microscopic analysis revealed almost all the samples showed presence of gaps between sealer and root dentine. Statistically significantly lesser number of interfacial gaps were present in the group I-obturated with Endosequence BC RCS, followed by group II-obturated with EndoRez. Group III-AH plus sealer was used showed maximum number of interfacial gap. The SEM examination of root-end-filled teeth showed a mean value of marginal gap of 0.9 in Group I whereas 2mean value of Group II and Group III were 1.85 and 2.35 respectively. The marginal gap between dentinal walls and filling material is less in Group I showing statistically significant difference when compared with Group II AND Group III. Significant P - value <0.05.

3.2 Discussion

Gutta percha is a inert material which most commonly used as filling material in root canal but does not provide three dimensional seal or mono-block effect on its own, so an flowable material in the form of sealer which seals inaccessible areas resulting in better treatment success [25].

Root canal sealers acts like lubricant for core material, improves the radio-opacity of core material, its antibacterial property and adaptation to the root canal wall reduces the chances of reinfection improving the outcome.

Depending upon the contents, sealers are grouped as - Zinc oxide eugenol containing sealer, Iodoform containing sealer, Calcium hydroxide containing sealer, Resin containing sealer, Polyacrylic acid containing sealer, Silicone based sealer, MTA based sealer, Calcium-silicate-Phosphate containing bioceramic sealers, Calcium-phosphate containing sealers.

In present study superior marginal adaptability was observed in Endosequence BC RCS followed by AH Plus and EndoRez. Similar results were found by Shinde et al in their study [26].

But when Zhang W el al compared another bioceramic sealer (iRoot) with AH Plus, they found no difference between marginal adaptation of both sealers and the root canal dentin however iRoot sealer showed superior adaptation with gutta percha cone when compared to AH plus [27].

Observations made by Souza SF et al and Ersahan S et al in their respective studies showed AH plus had shown better marginal adaptation than EndoRez and Resilon/ Epiphenay whereas in our study AH plus showed better marginal adaptation than EndoRez [28-29].

Bioceramic sealers can by of three types depending upon their major component- Calcium silicate based sealer, Mineral Trioxide Aggregate based sealer and Calcium Phosphate based sealer. Endosequence BC RCS and iRoot SP are calcium silicate based sealer [18].

The exact mechanism by which bioceramic sealer adapt to root dentin not known but various authors have postulated the following processes by which bioceramic sealer can adapt to root dentine-

1. Mechanical interlocking of sealer with the dentinal tubules by penetration of sealer component particle in the tubules by tubular diffusion [27].

2. ingress of the minerals into the intertubular dentin causes denaturing of the collagen fibers due to high alkaline pH of newly formed mineral infiltration zone [30-31].
3. Hydroxyapatite formed along the mineral infiltration zone due the interaction of phosphate ion with calcium silicate hydrogel and calcium hydroxide formed due to moisture present in the dentin [21].

Factors like physical and chemical properties of sealer, dentin permeability, filling technique and smear layer removal have influence on adaptability of sealer to dentin [18].

Endosequence BC RCS and EndoRez are premixed sealers they have advantage like homogenous consistency and their tips help in sufficient amount of sealer which has been carried till the apical end sealing the accessory canals also; whereas this may be compromised in manually manipulated AH plus sealer [32].

Smear layer contains organic substances trapped within inorganic dentine and it is generated during instrumentation in the canal. It was observed that smear layer contains organic & inorganic dentine along with fragments or parts of odontoblastic process, micro-organism & necrotic materials also [33].

Removal of smear layer improves sealing ability, creates better sealer adaptation to root dentin and thus lowering the chances of bacterial entry. Smear layer is removed by copious irrigation with numerous demineralizing agents [34].

The calcium silicate-based sealers release calcium hydroxide on hydration and can lead to the formation of calcium phosphate or calcium carbonate on the interface [35].

They bond to the root dentin by a biochemical process known as bio-mineralization by forming a crystalline bond [36].

The use of EDTA has been shown to affect the mineral infiltration zone which is responsible for the retention of the calcium silicate sealers to the radicular dentin.

Lee et al. reported that 17% EDTA showed unfavorable effects on hydration of calcium silicate-based sealer [37].

Yan et al. showed that 17% EDTA decreased the bond strength of calcium silicate-based sealers to the root canal wall [38].

Oxygen left behind from sodium hypochlorite irrigation inhibits polymerization in EndoRez sealer, so it is advised to thoroughly flush with EDTA and saline as final irrigation [17].

Keeping in view above points, in our study we used sterile saline as final irrigant in all three groups.

Shokouhinejad et al. showed that the smear layer did not affect the bond strength of an epoxy resin-based (AH Plus) and a calcium silicate-based sealer (EndoSequence BC) [39].

EndoSequence BC Sealer has superior flow, viscosity and thickness of the film as compare to AH plus sealer this may be one of the result for superior marginal adaptation of Endosequence BC RCS in our study [18,40].

Viscosity of the sealers is indirectly proportional to the penetration, higher the viscosity, lower the penetration which also depends on the composition of the sealer.

All specimens were vertically cut using diamond disc under continuous water flow. This may result in formation of smear layer. This smear layer was removed by cleaning the specimens in bath containing 17%EDTA and 3% NaOCl. This cleaning may have effect on sealer adaptation

Table 1. The mean ± SDs (μm) of gaps between the experimental sealers and dentinal walls

Sr No	Material Used	Minimum Value	Maximum Value	Mean Value	Standard Deviation
1	Endosequence BC RCS(n=25)	0.7	1.1	0.9	1.48
2	ENDOREZ (n=25)	1.5	2.2	1.85	0.49
3	AH PLUS(n=25)	1.9	2.8	2.35	0.64

in the tubules towards the cutting area and the dimension of tubule opening, but this will have same effect in all three groups.

In present study and given environment, Endosequence BC RCS has shown better marginal adaptation. This may be due to formation of mineral infiltration zone which nothing but formation of tag like structures by calcium silicate at calcium silicate and dentin interference. The calcium ion present in this zone reacts with the carbon dioxide in the tissue and forms calcite crystals. These crystals result in reduction in gaps and leakage and better adaptation [31,41-42].

4. CONCLUSION

Within the limitations of the study, Bioceramic based root canal sealer - Endosequence BC RCS showed better marginal adaptation and less number of marginal cracks & voids as compare to resin based sealers- EndoRez & AH plus. Further studies are required to co-relate the marginal adaptation property of these sealers to dye leakage, bacterial infiltration, fracture resistance etc.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. there is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

CONSENT

It is not applicable.

ETHICAL APPROVAL

Ethical clearance for the study was obtained from The Institutional Ethical Review Board And Research Committee at Rural Dental College And Hospital, Loni**.**

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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