Original Article

In vitro comparative evaluation of apical leakage using a bioceramic sealer with three different obturating techniques: A glucose leakage model

Tanvi S Agrawal, Shishir Singh, Rajesh S Podar, Gaurav Kulkarni, Anuprita Gadkari, Navin Agarwal

Department of Conservative Dentistry and Endodontics, Terna Dental College, Navi Mumbai, Maharashtra, India

Abstract

Context: Bioceramic sealers have improved sealing ability by forming an interfacial apatite layer that chemically bonds the sealer and radicular dentin thus decrease apical leakage.

Aim: This study aims to evaluate and compare the apical leakage of Cerafill RCS bioceramic sealer and gutta percha when used with three different obturating techniques.

Materials and Methods: Thirty-four extracted single-rooted premolars were decoronated and prepared up to size F3. Then, the specimens were randomly divided into 3 experimental groups (n = 10) cold lateral obturation technique, warm vertical obturation technique, single-cone obturation technique, positive and negative control groups (n = 2), according to the obturation technique used along with a bioceramic sealer. To evaluate apical leakage, all specimens were mounted in a glucose leakage model and assessed at 7 and 14 days using an ultraviolet-visible spectrophotometer.

Statistical Analysis: The results were subjected to ANOVA/Kruskal–Wallis ANOVA; followed by *post hoc* analysis using Bonferroni correction.

Results: Significant differences were found in the cumulative leakage of all the three experimental groups. Significantly higher leakage was found in groups obturated using single-cone obturation technique as compared to warm vertical compaction technique at both 7 and 14 days.

Conclusions: Warm vertical compaction showed a better sealing result than single-cone obturation techniques at all observation periods.

Keywords: Bioceramic sealers; cold lateral compaction; glucose leakage model; single-cone obturation technique; warm vertical compaction

INTRODUCTION

The accomplishment of root canal treatment depends greatly on achieving a three-dimensional seal of the canals. This seal is essential to avert the leakage of periapical exudates and reinfection, while also promoting healing.[1] The property of root canal sealers to prevent microleakage is a critical factor in achieving this apical seal.[2] However, there are various factors that can influence the occurrence of apical leakage, such as the quality of the obturating substances, the obturation techniques used, and the availability of the smear layer, all leading to treatment failure.[3]

A range of substances have been introduced for obturation, but despite this, gutta-percha remains the primary choice.

Address for correspondence:
Dr. Tanvi S Agrawal,
Department of Conservative Dentistry and Endodontics, Terna Dental College, Nerul, Navi Mumbai, Maharashtra, India.
E-mail: agrawaltanvi96@gmail.com

Date of submission : 08.05.2023
Review completed : 05.06.2023
Date of acceptance : 03.11.2023
Published : 13.01.2024

Access this article online	
Quick Response Code:	Website: https://journals.lww.com/jcde
	DOI: 10.4103/jcd.jcd_303_23

How to cite this article: Agrawal TS, Singh S, Podar RS, Kulkarni G, Gadkari A, Agarwal N. *In vitro* comparative evaluation of apical leakage using a bioceramic sealer with three different obturating techniques: A glucose leakage model. J Conserv Dent Endod 2024;27:76-81.

However, gutta-percha lacks complete bonding to the root structure. Bioceramic sealers were introduced to improve the sealing ability by forming an interfacial apatite layer that chemically bonds the sealer and radicular dentin.[4] In this study, a new ready-to-use injectable premixed sealer based on bioceramic technology, Cerafill RCS (PREVEST DenPro, Jammu Kashmir, India) was used in combination with gutta percha. It is an alumina-free calcium silicate-based material that requires water from dentin to set and harden. It is mainly composed of calcium silicates, calcium phosphates, zirconium oxide, bioactive glasses, calcium sulfate, calcium oxide, fillers, accelerators, and thickening agents. As per the manufacturer, this hydrophilic sealer forms hydroxyapatite on setting and chemically bonds to dentin.

There have been several proposed methods to attain a superior apical seal and prevent endodontic failure caused by leakage at the apex. However, results from previous studies have been inconsistent, with some indicating that the single-cone obturation technique is superior[5] to cold lateral compaction and warm vertical compaction, while others have suggested the opposite or comparable results.[6,7]

There has been no general consensus on the sealing ability of bioceramic sealers when used with different obturating methods. Hence, this study aimed to evaluate and compare the apical leakage of Cerafill RCS bioceramic sealer and gutta percha when used with three different obturating techniques.

MATERIALS AND METHODS

The Scientific Advisory Committee and the Institutional Ethics Committee Ref.No. TDCEC/23/2020 both gave their approval for this *in vitro* investigation. Thirty-four mandibular premolars, extracted for orthodontic and periodontal purposes were selected for the study. All the sample teeth were confirmed for single roots and single canal by 2 radiographs taken in bucco-lingual and mesio-distal views. Teeth with mature apices, straight roots, and straight canals were selected for the study. Teeth with calcified canals, previous endodontic treatment, decay, cracks, fractures, internal or external resorption, curved canals, and curved roots were excluded from the study. The teeth were decoronated at the cementoenamel junction, leaving 16 mm of root length. Two teeth were kept aside after decoronation and did not undergo further root canal preparation and obturation procedure. These teeth served as negative control.

Root canal preparation

To treat the remaining 32 teeth, access cavity preparation was done and the working length was determined by subtracting 1 mm from the length of a 15K file until the tip could barely be seen at the apex. Biomechanical preparation was carried out using ProTaper Gold rotary files (DENTSPLY Maillefer, Ballaigues, Switzerland) up to size F3. The root canals were copiously irrigated using 1 mL 5.25% NaOCl after each instrument. Final irrigation was done using 5 mL of 5.25% NaOCl, saline, 5 mL of 17% ethylenediaminetetraacetic acid and saline. Passive ultrasonic irrigant activation was done using 5.25% NaOCl and a no. 15 U file (Mani, India) attached to the scaler handpiece kept 1 mm short of the working length for 1 min.

Root canal obturation

The experiment commenced with the random division of the teeth into three experimental groups ($n = 10$) based on the obturation technique, and a control group ($n = 2$).

The first experimental group, Group 1, utilized the single-cone obturation technique to obturate 10 teeth. This involved selecting an F3 gutta-percha master cone that fit the working length and elicited a "tug back." After drying the canal with F3 absorbent paper points, the sealer was placed using manufacturer-provided intra-canal tips. The sealer was also applied to the tip of the master cone, which was subsequently inserted into the canal. Then, the mastercone was seared at the orifice.

For Group 2, 10 teeth were obturated using the cold lateral compaction technique. Mastercone selection and sealer placement were similar to Group 1. Then, lateral compaction was done using a finger spreader and accessory gutta-percha cones until the spreader could not penetrate the canal further. Finally, the mastercone and excess gutta percha was seared at the orifice.

For Group 3, 10 teeth were obturated using warm vertical compaction method. Here, a plugger was adjusted 3 mm short of the working length. Mastercone selection and sealer placement were carried out in a similar manner to Group 1. An activated heat carrier EndoPilot (Schlumbohm, Germany) at 200°C was used to sear off the mastercone at the orifice. The tip of the EndoPilot was then inserted slowly to a depth of 3 mm short of the working length and held there for 3–4 s before being allowed to cool. Next, the tip was removed with a single burst of heat wave for 1 s, which caused an increment of gutta-percha to come out. The canal was then back-filled with thermoplasticized gutta-percha by using a backfill device (Schlumbohm, Germany) to inject a 3–4 mm segment at a time, and each increment was compacted with a prefitted plugger.

To serve as a positive control, 2 roots were left unobturated after being prepared till size F3, allowing for 100% leakage. Meanwhile, 2 roots were not subjected to any root canal preparation or obturation to serve as the negative control group.

After obturation, all roots were checked for vertical root fractures under a microscope and then stored in a 100% RH incubator at 37°C for 1 day to allow the sealer to set. The experimental and positive control groups' specimens were coated with clear paint, excluding the top access and apex. The entire length of specimens in the negative control group was coated with two layers of clear paint to prevent any leakage.

Method of preparation of glucose leakage model [Figure 1]

Figure 1 shows the experimental setup involved using a leakage model as demonstrated by Xu *et al*.[8] The top part of the specimen was connected to an Eppendorf tube which was then connected to a plastic tube (15 cm). The entire setup was placed into a 5-mL plastic container containing 1 mL of 0.2% NaN3, and all connections were secured with M-Seal and sticky wax to prevent leakage. A solution of glucose (1 mol/L, pH 7.0) with 0.2% NaN3 was introduced into the Eppendorf tube through the plastic tube, creating a fluid pressure of 1.5 kPa by elevating the solution 14 cm above the top of the specimen. Any glucose that leaked through the root canal was collected in a plastic bottle. The entire setup was kept in a 100% RH incubator at 37°C for 7–14 days. The quantity of glucose that reached the apical reservoir was quantified using a glucose oxidase method and an ultraviolet-visible spectrophotometer at a wavelength of 340 nm.

RESULTS

The absorbance value obtained using the spectrophotometer was converted to concentration value using Lambert–Beer law.

A (absorbance) = C (concentration) × Path length × ε.

Here, the path length is the length of the path light takes to reach the sample. It is generally equal to the length of the cuvette (2 cm). ε is the extinction coefficient. Glucose has a molecular extinction coefficient of 1.27 × 104 L mol-1 cm-1.

The statistical analysis was conducted using IBM SPSS software (Version 17.0, IBM, New York, USA) and included one-way ANOVA test and *post hoc* Bonferroni test for multiple comparisons.

The negative control group did not exhibit any detectable leakage during all the observation times, while the positive control group had the highest level of leakage.

According to the results presented in Table 1 and Graph 1, teeth obturated using single-cone obturation technique showed the highest levels of leakage at both 7 and 14 days, while the teeth obturated using warm vertical compaction obturation technique had the lowest levels of leakage.

Furthermore, the glucose leakage was found to be significantly higher ($P < 0.05$) in the single-cone obturation technique group compared to the warm vertical compaction obturation technique group at both 7- and 14-day time intervals.

DISCUSSION

This study aimed to evaluate and compare the apical leakage of a bioceramic sealer when used with three different obturating techniques. Cerafill RCS is a new injectable bioceramic sealer. To date, no research has been conducted to investigate the effectiveness of this sealer in conjunction with different obturating methods.

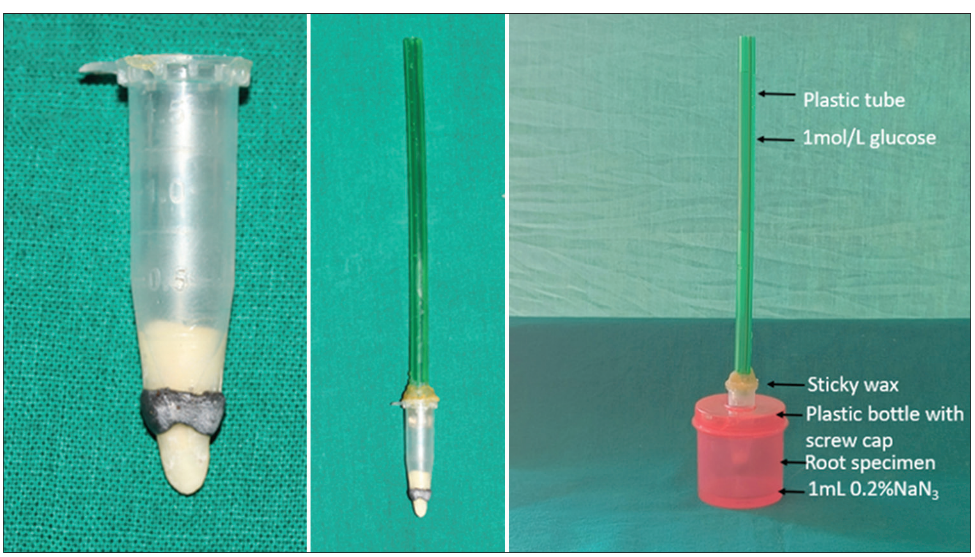
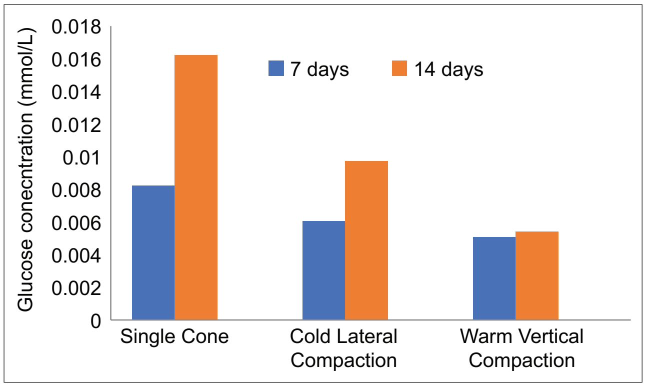


Figure 1: Glucose Leakage model

Table 1: Mean±standard deviation values of glucose concentrations (mmol/L)

Time interval	Mean±SD			Significance
	Single-cone	Cold lateral compaction	Warm vertical compaction	
7 days	0.00822±0.00154	0.00604±0.00329	0.005090±0.0018829	0.019*
14 days	0.01622±0.00376	0.00971±0.00916	0.005410±0.0020669	0.001*
Significance	0.001*	0.043*	0.332	

*Statistically significant values ($P<0.05$). SD: Standard deviation



Graph 1: Mean values of glucose for various obturating groups at 7 days and 14 days

A meticulous irrigation protocol was followed to remove the smear layer, open dentinal tubules so that it can lead to better sealer penetration and thus improve the chances of better apical seal.

There exist several approaches for measuring apical leakage. Nonetheless, the *in vitro* techniques for assessing leakage cannot be generalized to an *in vivo* situation, regarding both the nature and extent of leakage. However, laboratory tests remain the only effective preclinical screening procedure that can foretell or indicate clinical outcomes. Researchers have implemented different experimental methods for evaluating microleakage following obturation, such as radioisotopes,[9] dyes,[10] bacteria,[11] proteins,[12] endotoxins,[11] glucose penetration,[8] and computerized fluid filtration.[13]

It is possible for studies to yield inconsistent findings because of differences in tracers and criteria employed in leakage tests. An example of this is the varying reports on the effectiveness of the lateral compaction technique in contrast to the thermoplasticized compaction technique. In addition, the physicochemical attributes and particle size of the tracer material utilized in the experiment can also impact the outcomes.[14]

According to literature, the glucose leakage model operates on the principle of glucose filtration rate through the materials used for root canal obturation.[8] Glucose is chosen as the tracer due to its small molecular size (MW = 180 Da) and nutritional value for bacteria.[8] The model assumes that if glucose enters the root canal through the oral cavity, it can facilitate the growth of bacteria postbiomechanical preparation and obturation, potentially causing periapical inflammation.[15] The glucose leakage model is highly sensitive and can determine the cumulative amount of microleakage over time. Some studies propose that the total quantity of leakage may hold greater clinical significance than the rate of leakage, as the cumulative effect of microorganism infiltration could lead to periapical inflammation.[16,17] To achieve precise outcomes in the current research, the models were placed in a sealed incubator with 100% humidity throughout the study, to prevent any form of evaporation.

Throughout all the observation periods, it was evident that the warm vertical compaction technique presented the lowest amount of apical leakage. Conversely, the lateral compaction technique showcased higher leakage, which may be attributed to various factors. The utilization of accessory gutta-percha cones might have led to more voids between these cones. In addition, the cold and rigid gutta-percha cones may not have been able to adapt appropriately to the root canal wall and each other, resulting in inadequate sealing. Single-cone obturation technique exhibited a greater amount of glucose leakage, which can be due to less homogenous filling, leading to more voids and consequently more leakage. In contrast to the lateral compaction technique, the use of thermoplasticized gutta-percha in warm vertical compaction enabled the gutta-percha to penetrate the complex structures of the root canal system, resulting in better and more efficient sealing. Several studies have found similar results where greater leakage was observed in cold lateral compaction and single-cone obturation as compared with warm vertical compaction.[6,18,19] However, some studies have reported no significant difference in leakage between different obturation techniques.[7] In addition, some studies have reported higher leakage with the thermal method than when cold gutta-percha was used.[15,20] This difference may be due to the type of root canal sealer used, such as resin-based sealers undergoing faster polymerization shrinkage.[20]

The study by Xu *et al*.[19] reported very high glucose leakage values, which differed from the present study. This difference may be attributed to the resin-based sealer used by Xu *et al*.,[19] which tends to shrink or partially dissolve over time. On the other hand, the bioceramic sealer used in this study demonstrated a unique sealing mechanism. It forms interlocking mechanical connections through tubular diffusion of sealer particles and apatite crystal formation. In addition, it infiltrates the mineral content of the sealer into the intertubular dentin, creating a zone of mineral infiltration by denaturing the collagen fibers with a strong alkaline sealer. These distinctive properties of the bioceramic sealer contribute to more effective sealing and decreased leakage, which is a significant advantage over other conventional sealers.[21]

There have been several studies on the sealing ability and quality of obturations produced by these sealers. Yanpiset *et al*.[22] did a micro-computed tomography analysis and bacterial leakage test to study the quality of obturations and sealing ability using bioceramic-impregnated gutta percha cones or gutta percha, with TotalFill bioceramic sealer or AH Plus sealer. They found no significant difference in the number of voids and leakage between the groups using the single-cone obturating technique. Later, Antunovic *et al*.[23] studied the sealing ability of four bioceramic sealers along with AH plus resin-based sealer using a bacterial leakage model. Conversely, they found better sealing ability with TotalFill bioceramic sealer; as compared to other bioceramic sealers, BioRoot root canal sealer; MTA Fillapex, MTA Plus; and AH Plus sealer. However, the results of our study cannot be directly compared due to the individual differences in individual composition and different methods used for evaluating leakage.

The present study utilized a relatively small number of samples, which may call for reconsideration of the results and validity of the study. The length of the root filling was significantly longer (16 mm as compared to 12 mm–15 mm in the previous studies), and studies have reported that longer root fillings result in less leakage.[24] However, the results in this study may be influenced by the difficulty in maintaining a bacteria-free system in the glucose leakage model, which can lead to glucose consumption and the risk of water evaporation. Another limitation is that the obturating materials used may react with the tracer (glucose) used.[25]

The present study has certain limitations that need to be considered. First, the results of this study could be altered due to the presence of anatomical variations or apical deltas which are commonplace in the apical 3 mm of mandibular premolars. This study tested only one bioceramic sealer, hence the results cannot be extrapolated to all the bioceramic sealers. Within the limitations of the present study, one can note that there is a need for continual assessment of the sealing ability of bioceramic sealers over a prolonged period. There is also a need for validation of the results of this investigation using other contemporary methods of leakage detection, canal adaptation, and dentinal tubular penetration. Furthermore, it will be of great scientific significance if a comparison with other sealers of the same genre sealers can be done in future.

CONCLUSIONS

It can be deduced that for the bioceramic sealer used in this study, the warm vertical compaction technique provides significantly better sealing compared to the single-cone technique. Therefore, the null hypothesis was rejected.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Punia SK, Nadig P, Punia V. An *in vitro* assessment of apical microleakage in root canals obturated with gutta-flow, resilon, thermafil and lateral condensation: A stereomicroscopic study. J Conserv Dent 2011;14:173-7.
2. Farea M, Masudi S, Wan Bakar WZ. Apical microleakage evaluation of system B compared with cold lateral technique: *In vitro* study. Aust Endod J 2010;36:48-53.
3. Muliyar S, Shameem KA, Thankachan RP, Francis PG, Jayapalan CS, Hafiz KA. Microleakage in endodontics. J Int Oral Health 2014;6:99-104.
4. Sarkar NK, Caicedo R, Ritwik P, Moiseyeva R, Kawashima I. Physicochemical basis of the biologic properties of mineral trioxide aggregate. J Endod 2005;31:97-100.
5. Shetty KP, Satish SV, Luke AM, Badade AR, Kilaru KR. *In vitro* interrelationship between apical fill and apical leakage using three different obturation techniques. J Int Soc Prev Community Dent 2018;8:503-7.
6. Özkurt-Kayahan Z, Barut G, Ulusoy Z, Oruçoğlu H, Kayahan MB, Kazazoğlu E, *et al.* Influence of post space preparation on the apical leakage of calamus, single-cone and cold lateral condensation obturation techniques: A computerized fluid filtration study. J Prosthodont 2019;28:587-91.
7. Olczak K, Pawlicka H. Evaluation of the sealing ability of three obturation techniques using a glucose leakage test. BioMed research international. 2017.
8. Xu Q, Fan MW, Fan B, Cheung GS, Hu HL. A new quantitative method using glucose for analysis of endodontic leakage. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2005;99:107-11.
9. Dow PR, Ingle JI. Isotope determination of root canal failure. Oral Surg Oral Med Oral Pathol 1955;8:1100-4.
10. Johnson WT, Zakariasen KL. Spectrophotometric analysis of microleakage in the fine curved canals found in the mesial roots of mandibular molars. Oral Surg Oral Med Oral Pathol 1983;56:305-9.
11. Carratù P, Amato M, Riccitiello F, Rengo S. Evaluation of leakage of bacteria and endotoxins in teeth treated endodontically by two different techniques. J Endod 2002;28:272-5.
12. Messing JJ. An investigation of the sealing properties of some root filling materials. J Br Endod Soc 1970;4:18-22.
13. Tay KC, Loushine BA, Oxford C, Kapur R, Primus CM, Gutmann JL, *et al. In vitro* evaluation of a Ceramicrete-based root-end filling material. J Endod 2007;33:1438-43.
14. Kersten HW, Moorer WR. Particles and molecules in endodontic leakage. Int Endod J 1989;22:118-24.
15. Bernabé PF, Gomes-Filho JE, Bernabé DG, Nery MJ, Otoboni-Filho JA, Dezan E Jr., *et al.* Sealing ability of MTA used as a root end filling material: Effect of the sonic and ultrasonic condensation. Braz Dent J 2013;24:107-10.
16. Shemesh H, Wu MK, Wesselink PR. Leakage along apical root fillings with and without smear layer using two different leakage models: A two-month longitudinal *ex vivo* study. Int Endod J 2006;39:968-76.
17. Wu MK, Fan B, Wesselink PR. Diminished leakage along root canals filled with gutta-percha without sealer over time: A laboratory study. Int Endod J 2000;33:121-5.
18. Rajeswari P, Gopikrishna V, Parameswaran A, Gupta T, Kandaswamy D. *In-vitro* evaluation of apical micro leakage of thermafil and Obtura II sheated gutta percha in comparison with cold lateral condensation using fluid filtration system. Endodontology 2005;17:24.

19. Xu Q, Ling J, Cheung GS, Hu Y. A quantitative evaluation of sealing ability of 4 obturation techniques by using a glucose leakage test. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2007;104:e109-13.
20. Wu MK, Kašt'aková A, Wesselink PR. Quality of cold and warm gutta-percha fillings in oval canals in mandibular premolars. Int Endod J 2001;34:485.
21. Atmeh AR, Chong EZ, Richard G, Festy F, Watson TF. Dentin-cement interfacial interaction: Calcium silicates and polyalkenoates. J Dent Res 2012;91:454-9.
22. Yanpiset K, Banomyong D, Chotvorrarak K, Srisatjaluk RL. Bacterial leakage and micro-computed tomography evaluation in round-shaped canals obturated with bioceramic cone and sealer using matched single cone technique. Restor Dent Endod 2018;43:e30.
23. Antunovic M, Vukmanovic L, Budimir A, Kabil E, Anic I, Bago I. Evaluation of sealing ability of four bioceramic root canal sealers and an epoxy resin-based sealer: An *in vitro* study. Saudi Endod J 2021;11:66.
24. Mattison GD, Delivanis PD, Thacker RW Jr., Hassell KJ. Effect of post preparation on the apical seal. J Prosthet Dent 1984;51:785-9.
25. Shemesh H, Souza EM, Wu MK, Wesselink PR. Glucose reactivity with filling materials as a limitation for using the glucose leakage model. Int Endod J 2008;41:869-72.