

Comparative evaluation of the marginal adaptation of three different root canal sealers to root dentin using scanning electron microscope - An in-vitro study.

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Abstract

Background: The endodontic treatment is completed by the three- dimensional filling of the root canal system, which can be achieved by using an inert, dimensionally stable, and bio logically compatible material combined with a suitable sealer cement which should be bio compatible and cause minimal irritation. Sealer fills the gaps between gutta -percha cones and the root canal wall and voids between individual gutta- percha cones applied during root canal obturation.

Aim: To determine the penetrability and adaptation of different types of root canal sealers to root dentin.

Methods and materials: Thirty sound mandibular premolar teeth were decoronated and access opening was done. Instrumentation of the canals were done using hand Protaper files. Samples were then randomly divided into three groups (n = 10) based upon the sealer used. Group 1 - Samples were obturated using Bio -C sealer, Angelus, Group 2 – Samples were obturated using MTA Fill apex, Angelus, Group 3– Obturation was done using Endometha sone, Septodont. Later the

samples were vertically sectioned and the marginal gap between the sealer and root dentin interface was evaluated under scanning electron microscope. Statistical analysis was done using One-way ANOVA followed by Tukey's post hoc test.

Results: Bio ceramic sealer showed least marginal gap value. Coronal halves showed superior adaptation compared to apical halves.

Conclusion: Bio ceramic-based sealer showed best marginal adaptation

Keyword: root canal sealer, scanning electron microscope, marginal adaptation

Introduction

Endodontic treatment is accomplished by the three-dimensional filling of the root canal space, which leads to complete obliteration of the dentinal tubules after chemo-mechanical preparation. In above context, the term "filling" refers to completely obstructing the canal system and any anatomical abnormalities with biologically compatible and dimensionally stable inert material (often gutta-percha), together with a suitable sealer cement that should be bio compatible [1]. Total debridement of the root canal space, creation of a fluid-tight seal at the apex, and complete obturation of the root canal are the main goals of endodontic treatment [2].

“Sealers are the cements/ resins/ semiliquid/plastic which are used as binding agents to fill up the gap between root canal and obturating material. It is a mixture that usually hardens by chemical reactions.”

Endodontic sealers that are used to obturate the root canal, creates a 3-D hermetic seal throughout the root canal, along with the root apex, canal abnormalities, and minute inconsistencies between the walls of the dentinal tubules and the obturating material [3]. Sealers can expand into various open spaces of the root canal because to their fluidity, especially those that were not

prepared during the cleaning and shaping procedure. During the obturation process, sealers also act as lubricants.

For a number of reasons, the penetration of sealer cements into the dentinal tubules is contemplated to be a desirable outcome, as it will increase the contact between the obturating material and root canal dentine, enhance the sealing ability, and adaptation of the material may also be improved by mechanical interlocking. Factors that determine the adaptation and penetration of the sealer into the dentinal tubules include size of the particles of the sealer material, removal of smear layer, permeability of the dentinal tubules - number and diameter of dentinal tubules, dimension of the root canal along with the physical and chemical nature of the sealer cement [4].

Variation in physical and chemical properties of sealer cements influence the depth of penetration of sealer into dentinal tubules [5]. One of the major factor that influence the choice of filling material is the capacity of the sealer cement to consistently and effectively penetrate the dentinal tubules. As a result, it is important to evaluate how different types of root canal sealers adapt and penetrate into the dentinal tubules.

Hence, the present study was undertaken to compare and evaluate the marginal adaptation of three different root canal sealers to the dentinal tubules of the root canal using scanning electron microscope.

Materials and methods

Study design and location

The present prospective in-vitro comparative study was carried out in the Department of Paediatric and Preventive Dentistry, Darshan Dental College and Hospital, Udaipur, Rajasthan. This study was approved by the Institutional Ethical Committee (Protocol no. 2020-21/1058)

Source of data

Thirty sound permanent mandibular premolar teeth extracted for orthodontic purpose collected from Department of Oral and Maxillofacial Surgery of Darshan Dental College and Hospital, Rajasthan were used for the study. Premolars with non – calcified single canal were included in the study.

Preparation of samples

Superficial soft tissues debris were scrubbed off with a brush from the samples and they preserved in 10% formalin. Teeth were rinsed with distilled water and air dried. A standardized root length of 12mm was achieved by decoronating the samples at the cemento-enamel junction using a slow speed, water-cooled diamond disc bur. An ISO hand 10 no. K-file was used to assess the working length. Size 10 K-file was used to check the working length by introducing it into the root canal of each tooth up to the point until it became visible at the apex and then subtracting 1mm from the total length. The canals were instrumented with hand Protaper files

using crown-down technique. Continuous recapitulation of canals was done with 15K file along with repeated irrigations with 5.25% sodium hypochlorite to establish a progressive taper along the root canal preparation. Canals of all the samples were instrumented up to F2 hand Protaper file and final irrigation was done with 5.25% NaOCl irrigation followed by 17% EDTA and, 10 mL distilled water to eliminate the smear layer. Finally, the samples were dried using sterile paper points After drying the canals, Protaper GP cones was selected according to master apical file size. The fit of each master point was assessed by radiographs (RVG) to determine whether the point was fully seated up to the working length or not. Thereafter, the samples were divided into three groups based on the sealers used for obturation, composition of which is elaborated in table 1:

1. Group I- Bio-C Sealer (Angelus, Londrina, Brazil)
2. Group II- MTA-Fill apex (Angelus, Londrina, Brazil)
3. Group III- Endometha sone N (Septodont, France)

Table 1: Composition of materials used in the study.

Sealer	Product name	Brand	Composition
Bio ceramic-based sealer	Bio-C	Angelus, Londrina, Brazil	premixed calibrated syringes- composed of zirconium oxide, calcium silicates, calcium phosphate monobasic, calcium hydroxide, and various filling and thickening agents
MTA based sealer	MTA Fill apex	Angelus, Londrina, Brazil	2 paste system: Paste A - Salicylate resin, Bismuth Trioxide and Fumed Silica Paste B - Fumed Silica, Titanium Dioxide, Mineral Trioxide Aggregate (40%) and Base resin
Zinc oxide eugenol-based sealer	Endo metha sone N	Septodont, France	Powder is composed of Hydrocortisone acetate (1.0 g) with Excipients- thymol iodide, barium sulphate, zinc oxide, magnesium stearate and the liquid is composed of Eugenol

Group 1 (Bio-c sealer)

Bio- C sealer (bio ceramic based sealer) available as in a ready to use form was dispensed in a glass slab and gutta percha cone coated with the sealer is inserted into the canal up to the full working length and cold lateral condensation obturation technique was performed.

Group 2 (MTA fill apex)

The sealer was mixed using a self-mixing tip attached to a syringe. Then the sealer was applied to root canal space and corresponding Protaper Gutta-percha point coated with sealer was inserted to the working length and then seared off at the orifice level.

Group 3 (Endo metha sone)

According to the manufacturers' instructions, an appropriate amount of powder and liquid (2:1 Wt ratio) were dispensed onto the glass slab and were mixed with the spatula for 15–20 s until the mix was creamy and homo geneous.

After thorough drying of canals, endo metha sone was applied using lentulospiral, and canals were obturated with corresponding Protaper Gutta-percha points.

To assess the obturation, radiographs (RVG) were taken and access cavities was sealed with a temporary material.

The samples were vertically sectioned using a diamond disc and manually marked at the corona (9-10 mm from apex), and apical (2-3 mm from apex) thirds of each specimen before the scanning electron microscopic (SEM) analysis.

Scanning Electron Microscopic evaluation

Coded samples were mounted on metallic stubs, gold sputtered and viewed under a SEM. To calculate the marginal adaptation, maximum gap width i.e., the maximum distance between obturated material and root

canal dentin at coronal as well as apical halve was measured directly at $\times 2000$ magnification by two independent examiners unaware of the experimental groups to which the samples belonged using Smart-Sem Software.

Statistical analysis

Descriptive statistics was performed using statistical package for social sciences software (SPSS) version 23.0. Shapiro Wilk test was used to check whether the variables are normally distributed or not.

Parametric test was used for inferential statistics as the data was found to be normally distributed. One-way ANOVA followed by Tukey's test for pairwise comparison was used for inferential statistics. Statistical significance level was set at p-value less than 0.05

Results

Comparison of mean marginal gap value at coronal level

Overall significant difference ($p < 0.005$) was seen in the mean marginal gap value between the sealer-dentine interface at coronal level (Table 2) under scanning electron microscope (figure 1, A-C).

Mean marginal gap value between the sealer-dentine interface was seen significantly less in Bio-C (572.11nm) followed by MTA (2644) and Endo metha sone. (13388.9nm)

Comparison of mean marginal gap value at apical level

Overall significant difference ($p < 0.005$) was seen in the mean marginal gap value at apical level (Table 3) using bio-c sealer, MTA fill apex and endo metha sone under scanning electron microscope (figure 2, A-C).

Mean marginal gap value was significantly less in Bio-C sealer (700.5nm) followed by MTA fill apex (4671nm) and endo metha sone (22421nm).

Table 2: Mean and standard deviation values in different groups at coronal level.

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum	P value	
					Lower Bound	Upper Bound				
Coronal	Bio-c sealer	10	572.110	107.57	34.0183	495.155	649.065	432.6	747.1	<0.0001
	MTA fill apex	10	2644.00	2438.928	771.256	899.296	4388.704	780.0	7053.0	
	Endo-metha sone	10	13388.9	4255.088	1345.57	10344.99	16432.80	8146.0	21430.0	

*Level of significance was set at p value less than 0.05

Table 3: Mean and standard deviation values in different groups at apical level.

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum	P value	
					Lower Bound	Upper Bound				
Apical	Bio-c sealer	10	700.580	99.9524	31.6077	629.078	772.082	562.9	855.1	<0.0001
	MTA fill apex	10	4671.000	3650.5295	1154.398	2059.569	7282.431	960.0	9610.0	
	Endo- Metha sone	10	22421.20	11775.9434	3723.880	13997.198	30845.202	11523	51230.0	

*Level of significance was set at p value less than 0.0

Discussion

The cleaning, shaping, and sealing of the canal space is the cornerstone of an effective endodontic therapy. Due to advancement in scientific knowledge, endodontic treatments now provide remarkably predictable outcome [6].

Despite of complete instrumentation, micro-organisms remain in the root canal, which can percolate into periapical tissue and cause re-infection. So, to prevent bacteria and their by product from invading the apex, complete sealing of the root canal dentine is required.

The association of gutta percha and a sealer is the foundation of modern root canal filling techniques, which involve using two materials—the core and the sealer. The root canal sealer serves as a sealing material due to its fluidity, and it is also able to spread into any free spaces, particularly those that were not enlarged during the mechanical root canal preparation.

The core can be either cold or thermoplastic zed [7]. The gutta-percha is of hydrophobic nature and devoid of any adhesive properties, sealers help to adapt the same to the root canal dentin [8].

In our study, to eliminate variations in size, single canal mandibular premolars were chosen because they have approximately comparable buccolingual and mesiodistal dimensions [8].

Also access cavity, preparation samples were also decoronated at the cemento enamel junction and to obtain standardization in this study, the length of the specimens was kept same [9].

The combination of 5.25% NaOCl and 17% EDTA has been used as the final irrigation in our study. This combination allows the sealer to effectively penetrate the dentinal tubules by removing the smear layer from the coronal as well as the apical third [10].

By assessing the extent of microleakage various techniques have been used to evaluate the sealing ability of the root canal sealers to seal the dentinal tubules,

some of which are: dye leakage studies, microbial studies, electro chemical studies, fluid filtration technique, confocal-laser micro scope, radio isotope tracing etc., in our study sealing ability is determined by evaluating the adaptation of sealer cement to the dentinal walls of the root canal system under SEM. In SEM, the defects can be observed at the submicron level and at required magnification. Also, final evaluation can be done by preserving the micro photographs. According to Punithia PG and Shashikala K (2011) The scanning electron microscope has superior field depth, greater magnification, and resolution [11].

In present study, the adaptation and penetrability of three different types of sealers cements i.e. bio-c sealer, MTA fill apex, and endomethacrylate to root canal dentin was evaluated under, high magnification (2000x) scanning electron microscopy (SEM) were assessed and evaluated. The magnification of 2000 X was used because it permits better visualization and identification of the dentin-sealer interface [12].

The results of our study showed BIO-C sealer exhibited least marginal gap value compared to other sealers. These findings were in accordance with the previous studies done by Caceres C and Larrain MR et al. and Wang Y and Lui S et al [13].

Superior adaptation of the sealer is due to the hydrophilic properties. The presence of moisture in the root canal system initiates the setting reaction of sealer and lead to the production of hydroxyapatite, as a result of interaction between the phosphate ion and calcium silicate hydrogel plus calcium hydroxide.

The penetration of sealer cement into dentinal tubules will also generate micromechanical interlocking with root dentine and improves the resistance of the filling material which will favour better retention and

adaptation of the sealer to the dentine thus, enhancing its sealing ability.

MTA based sealer (MTA Fill apex) showed less value for mean marginal adaptation as compared to BIO-C sealer. According to Sarkar et al. [14] in 2009 when the MTA based sealers come in contact with phosphate-containing fluids, the set material releases calcium and hydroxyl ions and forms hydroxyapatite.

The formation of an interface layer by apatite with tag-like structures plays an important role in sealing, as this apatite layer gets deposited within the collagen fibrils and promotes controlled mineral nucleation on dentinal walls.

However, in our study, MTA Fill apex showed tiny tags under SEM, which could be related to the presence of erratic moisture content in the canals. The improper adhesion of the material caused due to poor micro tags formation during setting reaction could be the cause of inferior marginal adaptation of MTA based sealer.

Other sealer evaluated in our study was endomethacrylate, which showed significantly lowest value for marginal adaptation with root dentine as compared to Bio-C and MTA fill apex.

This is due to the fact that setting reaction between eugenol and zinc oxide, which leads to the production of an unstable substance i.e., zinc eugenolate, which gets hydrolysed in the presence of water and cause the release of free eugenol thus, reduces the marginal adaptation of the sealer to root dentine. The results were in accordance with previous studies done by Polineni S et al. [12] and Srinidhi V et al. [15]

However, the results of current study were not in accordance with Hirschberg et al. [16] and d. I. K. Pontoriero et al. [17] in which MTA fill apex and ZOE endodontic sealers, shows similar sealing ability.

The reason for this difference could be because, in our study the teeth samples were sectioned Bucco-lingually during which the gutta-percha may get withdrawn from the root canal thus modifying the results. Also the obturation technique and method of evaluating the sealing ability used in these studies were different.

Discrepancy between the penetration of sealers in apical and coronal region may be attributed to the lesser density and diameter of dentinal tubules present at the apical level, difficulty to remove smear layer from the apical third despite of using various chelating agents which might act as a physical barrier and due to the presence of sclerotic and poorly permeable dentin at apical region.

Conclusion

Within the limitations of the current in-vitro study apical halve shows the highest marginal gap between the sealer-root dentin interface irrespective of the sealer used. Bio-C sealer showed the highest marginal adaptation as compared to MTA fill apex and endometha sone.

However, still there is a shortage of information on bio ceramic sealers. Also, additional in-vivo researches are required to demonstrate the long-term clinical superiority of Bio-C sealer.

Legend Figures

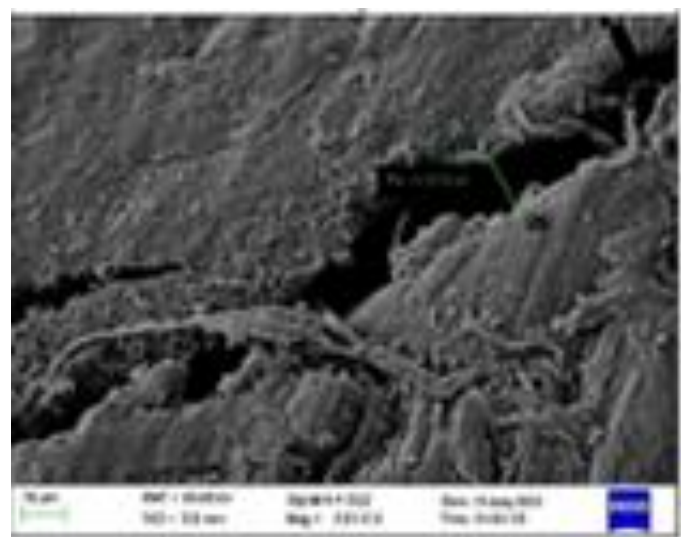
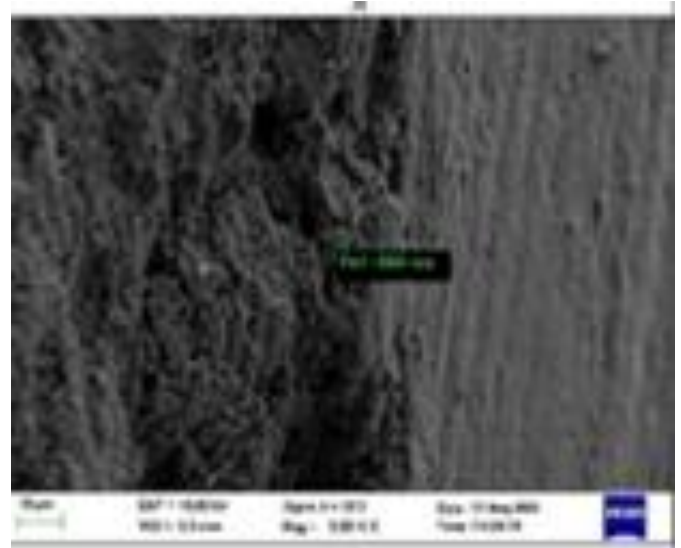
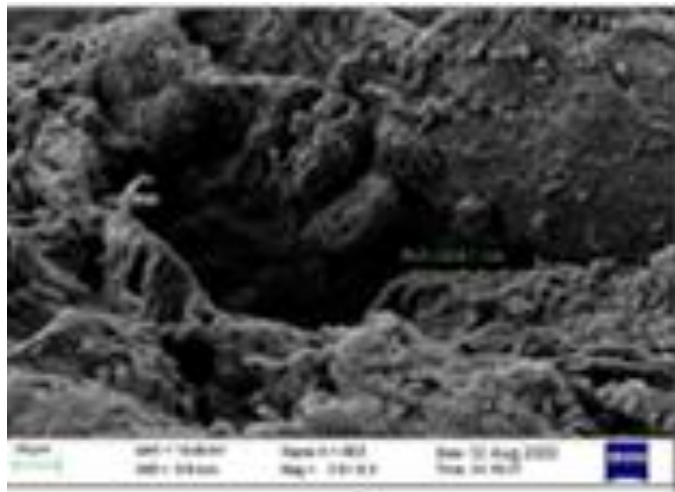
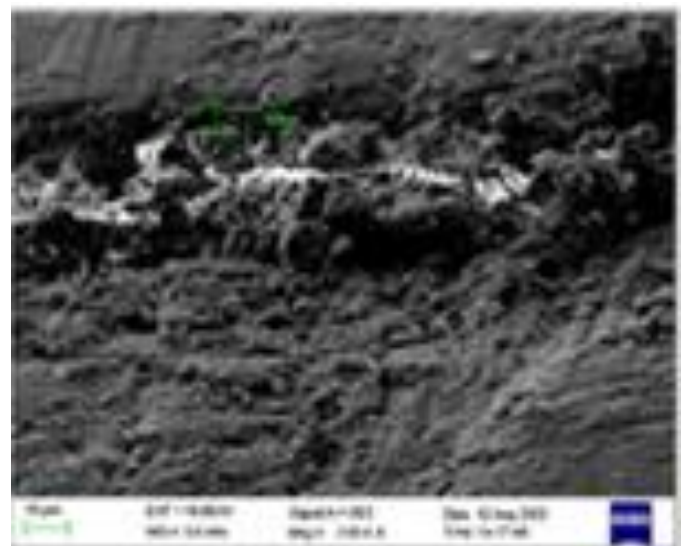


Figure 1: SEM images of samples at coronal level, A. Bio-C sealer, B. MTA fill apex, C. Endo metha sone



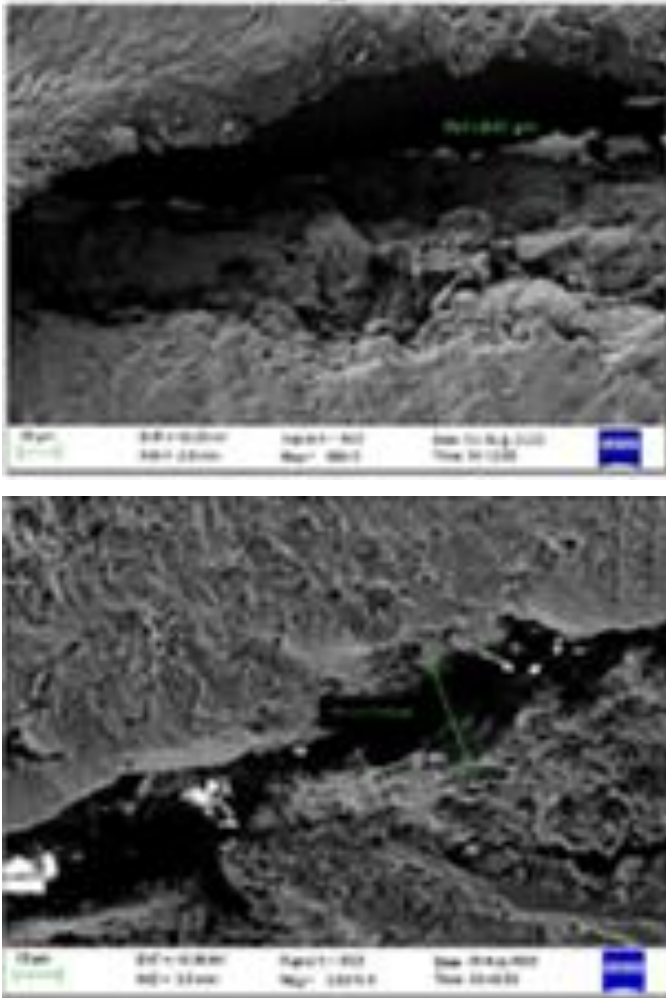


Figure 2: SEM images of samples at apical level, A. Bio-C sealer, B. MTA fill apex, C. Endo metha sone

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