RESEARCH ARTICLES

Comparison of apical sealing ability between bioceramic and zinc oxide eugenolbased sealer during root canal treatment, *in vitro*

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ABSTRACT

Obturation with a sealer material that provides an adequate apical sealing ability is required to prevent endodontic treatment failure due to microleakage. However, there are no sealers that meet all the physical and chemical properties to be able to hermetically seal the root canal system to date. Various sealer materials have been developed in recent years including the use of bioceramic materials which are claimed to have excellent biocompatibility to tissues. This study aimed to compare the apical sealing ability of bioceramic-based and zinc oxide eugenol (ZOE)-based sealer in root canal treatment. A total of 27 extracted mandibular premolars were decoronated to the standard root length of 14 mm. The root canals were prepared with a crown-down technique using manual instrument to file F3 (30/.09). The samples were then divided into three groups: obturation with bioceramic-based sealer (n=9); ZOE-based sealer (n=9); and control group (n=9). Microleakage was measured using a dye penetration method with 1% methylene blue and observed under stereomicroscope at x20 magnification. The mean of the maximum penetration length from the lowest to the highest was found in the bioceramic-based sealer group (0.825 mm), the ZOE-based sealer group (3.850 mm), and the control group (4.444 mm). One-way ANOVA test showed a significant difference in the maximum penetration length between the three groups (p<0.05). The post hoc LSD test showed a significant difference in the maximum penetration length between the bioceramic-based and ZOE-based sealer groups (p<0.001). Obturation with bioceramic-based sealer groups (p<0.001). Obturation with bioceramic-based sealer groups (p<0.001). Obturation with bioceramic-based sealer.

Keywords: apical sealing ability; bioceramic-based sealer; maximum penetration length; microleakage; ZOE-based sealer

INTRODUCTION

The three-dimensional obturation of the root canal system has been widely accepted as one of the key factors for successful endodontic treatment.^{1,2} Obturation with inadequate apical sealing can cause microleakage due to voids or gaps in the root canal which allow the entry of oral microorganisms, thus leading to endodontic treatment failure.^{2,3} A frequently used obturation material that has become the standard material to date is gutta-percha in combination with a root canal sealer.¹

A root canal sealer has the ability to fill the gaps between the gutta-percha and root canal walls, as well as to fill the accessory canals and areas that cannot be reached by the gutta-percha itself.^{4,5} All kinds of sealers in use so far are available and classified according to their

basic materials, one of them is zinc oxide eugenol (ZOE)-based sealer which is the most widely used standard sealers in root canal treatment because of its long history of successful use.6-9 Previous studies showed that ZOE-based sealers have mild analgesic properties, minimal dimensional changes, and relatively good adhesion to root canal walls; they are also easy to manipulate and radiopaque.^{10,11} However, these sealers contain formaldehyde and eugenol that could be a cytotoxic agent to periradicular tissue.^{12,13} Various modifications to ZOE-based sealers continue to be developed from the original formula by Rickett or Grossman to obtain a composition that is well tolerated by tissues and provides anti-inflammatory effects, including by making a ZOE-based sealer formulation without the formaldehyde content as used in the present study.7,13

There are no sealers that meet all the physical and chemical properties to be able to hermetically seal the root canal system to date, so various sealer materials have been developed in recent years including the use of bioceramic materials which are claimed to have excellent biocompatibility to tissues.^{1,14} The materials contained in bioceramicbased sealers such as calcium hydroxide and calcium phosphate can produce a high pH to increase antibacterial activity, as well as produce a chemical composition and crystal structure similar to dental hard tissue, thereby increasing the bond strength between the sealer and root canal dentin.^{1,6} These advantages are the reasons why bioceramic is one of the ideal sealer materials for obturation of the root canal system.14

Several studies have been conducted to evaluate the apical sealing ability of bioceramicbased sealers.^{1,15,16} However, there are no studies conducted to compare this material with the most commonly used standard sealer material, namely ZOE. This study aimed to compare the apical sealing ability of bioceramic-based and ZOEbased sealers in root canal treatment.

MATERIALS AND METHODS

The study used an experimental laboratory design. The study was conducted at the Radiology Installation of Sultan Agung Islamic Dental and Oral Hospital, Dental Preclinical Laboratory of Diponegoro University, and Biotechnology Laboratory of Diponegoro University. This study was approved by the Health Research Ethics Commission of the Faculty of Medicine Diponegoro University (182/EC/KEPK/FK-UNDIP/VII/2020).

Noncarious, intact, extracted mandibular premolar teeth with a single root, single canal, and fully formed apex were collected. All the teeth were extracted for orthodontic reasons. These teeth were visually inspected under a magnifying glass and transillumination with light-curing for the presence of any crack or resorption areas. Any teeth with cracks, root fractures, root caries, dental fillings, evidence of periapical resorptive processes, calcified canals, or multiple canals were excluded from the study. The samples of this study were divided randomly into three groups, namely obturation with bioceramic-based sealer (A), ZOE-based sealer (B), and control group (C). The sample size was obtained based on Federer's formula which resulted in $n \ge 8.5$. This calculation showed that the minimum number of samples needed for each group was nine teeth, so for the three groups, twenty-seven teeth were needed.

Twenty-seven selected teeth were stored in 10% formalin solution for 1 week followed by decoronation of the teeth to obtain a standard root length of 14 mm using a diamond disc along with a straight handpiece and micromotor. A #15 K-file with a rubber stopper was introduced into the root canal and the working length was determined by reducing 0.5 mm from its total length. The root canals were instrumented with the crown-down technique using Protaper Universal (Dentsply Maillefer, Switzerland) manual instruments to file size F3 (30/.09). The manufacturer's instructions were followed for the sequence of the instrument and the torque was used for biomechanical preparation. Between the uses of each file, the root canal irrigation was done with 1 ml of 5.25% NaOCI solution and recapitulation was performed using a #15 K-file to maintain its working length. Final irrigation was done by using 2 ml of 5.25% NaOCI solution, 2 ml of distilled water, followed by 2 ml of 17% EDTA and ended with 2 ml of distilled water. All the irrigating solutions were delivered through side vented 30-gauge needles and the root canals were dried with corresponding paper points. This protocol was employed for all the experimental groups as well as for the control group.

The roots were then filled as follows. Group A (n=9): the roots were obturated with Ceraseal (Meta Biomed, South Korea) and gutta-percha Protaper F3 using a single-cone technique, Group B (n=9): the roots were obturated with Endoseal (Prevest DenPro, India) and gutta-percha Protaper F3 using a single-cone technique, Group C (n=9): the roots were obturated with gutta-percha Protaper F3 using a single-cone technique without the use of sealer. Endoseal sealer was dispensed and mixed as per the manufacturer's instruction. Ceraseal

sealer was available as a premixed paste. Before the placement of the sealers, gutta-percha alone was placed to assess the tug-back and its fit to working length. Excess gutta-percha was then removed with a heat-carrier plugger 1 mm below the canal opening. The root canal orifices were sealed with a temporary filling material, namely Ceivitron (Dong Quan, Taiwan).

After the filling was complete, the density of the obturation results of all the samples was evaluated with a radiographic image. Subsequently, all the samples were stored at 37 °C temperature for 24 hours in an incubator (Memmert, Germany). Then the outer surfaces of the roots were covered with two-layer nail polish except 1 mm from the end of the apex, each layer was allowed to dry for 1 hour at a room temperature. All the samples were then soaked in 1% methylene blue solution for 72 hours at a room temperature. After the samples were removed from the 1% methylene blue solution, all the samples were then washed under running water and allowed to dry. The roots were sectioned buccolingually using a diamond disc so they split into two longitudinal halves. The section showing the clearer visualization of the dye and longer penetration was used for observations.

The observations were carried out under a stereomicroscope at x20 magnification. The data taken were the microleakage of the dye in the apical third obtained from the measurement of the maximum penetration length of the dye into the root canal in each of the samples using a millimeter grid and a sliding caliper. Two observers measured independently and the average score of the measurement results of the two observers was considered the final score. The data were collected and all the groups were compared using one-way analysis of variance (ANOVA) using SPSS Version 21.0 (IBM, US). Post-hoc least significant difference (LSD) followup test was performed to analyze the significance level between the groups where p<0.05 was considered significant. Intraclass correlation coefficient (ICC) test was also carried out at the beginning to assess the inter-rater reliability of the measurement results.

RESULTS

Figure 1 shows the differences of the maximum penetration length among the three groups which can be seen from the penetration of 1% methylene blue dye from the apical to the coronal direction vertically. The mean score and standard deviation of the maximum penetration length in the three groups are shown in Table 1. The results of this study showed that the highest mean of the maximum penetration length was found in Group C (4.444 mm) and the lowest mean of the maximum penetration length was found in Group A (0.825 mm), indicating that the mean leakage in Group A was the lowest compared to the other groups. The ICC test result showed the inter-rater

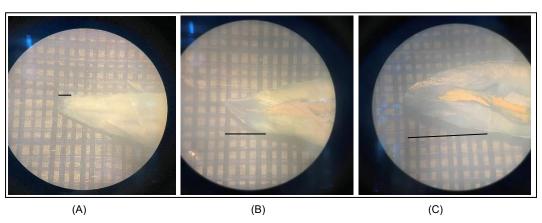


Figure 1. Microscopic image of the obturation of the bioceramic-based sealer group (A), the ZOE-based sealer group (B), and the control group (C) on observation with a x20 magnification stereomicroscope

Table 1. Mean, standard deviation (SD), and the result of the one-way ANOVA test on maximum penetration length of the three groups

| | Groups | Mean ± SD (mm) | P-value |
|----------------------------|-----------------------------|----------------|---------|
| Maximum penetration Length | Bioceramic-based sealer (A) | 0.825 ± 0.914 | |
| | ZOE-based sealer (B) | 3.850 ± 1.552 | 0.000* |
| | Control (C) | 4.444 ± 1.423 | |

*Significance: p<0.05

Table 2. Mean difference and the result of the post hoc LSD test on maximum penetration length between groups

| | Groups | | |
|-----------------|------------------|----------------------|---------------|
| | Bioceramic – ZOE | Bioceramic – Control | ZOE – Control |
| Mean difference | 3.025 | 3.619 | 0.594 |
| P-value | 0.000* | 0.000* | 0.351 |

*Significance: p<0.05

reliability of the measurement between the two observers with a coefficient of 0.984, categorized as a very good inter-rater agreement so the data used was reliable.

The result of the one-way ANOVA test showed p<0.05 as seen in Table 1. These results indicated a significant difference in the maximum penetration length among the three groups. The result of the post hoc LSD test in Table 2 showed that the maximum penetration length of the bioceramic-based sealer group was the lowest and significantly different with the other two groups (p<0.001). However, the difference of the maximum penetration length between the ZOEbased sealer group and the control group was not statistically significant (p>0.001).

DISCUSSION

Over the years, many techniques and materials have been used to obtain a dense and hermetic apical sealing, yet no root canal filling material has met the ideal characteristics of achieving this to date.¹⁷ This study was conducted in the apical third area of the root canal which is the most difficult area to clean, prepare, and obturate because it has a very complex anatomy where there are many lateral root canals.^{3,16} An adequate apical sealing is required to prevent microleakage which can lead to endodontic treatment failure.¹⁶ The samples used in this study were mandibular premolars which aimed to uniform the samples and to minimize anatomical variations. The samples were treated with a single-cone obturation technique with two different types of sealers, namely bioceramic and ZOE-based sealers, and with a control group without using sealers. Cohen (2018) suggested that a single-cone obturation technique was chosen because it is more efficient and allows a minimum working time than other obturation techniques. The single-cone obturation technique has a better ability to clinically preserve the coronal dentin and reduce the lateral pressure on the root canal.18

Various methods have been suggested to detect and evaluate microleakage. Observation of microleakage in this study was carried out by the dye penetration method of 1% methylene blue which was immersed for 72 hours. This method is more popular than other methods because of its simple procedure and reliable results.^{16,19,20} Theoretically, if the root canal filling does not allow the penetration of small molecules such as dyes, then the material has the potential to prevent leakage due to larger molecules such as bacteria.²⁰

The results of this study showed that the mean of the maximum penetration length from the lowest to the highest was found in the group treated with the bioceramic-based sealer, ZOE-based sealer, and the control group without the use of sealer. The maximum penetration length indicates the microleakage level of the sealer material against the methylene blue dye. The lower the microleakage level, the better the apical sealing ability of sealer materials. Based on these results, obturation with a bioceramic-based sealer provides a better apical sealing ability than that with a ZOE-based sealer. This result is in line with previous research by Hasnain et al and Asawaworawit et al which showed that a bioceramic-based sealer has a better apical sealing ability compared to various other sealers including epoxy resin and methacrylatebased sealers.^{1,4} Better results of bioceramicbased sealer are related to the hydrophilicity, biocompatibility, and its small particle size which is approximately ≤48 µm or even nanosized. These biocompatible materials tend to have mechanical properties similar to dental hard tissues, thus allowing the sealer particles to penetrate easily into the dentinal tubules and fill the lateral root canal. This penetration ability further produces a mechanical interlocking bond that enhances apical sealing ability.^{1,6,14} The bioceramic-based sealer also showed the expansion of up to 0.2% after setting reactions, its expansion ability together with chemical and micromechanical bonds could increase the bonds between the sealers and the root canal wall.1,21

The difference in the apical sealing ability between the bioceramic and ZOE-based sealers was found to be significant with p<0.05, indicating that ZOE-based sealers have a higher level of microleakage compared to bioceramic-based sealers. This result is in accordance with studies conducted by Lankar et al. and Aryanto where ZOE-based sealers showed the largest number of microleakage compared to other groups.^{16,22} The fact that ZOE-based sealer experiences larger microleakage compared to other sealer materials is presumably because it has a longer setting time, slow absorption in periradicular tissue, and low bond strength between the sealer material and root canal walls.²² According to Rahaswanti et al, the application of ZOE-based sealer tends to be difficult so there is often a lack of filling. In addition, the operator in this study lacked experience especially in endodontic treatment. That is why microleakage was found in this treatment group.¹⁰

In this study, the post hoc LSD test between the ZOE-based sealer and the control groups did not show any significant difference in the maximum penetration length with p>0.05, thus indicating that obturation with the ZOE-based sealer and without using any sealers gave unfavorable results in terms of the apical sealing. These results may be influenced by various limitations in this study such as incubation and sample immersion period. This study used an incubation period of 24 hours at 37 °C to provide an opportunity for the sealers to set. The bioceramic-based sealers were not affected by the incubation period because they have a fast setting time of three to four hours. However, this did not apply to the ZOE-based sealers due to a longer setting time of around 24-36 hours, and nonacceptable flow rates.14,15,22 The characteristics of the ZOE-based sealer caused some samples in this treatment group to experience an incomplete setting so the sealer material dissolved when the samples were immersed in the methylene blue dye. This assumption served as the basis for obtaining insignificant differences between the ZOE-based sealer and the control groups.

Ingle and Bakland (2008) suggested that the most common cause of endodontic failure is incomplete obturation of the root canals.²³ No obturation technique can fully produce cavity-free obturation including the gold standard, namely the lateral condensation obturation technique.²⁴ Various literature studies suggest that lateral condensation is a technique that has been used for a long time. This technique is clinically effective and does not require special equipment.^{24–26} The limitation of this study is that the stereomicroscope used could not be connected to any digital software so the maximum penetration length was measured manually. This limitation was minimized with the help of a millimeter grid that was placed under the longitudinal halves of the tooth and then measured using a sliding caliper. However, this method allowed for subjectivity, so the measurements were carried out by two observers independently to eliminate bias.¹⁹ These limitations did not affect the validity of the study.

For further research, it is necessary to conduct research to test the apical sealing ability of bioceramic and ZOE-based sealers with a longer incubation and sample immersion period to improve the accuracy of the results. In addition, there is a need for conducting research using other obturation techniques such as the lateral condensation technique which is the gold standard in root canal obturation.

CONCLUSION

Based on the results of research that has been done, it can be concluded that the obturation with a bioceramic-based sealer provides a better apical sealing ability than that with a ZOE-based sealer in root canal treatment.

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REFERENCES

- Hasnain M, Bansal P, Nikhil V. An in vitro comparative analysis of sealing ability of bioceramic-based, methacrylate-based, and epoxy resin-based sealers. Endodontology. 2017; 29(2): 146-150. doi: 10.4103/endo.endo 101 16
- 2. Sikri V, Sidhu B, Sroa R, Setia P. Apical sealing ability of two novel root canal sealers: an ex-

vivo study. J Int Clin Dent Res Organ. 2013; 5(1): 9-13. doi: 10.4103/2231-0754.134130

 Kumar NS, Palanivelu A, Narayanan LL. Evaluation of the apical sealing ability and adaptation to the dentin of two resin-based Sealers: An in vitro study. J Conserv Dent. 2013; 16(5): 449-453.

doi: 10.4103/0972-0707.117518

 Asawaworarit W, Pinyosopon T, Kijsamanmith K. Comparison of apical sealing ability of bioceramic sealer and epoxy resin-based sealer using the fluid filtration technique and scanning electron microscopy. J Dent Sci. 2020; 15(2): 186-192.

doi: 10.1016/ j.jds.2019.09.010

- de Melo TV, Prado MC, Hirata R, Fidel SR, da Silva EJNL, Sassone LM. Improved sealing ability promoted by calcium silicate-based root canal sealers. Braz J Oral Sci. 2018; 17: 1-8. doi: 10.20396/bjos.v17i0.8652651
- Al-Haddad A, Aziz ZACA. Bioceramic-based root canal sealers: a review. Int J Biomater. 2016; 2016:10. doi: 10.1155/2016/9753210
- Komabayashi T, Colmenar D, Cvach N, Bhat A, Primus C, Imai Y. Comprehensive review of current endodontic sealers. Dent Mater J. 2020; 39(5): 703-720. doi: 10.4012/dmj.2019-288
- Javidi M, Zarei M, Naghavi N, Mortazavi M, Nejat AH. Zinc oxide nano-particles as sealer in endodontics and its sealing ability. Contemp Clin Dent. 2014; 5(1): 20-24. doi: 10.4103/0976-237X.128656
- Tour Savadkouhi S, Fazlyab M. Discoloration potential of endodontic sealers: a brief review. Iran Endod J. 2016; 11(4): 250-254. doi: 10.22037/iej.2016.20
- Rahaswanti LWA. Evaluasi keberhasilan pengisian saluran akar dengan sediaan zinc oxide eugenol dan campuran calcium hydroxide dengan pasta iodoform. Intisari Sains Medis. 2017;8(1):1-7.
- Singh H, Markan S, Kaur M, Gupta G. Endodontic sealers: current concepts and comparative analysis. Dent - Open J. 2015; 2(1): 32-37. doi: 10.17140/DOJ-2-107

- Nikolić M, Gašić J, Mitić A, Popović J, Radovanović A, Barac R. Bone tissue response to endomethasone implanted into the rat mandible. Acta Fac Medicae Naissensis. 2015; 32(2):155-164. doi: 10.1515/afmnai-2015-0016
- Jain P. Current Therapy in Endodontics. 1st ed. Oxford: John Willey & Sons, Inc.; 2016. 264-267.
- Nasseh A. The rise of bioceramics. Endod Prac [Internet]. 2009 [cited 2020 Mar 29]:21-25. Available from: https://www.endoexperience. com/documents/bioceramicsealer.pdf.
- Ayer A, Manandhar TR, Agrawal N, Vikram M, Suwal P. A comparative study of apical microleakage of different root canal sealers by apical dye penetration. Bangladesh Journal of Medical Science. 2017; 16(2): 219-224. doi: 10.3329/bjms.v16i2.23421
- Aryanto M. Comparison of microleakage in root canals obturation using zinc oxideeugenol and epoxy resin based sealer. ICEASD 2019. Apr 1-2. Indonesia; 2019. doi: 10.4108/eai.1-4-2019.2287254
- Al-thawab E, Khursheed I. A comparative evaluation of apical sealing ability between AH26, MTA and pure portland cement: a vitro study. EC Dent Sci. 2019; 18(11): 11-18. doi: 10.13140/RG.2.2.33483.75049
- Cohen RG. Successful single-cone obturation. Insid Dent [Internet]. 2018 [cited 2020 Oct 6];14(3). Available from: https:// www.aegisdentalnetwork.com/id/2018/03/ successful-single-cone-obturation.
- Sardar P, Abidi S, Iqbal W, Meo A, Jawaed N, Khan R. An in-vitro assessment of apical sealing ability of resin based endodontic sealer at various time intervals. J Pak Dent Assoc. 2015; 24(2): 75-80.

- Sonmez I, Oba A, Sonmez D, Almaz M. In vitro evaluation of apical microleakage of a new MTA-based sealer. Eur Arch Paediatr Dent. 2012; 13(5): 252-255. doi: 10.1007/BF03262880
- Pawar S, Pujar M, Makandar S. Evaluation of the apical sealing ability of bioceramic sealer, AH plus & epiphany: an in vitro study. J Conserv Dent. 2014; 17(6): 579-582. doi: 10.4103/0972-0707.144609
- Lankar A, Mian RI, Mirza AJ, Siddiqui AA, Alam MK. A comparative evaluation of apical sealability of various root canal sealers used in endodontics. Int Med J. 2018; 25(1): 39-41.
- Ingle JI, Leif K. Bakland. Ingle's Endodontics
 6. Shelton, Connecticut: People's Medical Publishing House; 2008. 3-42.
- Al Qassab SJ, Hadi D Al, Maniangant Luke A. Evaluation of three different obturation techniques using three-dimensional cone beam computed tomography: in vitro study. Dentistry. 2016; 6(12): 6-10. doi: 10.4172/2161-1122.1000403
- 25. Samadi F, Jaiswal J, Saha S, Garg N, Chowdhary S, Samadi F, Tripathi VP. A comparative evaluation of efficacy of different obturation techniques used in root canal treatment of anterior teeth: an in vitro study. Int J Clin Pediatr Dent. 2014; 7(1): 1-5. doi: 10.5005/jp-journals-10005-1224
- Varun K, Harpreet S, Rajinder B, Samrity P. Qualitative and quantitative comparative evaluation of sealing ability of guttaflow, thermoplasticized gutta percha and lateral compaction for root canal obturation: a cohort, controlled, ex-vivo study. J Oral Health Dent Manag. 2013; 12(3):155-161.