Journal of Oral Science. doi.org/10.2334/josnusd.23-0235

Original Article

Evaluation of the root dentin bond strength and intratubular biomineralization of a premixed calcium aluminate-based hydraulic bioceramic endodontic sealer

Yu-Na Lee[1)], Min-Kyeong Kim[2)], Hee-Jin Kim[2)], Mi-Kyung Yu[1,3,4)], Kwang-Won Lee[1,3,4)], and Kyung-San Min*[1,3,4)]

[1)]Department of Conservative Dentistry, School of Dentistry, Jeonbuk National University, Jeonju, Republic of Korea
[2)]Department of Dentistry, Department of Dentistry, College of Medicine, Kosin University, Busan, Republic of Korea
[3)]Research Institute of Clinical Medicine of Jeonbuk National University, Jeonju, Republic of Korea
[4)]Biomedical Research Institute of Jeonbuk National University Hospital, Jeonju, Republic of Korea

Received June 28, 2023; Revised November 28, 2023; Accepted December 12, 2023; J-STAGE Advance Publication: ●●●● ●●, 2024

Abstract

Purpose: This study evaluated the dentin bonding strength and biomineralization effect of a recently developed premixed calcium aluminate-based endodontic sealer (Dia-Root Bio Sealer) in comparison with existing calcium silicate-based sealers.
Methods: The root canals of 80 mandibular premolars were filled with Dia-Root Bio Sealer, Endoseal MTA, EndoSequence BC Sealer, and AH Plus Bioceramic Sealer. Medial and apical specimens were then obtained by sectioning. The push-out bond strength was measured using the medial specimens, and the failure mode was recorded. Intratubular biomineralization in the apical specimens was analyzed using scanning electron microscopy and energy-dispersive X-ray spectroscopy (EDS). The data were analyzed using one-way analysis of variance followed by the Tukey test ($P < 0.05$).
Results: The push-out bond strength of Dia-Root Bio Sealer was significantly higher than that of the other tested materials, and a cohesive failure pattern was observed in all groups. Dia-Root Bio Sealer also exhibited a significantly higher degree of biomineralization than the other groups, and EDS analysis indicated that the biomineralized precipitates were amorphous calcium phosphate.
Conclusion: The results of this study indicate that Dia-Root Bio Sealer has the potential to be used as an adequate root canal sealer due to its favorable bonding performance.

Keywords: biomineralization, calcium aluminate, endodontic, push-out bond strength, sealer

Introduction

Since mineral trioxide aggregate (MTA) first appeared in the literature as a material for repair of root perforation in the 1990s, it has been widely used in the field of endodontics for purposes including root-end filling, apexification, and vital pulp treatment [1,2]. In essence, MTA is a calcium silicate (CS)-based hydraulic cement [3,4] that has been used successfully for about 30 years. Because the chemical composition has changed, and many derivatives have been introduced to improve performance, it is now classified as a hydraulic bioceramic material [5]. In addition, to improve manipulability and ease of application, a so-called “premixed” method has been developed, whereby cement powder is mixed with a non-aqueous vehicle. This type of sealer is now widely used in clinical practice [6,7].

Gutta-percha (GP) is the most frequently used ideal core filling material, but it does not adhere to the dentin surface of the canal [8]. Therefore, an endodontic sealer should be used to compensate for this insufficiency. For this purpose, the endodontic sealer should have adequate flowability to fill the gap between the GP and the root canal wall and show adequate strength of bonding to the root dentin. Therefore, the quality of the canal filling depends upon the sealer [9,10]. The strength of bonding between root canal filling materials and root dentin is primarily evaluated using the push-out test [11-15].

Calcium aluminate (CA) cement was developed to overcome the disadvantages of CS cements such as MTA. A previous study reported that CA cement was less porous and had better fluidity, more favorable handling properties, greater mechanical strength, and a smaller pore size than CS-based MTA [16]. In fact, CA is the most reactive component in Portland cement [17], and is known to exhibit rapid hydration and provide the compound’s initial mechanical strength [18]. Based on research showing improvements over CS-based dental cements, a premixed hydraulic bioceramic endodontic sealer with maximized CA content has been developed (Dia-Root Bio Sealer, DiaDent, Cheongju, Republic of Korea). However, no study has investigated its bonding performance with dentin. In addition, biomineralization—a phenomenon in which mineralized precipitates are generated in dentinal tubules by bioceramic endodontic sealers—has also been observed [19,20]. Until now, studies on intratubular biomineralization have been limited to qualitative evaluations based on simple observation [21], and no quantitative analysis has been performed. Accordingly, the purpose of the present study was to evaluate the bonding performance of Dia-Root Bio Sealer, including its push-out bond strength and biomineralization, in comparison with commercially available CS-based premixed sealers. The null hypothesis was that there would be no significant differences in dentin bond strength and the degree of biomineralization between the tested materials.

Materials and Methods

Preparation of specimens

A total of 80 extracted mandibular premolars with a straight, single root canal free of caries and cracks and with fully developed roots were obtained. The teeth were stored in 0.5% sodium hypochlorite (NaOCl; Sense Cleaner, SNS Dental, Incheon, Republic of Korea) until the experiment. All teeth were decoronated at the cementoenamel junction, and the length of each root was adjusted to approximately 15 mm. A size #10 K-File (Dentsply Sirona, Ballaigues, Switzerland) was then inserted into the root canal until the tip of the file was visible in the apical foramen. The canal length was determined by subtracting 0.5 mm from this length. All root canals were enlarged to F4 size using a ProTaper Universal nickel-titanium rotary instrument (Dentsply Sirona). Afterward, to remove the smear layer, the root canal was irrigated with 1% NaOCl for 10 min, then with 1 mL of 17% ethylenediaminetetraacetic acid (EDTA; Wako Chemical, Osaka, Japan) for 1 min. After irrigation with 10 mL of sterile distilled water for 1 min to remove the residual washing solution, the root canal was dried using a paper point (Diadent, Cheongju).

The samples were randomly divided into 4 groups (n = 20), and the root canals were filled with 4 root canal sealers: Dia-Root Bio Sealer (DB), Endoseal MTA (ES; Maruchi, Wonju, Republic of Korea), EndoSequence BC Sealer (BC; Brasseler USA, Savannah, GA, USA), and AH Plus Bioceramic Sealer (AB; Dentsply Sirona Tulsa Dental Specialties, Johnson City, TN, USA). The chemical composition of the tested products is listed in Table 1. All teeth were filled using a sealer-based obturation method with a single GP cone. Briefly, each root canal sealer was injected directly

*Corresponding author: Kyung-San Min, Department of Conservative Dentistry, School of Dentistry and Institute of Oral Bioscience, Jeonbuk National University, 567 Baekje-daero, Deokjin-gu, Jeonju-si 54896, Republic of Korea
Fax: +82-63-250-2129 Email: endomin@gmail.com

Table 1 Materials used in this study

Material	Composition (%)	MSDS issue date
Dia-Root Bio Sealer (Diadent, Cheongju, Republic of Korea)	calcium silicate (0-5), calcium aluminate (20-50), ytterbium trifluoride (10-40), zirconium dioxide (0-10), polyethylene glycol (5-30), hydrophobic amorphous fumed silica (0.01-5), hydroxypropyl methylcellulose (0.1-10), polyoxyethylene (20), sorbitan monooleic acid (0-10), light mineral oil (0-5)	2/12/2019
Endoseal MTA (Maruchi, Wonju, Republic of Korea)	natural pure cement (27.81), zirconium dioxide/bismuth trioxide (47.28), thickening agents (24.91)	27/5/2016
EndoSequence BC Sealer (Brasseler USA, Savannah, GA, USA)	zirconium oxide (35-45), tricalcium silicate (20-35), dicalcium silicate (7-15), calcium hydroxide (1-4)	15/1/2018
AH Plus Bioceramic Sealer (Dentsply Sirona Tulsa Dental Specialties, Johnson City, TN, USA)	zirconium dioxide (50-70), tricalcium silicate (5-15), dimethyl sulfoxide (10-30), lithium carbonate (<0.5), thickening agents, etc. (<6)	7/2/2021

MSDS: material safety data sheet

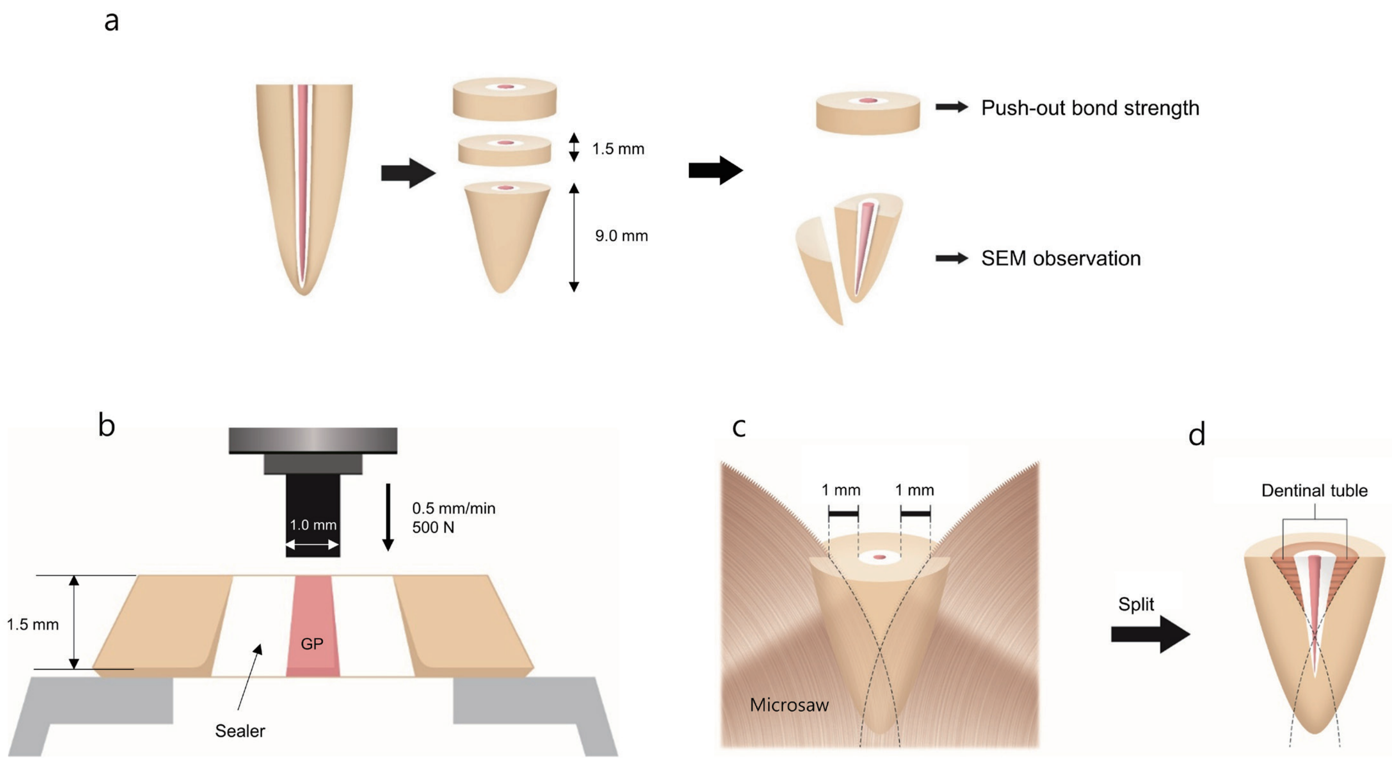


Fig. 1 Illustration of the experimental procedure. (a) The tooth filled with the tested materials was sectioned horizontally to obtain a sliced specimen and an apical segment. (b) Push-out bond strength was measured using the sliced specimen and a universal testing machine. (c-d) The apical segment was sectioned longitudinally and split for observation of the intratubular biomineralization.
SEM: scanning electron microscope; GP: gutta-percha

into the root canal up to the apical third, and then the selected master GP cone (taper: 0.04, size: 40) was inserted into the root canal slowly. The extra GP cone was cut at the canal orifice level using Duo-Alpha (B&L Biotech, Ansan, Republic of Korea). No additional GP cone was used. The specimens were stored in phosphate-buffered saline (PBS) (HyClone Laboratories, Logan, UT, USA) to promote biomineralization at room temperature for 30 days.

Evaluation of push-out bond strength and failure mode

The tooth specimens were embedded in acrylic resin (Ortho-Jet Acrylic, Lang Dental MFG, Wheeling, IL, USA). Each buried specimen was cut perpendicular to the longitudinal axis using a low-speed diamond saw (Isomet, Buehler, Evanston, IL, USA). Consequently, a specimen 1.5 ± 0.2 mm thick was obtained from the middle of the root, which was used to measure the push-out bond strength (Fig. 1a). The remaining apical specimens were stored in PBS until subsequent testing for biomineralization. A custom jig was fabricated to fix the specimen in place for measuring the push-out bond strength of the filling material. A metal rod with a cylindrical tip (1.0 mm in diameter) fixed in the superior portion of a universal testing machine (Z020, Zwick Roell, Ulm, Germany) was positioned over the specimen. Then, a push-out force of 500 N was applied to the filling material in the specimen's center in an apical-to-coronal direction, with a cross-head speed of 0.5 mm/min (Fig. 1b). The maximum failure load was recorded in N and converted to MPa by applying the following formula.

$$\text{Push-out bond strength (MPa)} = \frac{\text{Maximum load (N)}}{\text{Adhesion area of root filling (A) (mm}^2)}$$

The bonding area (A) in the filled root canal was obtained by applying the following formula, where π is the constant 3.14, r1 and r2 are the smaller and larger radii, and h is the thickness of the section in millimeters.

$$A = \pi(r1 + r2) \times \sqrt[2]{(r1 - r2)^2 + h^2}$$

After the push-out bond strength had been measured, the failure mode was evaluated by observing the specimen under a stereoscopic microscope (MZ16FA, Leica, Wetzlar, Germany) at ×30 magnification. Failure modes were classified as cohesive failure (dentin wall completely covered with the root canal sealer), adhesive failure (root canal sealer not visible on the dentin wall), and mixed failure (both cohesive and adhesive failures).

Evaluation of intratubular biomineralization

The specimens were cut using a method employed previously for observing biomineralization [5]. The apical section was cut using a low-speed diamond saw on both sides down the longitudinal axis of the tooth, leaving a distance of 1 mm to the filling material (Fig. 1c). The remaining parts were then split, and the split surface was observed with a scanning electron microscope (SEM; SU8230, Hitachi, Tokyo, Japan) to evaluate biomineralization in the dentinal tubules (Fig. 1d). In addition, the chemical composition of the mineralized precipitates was determined using energy-dispersive X-ray spectroscopy (EDS) (7593-H; Horiba, Tokyo, Japan).

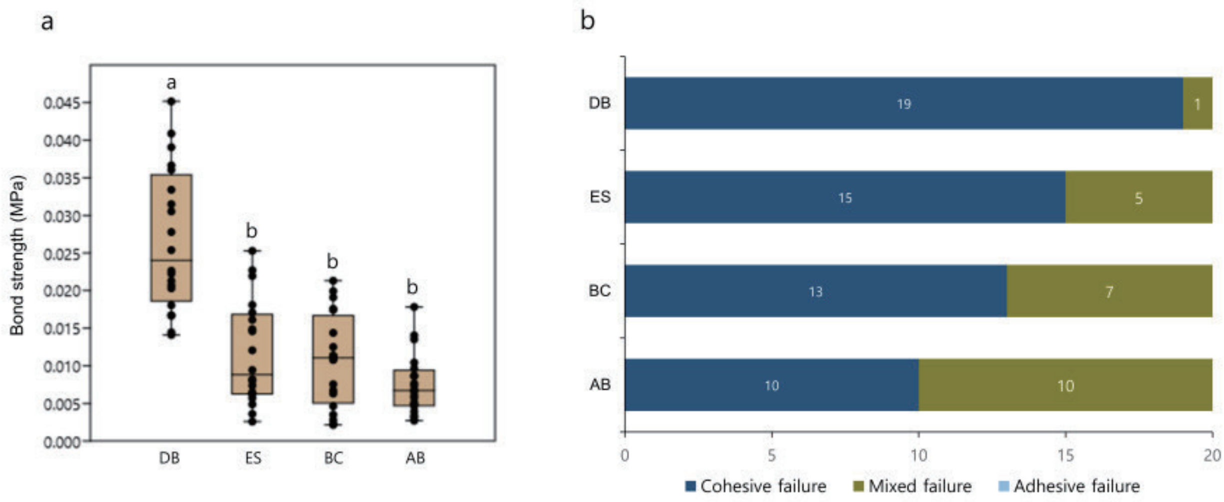


Fig. 2 Push-out bond strength and failure modes of the tested materials. (a) Bar chart showing the bond strength (mean ± standard deviation) of the tested material groups. (b) Failure mode distribution according to filling material. Different letters indicate statistically significant differences ($P < 0.05$).
DB: Dia-Root Bio Sealer; ES: Endoseal MTA; BC: EndoSequence BC sealer; AB: AH Plus Bioceramic Sealer

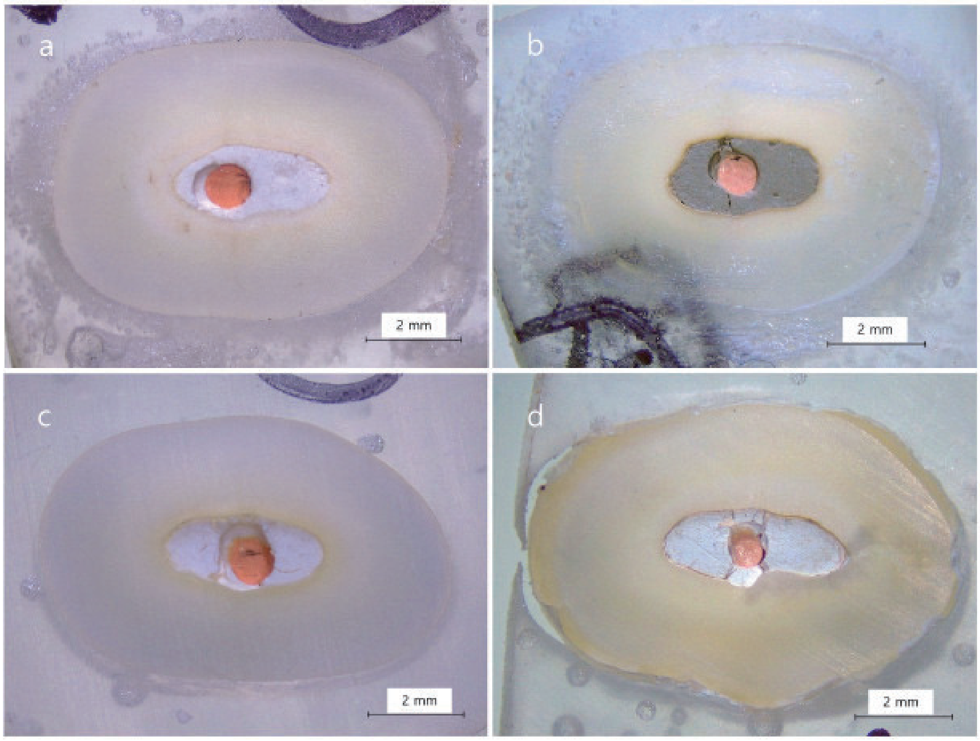


Fig. 3 Failure mode analysis using a stereomicroscope at ×30 magnification. (a-d) Representative images of DB, ES, BC, and AB, respectively.
DB: Dia-Root Bio Sealer; ES: Endoseal MTA; BC: EndoSequence BC sealer; AB: AH Plus Bioceramic Sealer

SEM images were obtained by selecting 5 points on the exposed dentinal tubule randomly for each tooth. The acquired images were analyzed using the Image J program (version 1.53s, National Institutes of Health, Bethesda, MD, USA). The procedure for image processing was as follows: (i) polygon selection of dentinal tubules, (ii) calculation of the pixel area corresponding to dentinal tubules, (iii) conversion of binary images and separation of precipitate and background by image threshold adjustment, (iv) calculation of pixel area corresponding to the precipitate inside the dentinal tubule, and (v) calculation of the sediment area relative to the dentinal tubule area (precipitate/dentinal tubule ratio). In step (iii), since the precipitate appeared brighter than the rest of the empty tubule space, the image was considered binary and a threshold value was used to separate the background (empty space) from the precipitate. An appropriate threshold value was applied by selecting a value that excluded the background, while including all precipitates in each image.

Statistical analysis

In order to determine the appropriate number of specimens required for the experiment, power analysis was performed using the G*Power 3.1 program (University of Düsseldorf, Düsseldorf, Germany) with the following assumptions: F-test, effect size = 0.48, power = 0.95. The normality and homoscedasticity of the data were confirmed using the Shapiro-Wilk test and Levene test, respectively. Consequently, one-way analysis of variance (ANOVA) and the Tukey *post hoc* test were used. The statistical analyses were performed using PAST version 4.03 [22]. A P-value of less than 0.05 was considered to indicate statistical significance.

Results

Push-out bond strength and failure mode

The push-out bond strength in the DB group was significantly higher than that in the other groups ($P < 0.05$), and no statistically significant difference was observed among the other 3 groups ($P > 0.05$) (Fig. 2a). When the failure modes were analyzed, no group showed cases of adhesion failure, and cohesive failure was the most common result. Mixed failures occurred most often with AB, followed by BC, ES, and DB (Figs. 2b, 3).

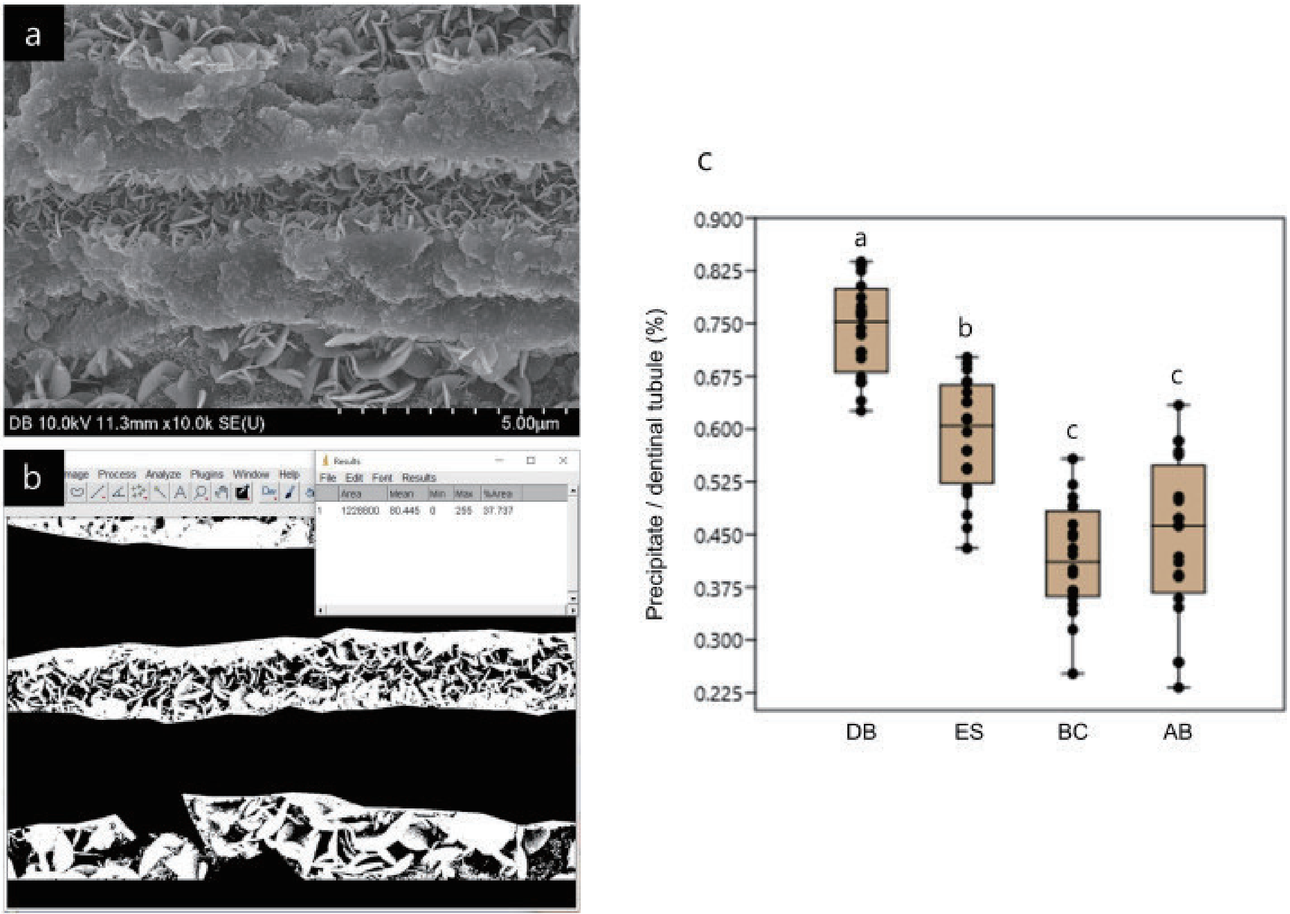


Fig. 4 Intratubular biomineralization. (a-b) Representative SEM images for evaluation of intratubular biomineralization. (c) Quantitative analysis of intratubular biomineralization in the tested groups (mean ± standard deviation). Different letters indicate statistically significant differences ($P < 0.05$).
DB: Dia-Root Bio Sealer; ES: Endoseal MTA; BC: EndoSequence BC sealer; AB: AH Plus Bioceramic Sealer

Intratubular biomineralization

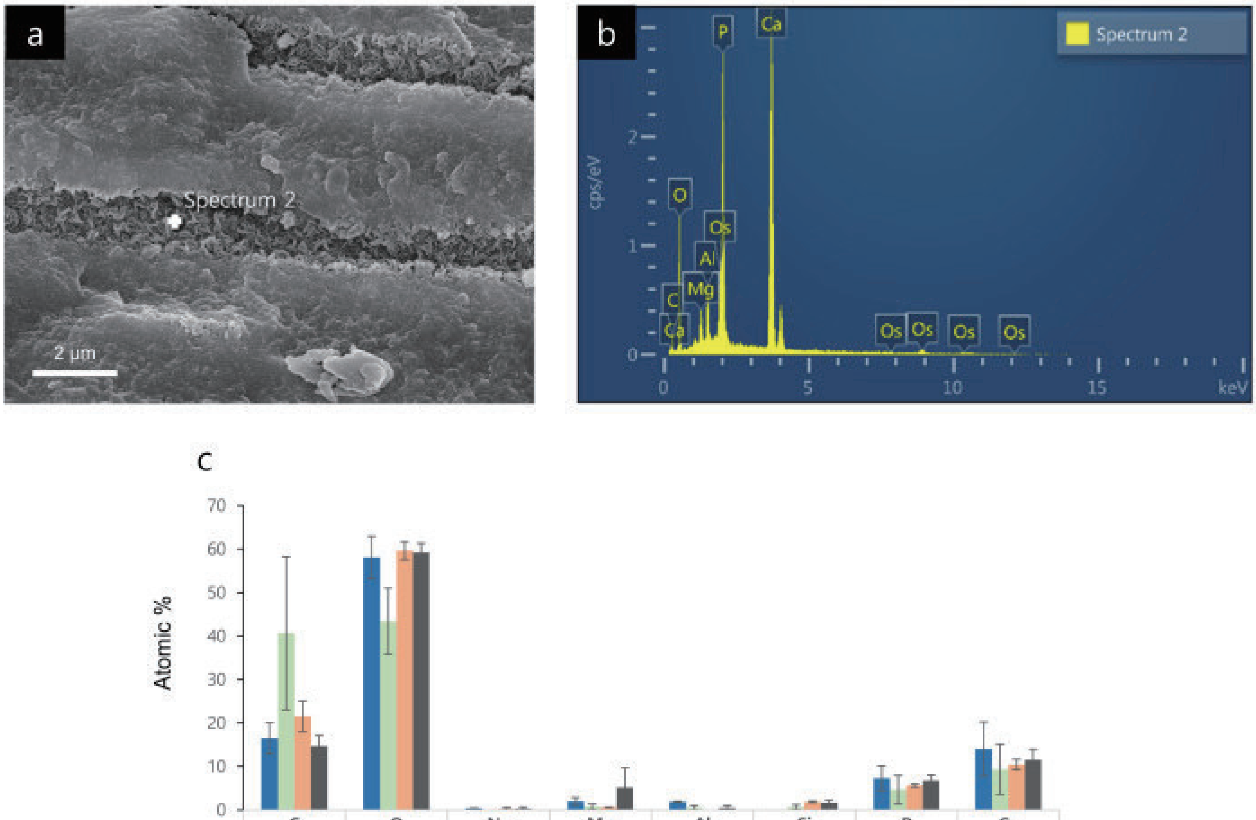
As shown in Fig. 4a and 4b, small flake-like precipitates were observed along the dentinal tubules in all groups. Quantitative analysis revealed that the DB group had significantly more biomineralization than the other groups ($P < 0.05$) (Fig. 4c). In addition, significantly more biomineralization occurred in the ES group than in the BC and AB groups ($P < 0.05$). EDS analysis revealed that the average atomic ratios of calcium to phosphorus were 1.9 for DB, 2.1 for ES, 1.8 for BC, and 1.7 for AB, being generally higher than the 1.67 ratio for hydroxyapatite (HA) (Fig. 5).

Discussion

In this study, the bonding performance of the newly developed CA-based premixed bioceramic sealer, DB, was evaluated in terms of its potential to fill root canals. For this purpose, the push-out bond strength of the DB sealer was compared with that of commercially available CS-based premixed sealers. It was found that DB had significantly higher push-out bond strength than the other sealers (Fig. 2a). It is possible that differences in composition may affect the bond strength of sealers [23]. According to the manufacturer's information, DB contains CA as a base material and ytterbium trifluoride (YbF_3) as a radiopacifier. In contrast, BC and AB contain zirconium oxide (ZrO_2) to provide radiopacity. YbF_3 is a radiopaque substance used frequently for dental cements, and shows comparable radiopacity to bismuth oxide (Bi_2O_3) and higher radiopacity than ZrO_2 [24]. This suggests that less YbF_3 could be included to provide radiopacity comparable to that of ZrO_2. Indeed, BC and AB have ZrO_2 contents of 35-45% and 50-70%, respectively, and 47% of ES is a mixture of ZrO_2 and Bi_2O_3. In contrast, the manufacturer of DB indicated that its YbF_3 content is 10-40%. ES, BC, and AB have CS contents of 27%, 27-50%, and only 5-15%, respectively. Consequently, it might be possible to incorporate more materials such as CA and CS into sealers. Indeed, according to the manufacturer's information, the CA and CS content of DB is 20-55%, which may contribute to its higher bond strength. Oliveira et al. [25] reported that MTA exhibited significantly higher bonding strength values than CS-based premixed sealers, due to the sealers' lower CS content [6,26]. Furthermore, YbF_3 increased the compressive strength of CS cement when its content was 20-30% [27]. In this study, failure occurred mainly in the materials (i.e. cohesive), and not at the sealer-dentin interface (i.e. adhesive or mixed). These results indicate that bond strength values rely primarily on the intrinsic strength of the materials. However, other additives such as vehicles and thickening agents may cause differences in bond strength [28].

After the push-out bond strength test, observation of failure modes showed that either cohesive or mixed failure occurred in all experimental groups, with no instances of adhesive failure. Notably, more than half of the specimens in all experimental groups showed cohesive failure. This high cohesive failure rate may have resulted from the low strength of the sealers. In a recent study by Park et al. [5], which evaluated the push-out bond strength of premixed bioceramic cements for root-end filling and vital pulp therapy, cohesive failure occurred most often. However, adhesive failures were also noted. The difference can be explained by the lower intrinsic strength of the premixed sealers, which enables possible retreatment if initial endodontic treatment fails. In this respect, it is possible that the intrinsic strength of the material itself exceeds the strength of bonding with dentin (adhesive failure) in the case of root-end filling materials. However, for the endodontic sealer, the material was weaker than the bonding strength and cohesive failures were more likely to occur.

In this study, the degree of intratubular biomineralization was analyzed quantitatively. Confirmation of intratubular biomineralization might provide indirect evidence for the sealing ability of a root canal filling [20]. Reyes-Carmona et al. [19] also stated that biomineralization could contribute to strengthening the resistance of MTA to dislodgement from dentin. In the present study, DB showed a significantly higher degree of biomineral-



	C	O	Na	Mg	Al	Si	P	Ca
DB	16.53	58.06	0.41	2.00	1.82	0.00	7.27	14.03
ES	40.63	43.43	0.00	0.73	0.56	0.66	4.64	9.32
BC	21.53	59.6	0.31	0.62	0.00	1.84	5.62	10.44
AB	14.69	59.23	0.32	5.17	0.49	1.69	6.78	11.56

Fig. 5 EDS analysis of the chemical characteristics of intratubular precipitate. (a) SEM image showing the precipitate (white cross). (b) Representative graph of EDS analysis of the precipitate. (c) Semi-quantitative analysis of the chemical composition of the crystalline area, denoted with a white cross (mean ± standard deviation).
EDS: energy-dispersive X-ray spectroscopy; SEM: scanning electron microscopy; DB: Dia-Root Bio Sealer; ES: Endoseal MTA; BC: EndoSequence BC sealer; AB: AH Plus Bioceramic Sealer

ization than the other sealers. The high degree of biomineralization in the DB group might have contributed to the increase in push-out bond strength, since both properties depend upon each sealer's respective chemical composition. Majeed et al. [29] reported that the biomineralization ability of calcium-based cement was directly proportional to the quantity of calcium ions released and the presence of phosphate. The semi-quantitative EDS analysis revealed that all tested groups contained large amounts of calcium and phosphorus, although DB showed the highest atomic percentage of calcium in the precipitates (Fig. 5). Since ytterbium has a higher atomic mass (173.04 u) than zirconium (91.224 u), it is possible that the required radiopacifier content of the sealer could be reduced when manufacturing DB, and that more calcium-based materials could be incorporated instead. In addition, the average calcium-to-phosphorus ratios were 1.9 in DB, 2.1 in ES, 1.8 in BC, and 1.7 in AB, being generally higher than the 1.67 ratio observed in HA or the 1.50-1.70 ratio observed in human enamel and dentin surfaces. This means that the precipitate produced in this study was closer to amorphous calcium phosphate than to crystalline HA.

As noted, previous studies of bioceramic sealer biomineralization have been limited to qualitative analysis. Therefore, the present study adopted quantitative analysis of precipitate formation in dentinal tubules for the first time. Briefly, 5 points were randomly selected on each specimen by an observer unrelated to the experiment, and normality and equal variance were confirmed before comparison of mean values. This allowed one-way ANOVA, a parametric statistical test, to be used. However, during the evaluation process for image analysis, accuracy may have been limited because the polygons for the dentinal tubules were selected manually. In addition, for differentiation of the precipitate from the background using binary image conversion and image threshold adjustment, the threshold value was selected arbitrarily. Nevertheless, it is noteworthy that this is the first study to have analyzed the degree of biomineralization in dentinal tubules quantitatively.

This study employed a sealer-based filling technique using a single GP cone. This is easier to implement than other obturation methods such as lateral compaction and warm vertical compaction. Consequently, it is less sensitive to operator variations and potentially less damaging to the root canal dentin [30]. Notably, use of a hydraulic bioceramic sealer is believed to be a simpler technique for obturation of all root canal systems [31]. Therefore, this can be considered a more appropriate method for evaluation of the bonding performance of hydraulic bioceramic root canal sealers.

In conclusion, DB, a CA-based hydraulic bioceramic root canal sealer, was shown to exhibit higher bond strength and better intratubular biomineralization than CS-based sealers. Therefore, the null hypotheses were rejected. The present results suggest that DB has the potential to be an adequate root canal sealer in view of its favorable bonding performance.

Ethical Statements

Ethical approval for the use of human teeth was obtained from the institutional review board of Kosin University Gospel Hospital (KUGH-022-08-033).

Conflicts of Interest

None declared

Funding

This study was supported by the Biomedical Research Institute, Jeonbuk National University Hospital, in 2023.

References

1. Lee SJ, Monsef M, Torabinejad M (1993) Sealing ability of a mineral trioxide aggregate for repair of lateral root perforations. J Endod 19, 541-544.
2. Parirokh M, Torabinejad M (2010) Mineral trioxide aggregate: a comprehensive literature review—part I: chemical, physical, and antibacterial properties. J Endod 36, 16-27.

3. Camilleri J, Montesin FE, Brady K, Sweeney R, Curtis RV, Ford TR (2005) The constitution of mineral trioxide aggregate. Dent Mater 21, 297-303.
4. Asgary S, Parirokh M, Eghbal MJ, Stowe S, Brink F (2006) A qualitative X-ray analysis of white and grey mineral trioxide aggregate using compositional imaging. J Mater Sci Mater Med 17, 187-191.
5. Park JH, Kim HJ, Lee KW, Yu MK, Min KS (2023) Push-out bond strength and intratubular biomineralization of a hydraulic root-end filling material premixed with dimethyl sulfoxide as a vehicle. Restor Dent Endod 48, e8.
6. Loushine BA, Bryan TE, Looney SW, Gillen BM, Loushine RJ, Weller RN et al. (2011) Setting properties and cytotoxicity evaluation of a premixed bioceramic root canal sealer. J Endod 37, 673-~~637~~.
7. Guivarc'h M, Jeanneau C, Giraud T, Pommel L, About I, Azim AA et al. (2020) An international survey on the use of calcium silicate-based sealers in non-surgical endodontic treatment. Clin Oral Investig 24, 417-424.
8. Ordinola-Zapata R, Bramante C, Graeff M, Perochena A, Vivan R, Camargo E et al. (2009) Depth and percentage of penetration of endodontic sealers into dentinal tubules after root canal obturation using a lateral compaction technique: a confocal laser scanning microscopy study. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 108, 450-457.
9. Apicella MJ, Loushine RJ, West LA, Runyan DA (1999) A comparison of root fracture resistance using two root canal sealers. Int Endod J 32, 376-380.
10. Rajput JS, Jain RL, Pathak A (2004) An evaluation of sealing ability of endodontic materials as root canal sealers. J Indian Soc Pedod Prevent Dent 22, 1-7.
11. Gesi A, Raffaelli O, Goracci C, Pashley DH, Tay FR, Ferrari M (2005) Interfacial strength of Resilon and gutta-percha to intraradicular dentin. J Endod 31, 809-813.
12. Skidmore LJ, Berzins DW, Bahcall JK (2006) An in vitro comparison of the intraradicular dentin bond strength of Resilon and gutta-percha. J Endod 32, 963-966.
13. Ungor M, Onay EO, Orucoglu H (2006) Push-out bond strengths: the Epiphany-Resilon endodontic obturation system compared with different pairings of Epiphany, Resilon, AH Plus and gutta-percha. Int Endod J 39, 643-647.
14. de Sá MA, Nunes E, Antunes AN, Brito Júnior M, Horta MC, Amaral RR et al. (2021) Push-out bond strength and marginal adaptation of apical plugs with bioactive endodontic cements in simulated immature teeth. Restor Dent Endod 46, e53.
15. Yaduka P, Kataki R, Roy D, Das L, Goswami S (2021) Effects of radiation therapy on the dislocation resistance of root canal sealers applied to dentin and the sealer-dentin interface: a pilot study. Restor Dent Endod 46, e22.
16. Oliveira IR, Pandolfelli VC, Jacobovitz M (2010) Chemical, physical and mechanical properties of a novel calcium aluminate endodontic cement. Int Endod J 43, 1069-1076.
17. Liu WN, Chang J, Zhu YQ, Zhang M (2011) Effect of tricalcium aluminate on the properties of tricalcium silicate-tricalcium aluminate mixtures: setting time, mechanical strength and biocompatibility. Int Endod J 44, 41-50.
18. Oh SH, Choi SY, Choi SH, Lee YK, Kim KN (2004) The influence of lithium fluoride on in vitro biocompatibility and bioactivity of calcium aluminate pMMA composite cement. J Mater Sci Mater Med 15, 25-33.
19. Reyes-Carmona JF, Felippe MCS, Felippe WT (2010) The biomineralization ability of mineral trioxide aggregate and Portland cement on dentin enhances the push-out strength. J Endod 36, 286-291.
20. Yoo YJ, Baek SH, Kum KY, Shon WJ, Woo KM, Lee W (2016) Dynamic intratubular biomineralization following root canal obturation with pozzolan-based mineral trioxide aggregate sealer cement. Scanning 38, 50-56.
21. Yoo YJ, Lee YS, Yoo JS, Perinpanayagam H, Yoo CS, Kang HS et al. (2017) Intratubular biomineralization in a root canal filled with calcium-enriched material over 8 years. Materials (Basel) 10, 1388.
22. Hammer Ø, Harper DAT, Ryan PD (2001) PAST: paleontological statistics software package for education and data analysis. Palaeontol Electron 4, 1-9.
23. Watson TF, Atmeh AR, Sajini S, Cook RJ, Festy F (2014) Present and future of glass-ionomers and calcium-silicate cements as bioactive materials in dentistry: biophotonics-based interfacial analyses in health and disease. Dent Mater 30, 50-61.
24. Collares FM, Ogliari FA, Lima GS, Fontanella VR, Piva E, Samuel SM (2010) Ytterbium trifluoride as a radiopaque agent for dental cements. Int Endod J 43, 792-797.
25. Oliveira DS, Cardoso ML, Queiroz TF, Silva EJ, Souza EM, De-Deus G (2016) Suboptimal push-out bond strengths of calcium silicate-based sealers. Int Endod J 49, 796-801.
26. Koch KA, Brave D (2009) EndoSequence: melding endodontics with restorative dentistry, part 3. Dent Today 28, 88, 90, 92 passim.
27. Antonijevic D, Medigovic I, Zrilic M, Jokic B, Vukovic Z, Todorovic L (2014) The influence of different radiopacifying agents on the radiopacity, compressive strength, setting time, and porosity of Portland cement. Clin Oral Investig 18, 1597-1604.
28. Sanz JL, López-García S, Rodríguez-Lozano FJ, Melo M, Lozano A, Llena C et al. (2022) Cytocompatibility and bioactive potential of AH Plus Bioceramic Sealer: an in vitro study. Int Endod J 55, 1066-1080.
29. Majeed A, AlShwaimi E (2017) Push-out bond strength and surface microhardness of calcium silicate-based biomaterials: an in vitro study. Med Princ Pract 26, 139-145.
30. Kim JA, Hwang YC, Rosa V, Yu MK, Lee KW, Min KS (2018) Root canal filling quality of a premixed calcium silicate endodontic sealer applied using gutta-percha cone-mediated ultrasonic activation. J Endod 44, 133-138.
31. Kooanantkul C, Shelton RM, Camilleri J (2023) Comparison of obturation quality in natural and replica teeth root-filled using different sealers and techniques. Clin Oral Investig 27, 2407-2417.