Original Article

An *in vitro* Comparative Evaluation of the Sealing Ability of Five Different Root-end Filling Materials under Confocal Laser Microscopy

Abstract

Aim: The purpose of this *in-vitro* study was to compare and evaluate the best sealing ability of five different root end filling materials i.e. Silver Amalgam, RMGIC, Cermet Cement, MTA Angelus and Biodentine using ConFocal Laser Scanning Microscope. **Methods and Material:** 90 extracted caries free, maxillary incisor teeth were collected and were root canal treated using standardized technique. Apical root resections followed by retrograde cavity preparation were done with ultrasonic retrotip. The teeth were divided into six groups depending upon different root end filling materials (Amalgam, RMGIC, Cermet cement, MTA, Biodentine) and one control group and apical leakage was observed under confocal laser scanning microscope. **Statistical Analysis Used:** The data was analyzed by ANOVA and *Post Hoc* test. **Results:** The mean dye penetration of different groups were Group I (Control Group) 0.00±(0.00) mm, Group II (Silver amalgam) 3.00±(0.00) mm, Group III (RMGIC) 1.84±(0.26) mm, Group IV (Cermet cement) 1.83 (0.25) mm, Group V (MTA) 1.25±(0.12) mm, Group VI (Biodentine) 0.26±(0.21) mm. **Conclusion:** It was concluded that Biodentine exhibits best sealing ability followed by mineral trioxide aggregate, followed by Cermet Cement and RMGIC, whereas silver amalgam exhibited least sealing ability.

Keywords: *Biodentine, Glass ionomer cement, Microleakage, Mineral trioxide aggregate, Rhodamine B, Root End Filling*

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Introduction

Root canal system is very complex in nature due to the presence of lateral canals and ramifications at the apical end of the root that's why it is very difficult to clean it completely. Therefore, root canals cannot always be treated using an orthograde approach.[1]

On failure of primary endodontic therapy, one can choose either to retreat the tooth nonsurgically with an orthograde root filling or surgically with apicoectomy and a retrograde root-end filling. Most ramifications (98%) are present at the most apical part of the root with (93%) lateral canals. Therefore, to achieve the healing of apical bone, resection of most apical 3 mm of the root is advised.[2]

The ideal root-end filling material should adhere to tooth tissue and "seal" the root-end 3-dimensionally. It should inhibit the growth of pathogenic microorganisms, stable under moisture, well tolerated by periradicular tissues with no inflammatory reactions; nontoxic should stimulate the regeneration of normal periodontium.[3,4]

The microleakage of root-end filling materials is of crucial importance. Therefore, the aim and objectives of this study were to evaluate and compare the sealing ability of different root-end filling materials by assessing their apical microleakage from the prepared root ends of the teeth.

Materials and Methods

Materials used in the study

1. High-copper amalgam (DPI, India)
2. Resin-modified glass ionomer cement (GIC) (GC Corporation, Japan)
3. Cermet cement (Promedica Medfil silver, Germany)
4. Mineral trioxide aggregate (MTA) (Angelus, Brazil)
5. Biodentine (Septodont, France)
6. Rhodamine b dye (Ases Chemical Works, India).

Equipment used

1. Confocal laser scanning microscope (IMTECH, Chandigarh, India).

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Methodology

Ninety human maxillary incisors were collected and decoronated at the level of cemento-enamel junction using high-speed cutting disc. The working length was calculated with K files (Mani Inc.). The canal was prepared with hand K files (Mani Inc.) up to size #40K file by standardized technique. Copious irrigation was done with 3% sodium hypochlorite solution. The samples were then obturated with lateral condensation technique. After that, the coronal access cavities were sealed with Type II GIC (Shofuinc, Japan). Then, all the samples were kept in incubator at 37°C for 24 h.

Later on, the apical 3 mm of root ends were resected at transversely 90° along the long axis of the tooth with diamond disc using straight handpiece. Then, 3 mm retrograde cavity was prepared with ultrasonic tip (ED11) in all the samples. The samples were then divided into six groups depending on different root-end filling materials.

- Group I (control group): In this group, all 15 samples were coated with two coats of nail varnish and sticky wax to prevent the penetration of dye solution
- Group II (High-copper amalgam): In this group, retrograde filling was done with high-copper amalgam
- Group III (RMGIC): In this group, RMGIC in a ratio of 3:1 powder liquid was taken according to the manufacturer's instructions and mixed on a paper pad with plastic spatula by folding method for 10–15 s to create a glossy consistency. After the placement of material in cavities, curing was done for 20 s by light-curing unit (WoodPecker Light Emitting Diode (LED) unit) with an intensity of 1200 mV/cm^2
- Group IV (Cermet Cement): Silver reinforced GIC was mixed in a ratio of 7:1 powder: Water according to the manufacturer's instructions on a mixing pad with the plastic spatula for 40–50 s and was filled in all the cavities
- Group V (MTA): MTA was prepared by mixing powder and liquid in a ratio of 3:1 on the mixing slab until a creamy consistency was achieved, which was then retrofilled in the prepared cavities
- Group VI (Biodentine): Biodentine was prepared by adding powder and liquid in a capsule and then mixing it in centrifugation machine (R-4C, Remi Lab instruments, Mumbai, India) for 30 s, at 4300 oscillations per minute and filled in prepared cavities. Then, all the samples prepared were covered in wet gauze pieces and placed in 100% humidity for 24 h in an incubator at 37°C.

The samples were then coated with two layers of nail varnish followed by a layer of approximately 2 mm of sticky wax to the external surface of each root except for the apical section, and all the samples were stored in 100% humidity for 24 h [Figure 1].

A solution of Rhodamine B was prepared, and all the samples were immersed in it for 24 h. Followed by longitudinal sections of each tooth was prepared by using a diamond disc [Figure 2] and observed under confocal laser scanning microscope to check the extent of dye penetration under green light of 546 nm wavelength, Dye gave a Red-Orange fluorescencent appearance.

Figure 1: Samples of (a) Group I (control), (b) Group II (amalgam), (c) Group III (RMGIC), (d) Group IV (Cermet cement), Group V (mineral trioxide aggregate), and Group VI (Biodentine) covered with nail varnish and sticky wax and filled with respective root-end filling materials

Under green light 546 nm wavelength, dye gave a Red-Orange fluorescent appearance [Figure 3]. The depth of dye penetration was calculated in mm using Scale provided by the Zeiss LSM software (Jena, Germany) one-way ANOVA, and independent samples *t*-test was used for the statistical analysis.

Results

The results showed that the Group VI (Biodentine) had the least dye penetration of 0.26± (0.21) mm as compared to other materials. Whereas Group V (MTA) showed the mean dye penetration of 1.25± (0.12) mm followed by Group IV (Cermet cement) 1.83± (0.25) mm and Group III (RMGIC) 1.84± (0.26) mm with Group II (amalgam) showing the highest amount of mean dye penetration of 3.00± (0.00) mm. In Group I (negative control) dye could not penetrate and showed the mean value of 0.00± (0.00) mm [Table 1 and Graph 1].

Multiple comparisons of values showed that there was a highly significant difference observed between different groups ($P < 0.01$), but in case of Group III (RMGIC) and Group IV (Cermet cement), no significant difference was observed.

Discussion

The success of surgical endodontics mainly depends on achieving a fluid tight apical seal. Preparation of apical end

Figure 2: Longitudinal section of all the samples after due penetration (a) Group I (control), (b) Group II (amalgam), (c) Group III (RMGIC), (d) Group IV (Cermet cement), (e) Group V (mineral trioxide aggregate), and (f) Group VI (Biodentine)

with ultrasonic tips allows better access to the resected apical root area with root-end cavity of adequate dimensions. This prepared cavity is filled with root-end filling material to create a apical seal that prevents the seepage of muscles and its by-products to allow the healing of periapical lesions.[5,6]

In the present study, all the root-end filling materials showed some amount of dye penetration except the control group, in which no dye penetration was allowed by sealing the resected apical section of the roots. Hence, from this observation, we can conclude that the recent material Biodentine (Group VI) exhibited least apical microleakage among all the other materials tested. This may be due to the lower setting time, i.e., 12 min and formation of tag-like structures composed of calcium or phosphate-rich crystalline deposits between the tooth and root-end filling materials.[8] This was in accordance to the studies by Pathak,[7] Nanjappa *et al*.,[8] Kokate and Pawar[9] where Biodentine showed better sealing ability as compared to other materials.

MTA also showed promising results with optimum sealing ability because of its hydrophilic nature and expand when allow to set under moisture which tends to fill the gap between the dentin and root-end filling material which is in accordance to studies by Gundam *et al*.,[10] Froughreyhani *et al*.[11] (RMGIC) and Group IV (Cermet cement) which is the enhanced form of conventional GIC exhibited nonsignificant difference in dye penetration because of moisture contamination and gap formation between the tooth and root-end filling material.[12,13]

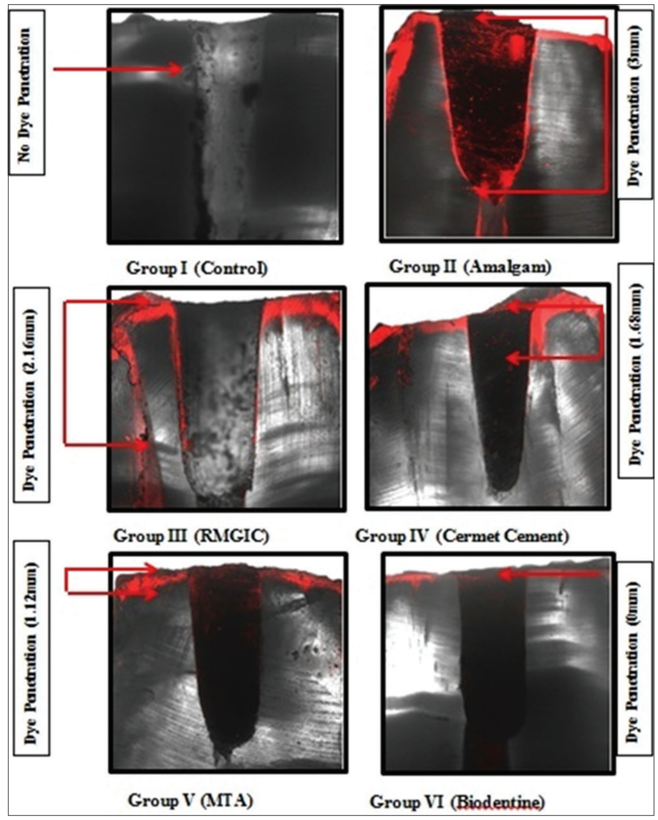


Figure 3: Confocal images of samples

Silver amalgam showed an increase in microleakage due to volume changes during its setting, which produced a continuous gap along the length of the interface. Other disadvantages of amalgam are the scattering of particles into the surrounding tissues, corrosion, and setting properties, which allow dimensional changes and thus increase in microleakage.[14,15]

For the proper evaluation of samples, confocal laser scanning microscope was used because a shape and clear images of samples can be achieved by excluding the light which is not produced by microscope's focal plane. From the samples, images with better contrast and lesser haziness can be obtained than that of traditional microscopes. No specific sectioning procedure is required for the confocal microscope; therefore, the occurrence of artifacts are low in comparison to scanning electron microscope.[16]

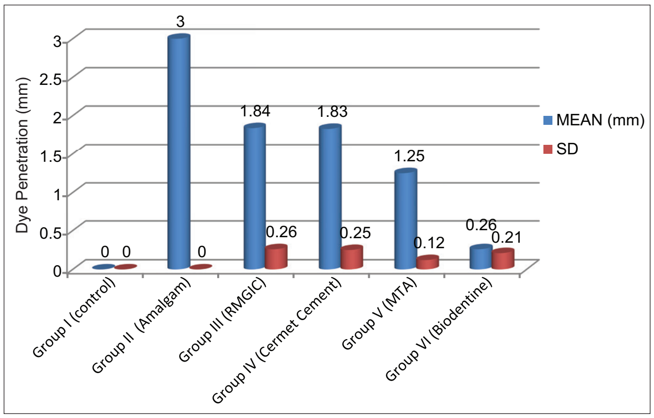
Conclusion

In the current *in vitro* study, sealing ability of different root-end filling material was scanned under confocal laser scanning microscope. Out of all material, Biodentine showed least microleakage and silver amalgam showed maximum. It can be concluded that Biodentine has better sealing ability among the other root-end filling material tested. Furthermore, its bioactive property can be beneficial in initiating the healing of the periapical lesions.

Table 1: Demonstrates the intra group comparison of mean and standard deviation values of dye penetration at the apical end of different root end filling materials tested using One-way ANOVA

Groups	*n*	Mean (mm)±SD	Minimum	Maximum	*P*
Group I (control)	15	0.00±0.00	0.00	0.00	<0.0001
Group II (amalgam)	15	3.00±0.00	3.00	3.00	
Group III (RMGIC)	15	1.84±0.26	1.37	2.25	
Group IV (Cermet Cement)	15	1.83±0.25	1.48	2.41	
Group V (MTA)	15	1.25±0.12	1.12	1.52	
Group VI (Biodentine)	15	0.26±0.21	0.00	0.76	

SD: Standard deviation; MTA: Mineral trioxide aggregate



Graph 1: (Bar Diagram): the intra group comparison of mean and standard deviationvalues of dye penetration at the apical end of different root-end filling materials tested

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Conflicts of interest

There are no conflicts of interest.

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