



## Dentinal Tubule Penetration of Various Root Canal Sealers after Removal of Calcium Hydroxide Intracanal Medicament: An In Vitro Confocal Laser Scanning Microscopic Study

Shreya Patil<sup>1,2\*</sup>, Charudutt Patil<sup>3</sup>, Ashish Medha<sup>4</sup>, Namrata Patil<sup>5</sup>, Priyatam Karade<sup>6</sup>

1. Department of Conservative Dentistry and Endodontics, Pandit Deendayal Upadhyay Dental College and Hospital, Solapur, Maharashtra, India
2. Department of Conservative Dentistry and Endodontics, Krishna Vishwa Vidyapeeth, Karad, Maharashtra, India
3. Department of Prosthodontics, Crown and Bridge, Saraswati Dhanwantari Dental College and Hospital, Parbhani, Maharashtra, India
4. Department of Conservative Dentistry and Endodontics, Vasantdada Patil Dental College and Hospital, Kavalapur, Sangli, Maharashtra, India
5. Department of Oral Pathology and Microbiology, Saraswati Dhanwantari Dental College and Hospital, Parbhani, Maharashtra, India
6. Department of Conservative Dentistry and Endodontics, Vasantdada Patil Dental College and Hospital, Kavalapur, Sangli, Maharashtra, India

| Article Info                                                                                                                                                                                                                                                                                                                                                                                                                                              | ABSTRACT                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                   |
|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p><b>Article type:</b><br/>Original Article</p> <hr/> <p><b>Article History:</b><br/>Received: 18 Jun 2025<br/>Accepted: 20 Dec 2025<br/>Published: 15 Jun 2026</p> <hr/> <p><b>* Corresponding author:</b><br/>Department of Conservative Dentistry and Endodontics, Pandit Deendayal Upadhyay Dental College and Hospital, Solapur, Maharashtra, India<br/>Email: <a href="mailto:shreyadeshpande9900@gmail.com">shreyadeshpande9900@gmail.com</a></p> | <p><b>Objectives:</b> This study compared the penetration depth of three different root canal sealers—AH Plus resin-based, Apexit Plus calcium hydroxide (CH)-based, and BioRoot RCS bioceramic-based sealers—into dentin after removal of CH intracanal medicament.</p> <p><b>Materials and Methods:</b> In this in vitro study, 60 extracted single-rooted mandibular premolars were decoronated with a diamond disc under water cooling to a standard length of 16mm; 12 samples were selected as control, and the rest received CH mixed with rhodamine dye as an intracanal medicament. Two weeks later, CH was removed by manual filing and irrigant activation using EndoActivator. Samples were then divided into 3 groups (n=12) depending upon the sealer used as AH Plus, Apexit Plus, and Bioroot RCS sealer. All sealers were combined with fluorescein dye, and obturation was performed using F3 gutta-percha with lateral compaction technique. After 2 weeks, the teeth were transversally sectioned into coronal, middle, and apical thirds. The penetration depth of sealers was measured via an imaging software. Statistical analysis was performed using ANOVA followed by the Tukey's post-hoc test (<math>\alpha=0.05</math>).</p> <p><b>Results:</b> A statistically significant difference was observed among the groups (<math>P&lt;0.05</math>). The penetration depth of sealers was the lowest in the apical third, and the highest in the coronal third in all three groups. BioRoot RCS showed the highest penetration depth at all three levels, despite CH medicament remnants.</p> <p><b>Conclusion:</b> Bioroot RCS bioceramic sealer was superior to AH Plus resin-based and Apexit Plus CH-based sealers regarding penetration depth into dentin after removal of CH intracanal medicament.</p> <p><b>Keywords:</b> Calcium Hydroxide; Epoxy Resin-Based Root Canal Sealer; Microscopy; Confocal</p> |
| <p>➤ <b>Cite this article as:</b> Patil S, Patil C, Medha A, Patil N, Karade P. Dentinal Tubule Penetration of Various Root Canal Sealers after Removal of Calcium Hydroxide Medicament: An In Vitro Confocal Laser Scanning Microscopic Study. <i>Front Dent.</i> 2026;23:26. <a href="http://doi.org/10.18502/fid.v23i26.22557">http://doi.org/10.18502/fid.v23i26.22557</a></p>                                                                        |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |

## INTRODUCTION

The aim of root canal treatment is to remove the infected tissues, eradicate the microorganisms from the root canals, neutralize any agents that might remain in the root canal system, and prevent reinfection [1]. To effectively remove bacteria and infected tissues, chemomechanical preparation employing either manual or rotary tools along with sodium hypochlorite is considered an effective method in cleaning and shaping of root canals [2]. To remove the remaining microorganisms after biomechanical preparation and to avoid an interappointment reinfection, the use of intracanal medicaments has been suggested [3].

Calcium hydroxide (CH) is recognized as the gold-standard intracanal medicament, but it is essential to completely remove it from the root canals prior to obturation to prevent any potential adverse effects on the treatment results. Remaining CH may result in microleakage at the apex and decrease sealer compatibility; it can also interact with zinc oxide eugenol, diminishing the bond strength to dentin [3]. The thorough and foreseeable removal of CH before root canal obturation is important.

Three-dimensional obturation is the main goal of root canal treatment to seal all portals of exit [4]. Root canal sealers are essential as they aid in sealing the root canals by filling the re-entrances, irregular areas, and dentinal tubules—spaces that cannot be accessed by endodontic instruments—thereby improving the fit of the core filling material at the dentin-material interface [5]. According to the literature, we have no accessible method or irrigant that can eliminate all residual CH from the root canal system so far, and these residues influence the bond strength of endodontic sealers to dentin, the ability of sealers to penetrate into the dentinal tubules, and the compatibility of root canal sealers [6]. Apexit Plus is a CH-based root canal sealer that is both radiopaque and non-shrinking, featuring reduced solubility in tissue fluids and a slight expansion during setting, which ensures a durable seal in root canals [7]. AH Plus has good physicochemical properties, low solubility and disintegration, good adaptation, antibacterial

action, and also good biological properties [7]. Bioroot RCS exhibits favorable bioactive properties through the release of calcium ions, possesses significant alkalizing capability, has the ability to form hydroxyapatite, and demonstrates good radiopacity [8].

This study aimed to compare the dentinal tubule penetration of three types of sealers namely AH Plus resin-based sealer, Apexit Plus CH-based sealer, and Bioroot RCS bio-ceramic-based sealer after removal of CH intracanal medicament using confocal laser scanning microscopy (CLSM).

## MATERIALS AND METHODS

This in vitro study was accepted by the Ethical and Research Committee of Vasantdada Patil Dental College and Hospital in which it was performed (2344/18-19).

Eligibility criteria- 60 Extracted human single-rooted mandibular premolars with a single canal and fully formed apices were included in this study. Teeth with cracks, fractures, multiple canals, root resorption, calcified canals, open apices, anatomical anomalies, or previous endodontic treatment were excluded from the study.

The sample size was determined using G\*Power software (version 3.1.9.7; Heinrich Heine University, Düsseldorf, Germany) for a one-way analysis of variance (ANOVA). Assuming a large effect size (Cohen's  $f=0.40$ ), a significance level ( $\alpha$ ) of 0.05, a statistical power of 80% ( $1-\beta=0.80$ ), and five study groups, yielding a minimum of 60 specimens (12 per group). 60 Extracted human single-rooted mandibular premolars with a single canal and fully formed apices were included in the study. Teeth with cracks, fractures, multiple canals, root resorption, calcified canals, open apices, anatomical anomalies, or previous endodontic treatment were excluded from the study.

The teeth were cleaned of soft tissue and calculus before storage in 0.9% thymol solution. The crowns were cut using a diamond disc under water cooling to achieve a standardized root length of 16mm. Access to the canal was created, and a #10 K-file (Mani Inc., Utsunomiya, Tochigi, Japan) was inserted

into the canal until its tip was visible at the apical foramen. The working length was determined to be 1 mm short of the root apex. Irrigation was performed with 2mL of 2.5% sodium hypochlorite (NaOCl) and 5mL of 17% EDTA. Canals were shaped to F3 ProTaper (Dentsply Maillefer, Ballaigues, Switzerland) and were filled with CH (Prevest Denpro, Jammu, India) as intracanal medicament mixed with 0.1% rhodamine B (Sigma-Aldrich, St. Louis, MO, USA) in 1:1 ratio. This mixture was introduced into the canals using a #30 lentulo spiral (Mani Inc, Tochigi, Japan). The specimens were stored at 37°C in 100% humidity for 2 weeks [6].

The 36 samples were instrumented with #30 H files (Mani Inc., Tochigi, Japan) to remove CH mechanically. The samples were irrigated with 5mL of 2.5% NaOCl and 5mL of 17% EDTA followed by 5mL of saline. The irrigant was activated using an EndoActivator (Dentsply Maillefer, North Carolina, United States) for efficient removal of CH.

60 Samples were randomly divided into five groups (n=12) depending on the type of sealer used:

*-Group A:* Positive control; The canals were instrumented with ProTaper rotary file system (Dentsply Maillefer, North Carolina, United States), irrigated, and filled with CH mixed with 0.1% rhodamine B dye.

*-Group B:* Negative control; The canals were prepared similarly with no intracanal CH placement, and were obturated.

The remaining samples were filled with a mixture of rhodamine B dye and CH powder. After incubation, the samples were instrumented with H files to eliminate CH mechanically and were irrigated and activated using an EndoActivator (Dentsply Maillefer, North Carolina, United States) to aid in the removal of CH medicament.

They were divided as follows:

*-Group C:* AH Plus (Dentsply Sirona, Konstanz, Germany) root canal sealer

*-Group D:* Apexit Plus (Ivoclar Vivadent, Schaan, Liechtenstein) root canal sealer

*-Group E:* Bioroot RCS (Septodont, Saint-Maur-des-Fosses, France) root canal sealer

Each of the sealers were mixed with 0.1%

fluorescein and delivered into the canals using a lentulo spiral. The lateral compaction technique was used for obturation. The samples were kept for 15 days at 37°C with 100% relative humidity to promote the full setting of the sealers.

The samples were sectioned transversely at 2, 5, and 8mm from the apex to represent the apical, middle, and coronal sections, respectively. The sections were captured using CLSM (Carl Zeiss, Germany) along with an epifluorescence method where the absorption and emission wavelengths are 540/590nm for rhodamine B and 536/617nm for fluorescein. The images were assessed in Zen software (Carl Zeiss, Germany) to calculate the deepest penetration depth of sealer in the maximum intensity projection. One observer who was blinded to the groups performed the measurements, which were repeated twice for intra-rater reliability (ICC=0.94).

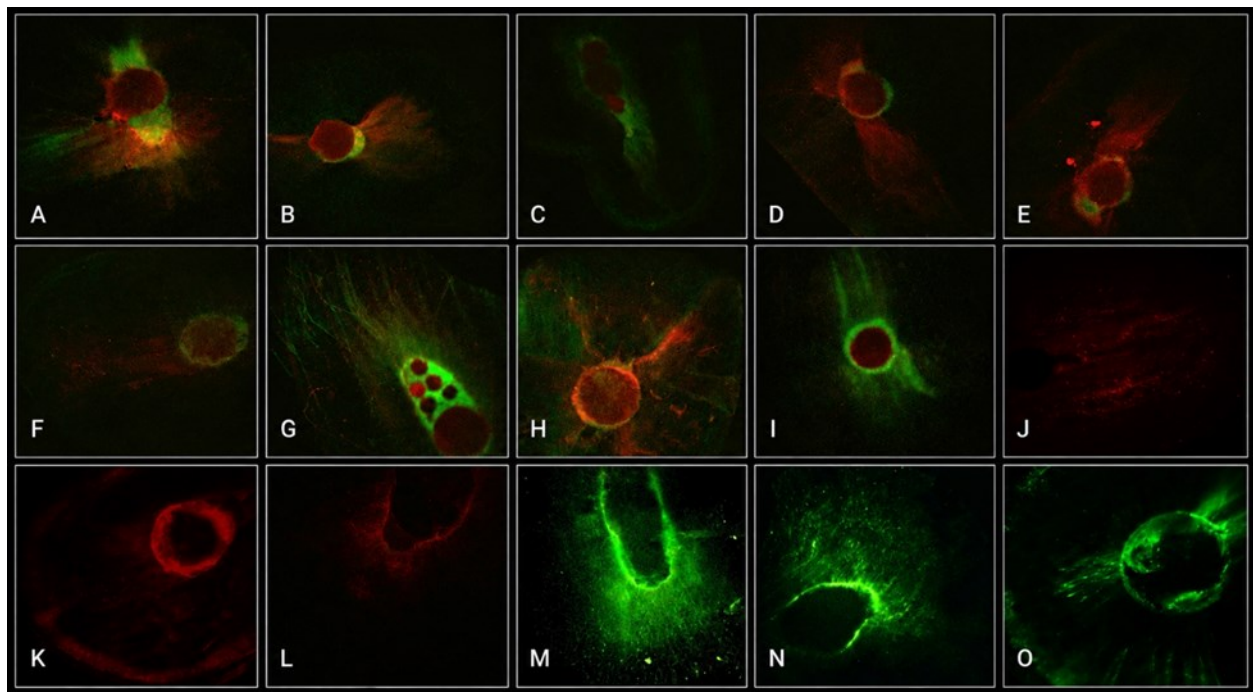
Descriptive statistics were expressed as mean  $\pm$  standard deviation. Normality and homogeneity of variance were assessed using the Shapiro-Wilk and Levene's tests, respectively. Five groups were compared for the mean dentinal penetration of sealers by ANOVA, and Tukey's post-hoc test was conducted for pairwise comparisons.  $P < 0.05$  was regarded as statistically significant. The analysis was performed using SPSS (Version-19, IBM [International Business Machines Corporation], Armonk, New York, USA).

## RESULTS

The coronal section showed the highest sealer penetration, followed by the middle and apical sections for all three sealers (Fig. 1). According to Table 1, the sealer penetration depth was the highest for Bioroot RCS, followed by AH Plus and Apexit Plus sealers ( $P < 0.001$ ). Pairwise comparisons demonstrated significantly greater dentinal tubule penetration in the negative control group than in the positive control, AH Plus, Apexit Plus, and BioRoot RCS groups (all  $p < 0.001$ ). BioRoot RCS exhibited significantly greater penetration than the

positive control, AH Plus, and Apexit Plus (all  $p < 0.001$ ). AH Plus showed significantly greater penetration than Apexit Plus

( $p = 0.018$ ). No significant differences were observed between the positive control and AH Plus ( $p = 0.164$ ) or Apexit Plus ( $p = 0.913$ ).



**Fig 1.** Representative CLSM images in each group: AH Plus/CH: (A) Coronal, (B) Middle, (C) Apical; Apexit Plus/CH: (D) Coronal, (E) Middle, (F) Apical; Bio-Root RCS/ CH: (G) Coronal, (H) Middle, (I) Apical; CH: (J) Coronal, (K) Middle, (L) Apical; Bio-Root RCS: (M) Coronal, (N) Middle; (O) Apical.

**Table 1.** Descriptive statistics for dentinal penetration of sealers (in  $\mu\text{m}$ ) in the apical, middle and coronal thirds in the five groups

| Descriptive Statistics   |    |         |         |          |                |
|--------------------------|----|---------|---------|----------|----------------|
| Group-Section            | N  | Minimum | Maximum | Mean     | Std. Deviation |
| Positive control-Coronal | 12 | 258.31  | 405.88  | 321.0358 | 50.90998       |
| Positive control-Middle  | 12 | 167.00  | 343.19  | 244.1600 | 59.61749       |
| Positive control-Apical  | 12 | 101.45  | 258.31  | 181.8008 | 45.32948       |
| Negative control-Coronal | 12 | 526.87  | 878.45  | 712.3575 | 108.26242      |
| Negative control-Middle  | 12 | 406.53  | 787.90  | 602.4317 | 125.71270      |
| Negative control-Apical  | 12 | 333.21  | 688.21  | 485.9242 | 120.08616      |
| AH Plus-Coronal          | 12 | 231.84  | 465.70  | 372.4775 | 70.85693       |
| AH Plus-Middle           | 12 | 217.17  | 443.73  | 300.2942 | 73.70639       |
| AH Plus-Apical           | 12 | 141.22  | 378.87  | 236.0850 | 62.98630       |
| Apexit Plus-Coronal      | 12 | 173.89  | 442.57  | 302.5475 | 70.96158       |
| Apexit Plus-Middle       | 12 | 137.01  | 322.73  | 235.2942 | 59.66698       |
| Apexit Plus-Apical       | 12 | 90.89   | 211.45  | 147.8883 | 43.00144       |
| Bioroot RCS-Coronal      | 12 | 455.99  | 622.97  | 525.4367 | 46.86448       |
| Bioroot RCS-Middle       | 12 | 369.17  | 562.72  | 417.9983 | 53.10209       |
| Bioroot RCS-Apical       | 12 | 245.87  | 430.61  | 337.9067 | 48.20706       |

N: number; Std deviation: Standard deviation

## DISCUSSION

The main objective of endodontic treatment is the removal of microorganisms and pulpal remnants from contaminated root canals, and then to adequately seal the disinfected root canals. Intracanal medicaments play a crucial role in destroying the microorganisms; they help prevent the regrowth of bacteria and enhance bacterial suppression [9]. Hereby, these medicaments disinfect the root canal system to prepare it for a biologically compatible obturating material.

Nowadays, CH is extensively used as a root canal medicament, but its removal before obturation is of utmost importance [10]. Penetration of endodontic sealers into the dentinal tubules could naturally refine the sealing of root canals by increasing the contact surface area of materials with instrumented canal walls [11]. Remnants of CH can restrict the penetration of sealers into the dentinal tubules, impede the adhesion of sealers, increase the apical microleakage, and could lead to negative chemical interactions with sealers, making the prognosis uncertain [12]. A study has been conducted to determine the best protocol for eliminating CH from root canals before obturation. The most frequently used technique is the use of master apical file in combination with irrigating solutions. The use of rotary files and sonic and ultra-sonically agitated tips was also recommended [13]. In order to effectively wash out the debris, sonic or ultrasonics increase the agitation and shearing stress of irrigants in the root canal walls [14]. However, none of the irrigant activation methods were able to eliminate CH remnants from the canals, specifically in the apical third [15].

The EndoActivator is an acoustically operated root canal irrigation device with a portable handpiece and disposable flexible polymer tips in various sizes [16]. Alturaiki et al., [17] stated that EndoActivator outperformed the Endovac system in terms of eliminating CH from the coronal, middle, and apical thirds of the root canal system. The ability to remove CH from root canals by EndoActivator in combination with irrigation is because of its primary function to generate vigorous

intracanal fluid agitation through acoustic streaming and cavitations. It enhances the penetration, circulation, and flow of irrigant into more remote sites of root canals [18]. According to Gupta et al., [19] when used in conjunction with various rotary file systems, EndoActivator significantly improves root canal debridement. When it came to removing CH, it performed noticeably better in the coronal third than in the apical third. This can be due to more velocity and an increase in volume of irrigant solution that reaches the coronal portion of the root canal during irrigation [19]. In contrast to traditional needle irrigation, it is hypothesized that EndoActivator promotes a greater flow of irrigant into the accessory canals and apical region.

In accordance with the results obtained in this study, CH cannot be entirely eliminated from the dentinal tubules, irrespective of the irrigants or the irrigation system used, and hence acts as a major hindrance affecting the penetration of sealers. Chelating solutions like EDTA in combination with NaOCl used during canal preparation reduce the accumulation of debris and facilitate CH removal.

CLSM was used to investigate the penetration depth of root canal sealers in this study because it could provide a concise view of the presence and distribution of sealers. Additionally, it promotes the use of fluorescence to visualize the resin-dentin interface and is associated with the penetration of fluorescently tagged sealer [20]. The use of Zen software allowed the calculation of sealer or CH penetration depth without the influence of the autofluorescence signal.

The long-term success of non-surgical root canal therapy is determined by the root canal sealer selection. An ideal sealer shows strong antibacterial activity, creates a microscopic seal that prevents bacteria from passing through the root canals, and accomplishes these objectives without producing cytotoxicity or an inflammatory reaction [21]. As there is minimal chemical adhesion taking place between dentin and sealers, the penetration depth of endodontic sealers is of growing clinical importance. Flow and adhesiveness of the sealer are important, but flowability allows

filling the irregularities, isthmuses, apical deltas, and accessory canals; however, a high flow can lead to apical extrusion, resulting in injury to periapical tissues [22].

Obturation with minimal voids and gaps is the desired prudent outcome of endodontic treatment, which relies on effective surface adaptation and reduced shrinkage of the sealer between the core material and the root dentin. The diameter and density of the dentinal tubules are greater at the coronal and middle thirds, whereas they are minimal at the apical third; this parameter significantly impacts sealer penetration [23].

It has been noted that, irrespective of the type of sealer or obturation technique applied, the percentage of segments with sealer penetration increased from the apical to the coronal third. This phenomenon can be attributed to the higher number and larger diameter of dentinal tubules, and the effective elimination of smear layer in the coronal and middle parts of the root canal [24].

The penetration depth of AH Plus may be because of the sealer integrity and unique property of being drawn by capillary forces into the dentin. AH Plus was chemically cured, which might have permitted compensation for polymerization shrinkage [25]. AH Plus exhibited better penetration in the coronal third than the middle and apical thirds due to its small particle size and film thickness, which is in the range of 20–25µm. AH Plus demonstrates excellent biocompatibility, tissue tolerance, long-term dimensional stability, and sealing ability, while the presence of silicone oil content of the sealer raises surface tension, leading to shrinkage at the sealer–dentin interface [26].

Apexit Plus is an example of CH-containing sealer which has been claimed to stimulate osteogenesis and cementogenesis, and also produces an antimicrobial environment [21]. The slight setting expansion, in association with low solubility, permits proper sealing but it shows low adhesion to dentin because of low cohesion of its molecules [7].

Bioroot RCS, a tricalcium silicate-based sealer, releases high levels of calcium, i.e., almost double as compared to the other sealers. When

it comes in proximity with physiological solutions, it forms an interfacial calcium phosphate layer, forming a chemically stable bond to dentin [27]. When it comes in contact with tissue fluids, it forms calcium phosphate paste and releases osteogenic and angiogenic growth factors, indicating its excellent bioactivity. It interlinks with dentin, forming mineral plugs within the dentinal tubules and thus increasing its bioactivity. This sealer showed the greatest dentinal tubule penetration in all three sections, compared with the other sealers used in this study. Irrespective of the location in the canal, the bioceramic-based root canal sealers show superior performance compared to epoxy resin-based sealers regarding the rate of dentinal penetration, as shown by Shieh et al., [28]. The remarkable performance was attributed to its small particle size, hydrophilic nature, and low contact angle allowing the sealer to spread easily on the dentin and penetrate inside, filling the lateral and accessory micro-canals. It forms a chemical bond to root dentin that contributes to a seamless bond between the sealer and dentinal walls, making it an ideal and efficient sealer [23]. According to Khullar et al., [29] Bioroot RCS demonstrated a noticeably greater depth of penetration than resin-based and CH-based sealers at all root regions. The results of the current study are in line with those of Pius et al., [23] which compared the gap formation and penetrability of AH Plus, Sealapex, and Bioroot RCS root canal sealers into dentin, and revealed that the bioceramic sealer had a superior penetrability and minimum gap formation compared with epoxy resin-based and CH-based sealers. Dsouza et al., [30] studied the depth of penetration of four different sealers using CLSM and reported that Bioroot RCS had greater sealing potential and a higher penetration depth into the dentinal tubules.

As it is an *in vitro* study, it limits the direct clinical applicability of the findings and dentinal tubule penetration alone may not predict the overall clinical performance of root canal sealers. Future *in vivo* and clinical studies are needed to validate these findings and correlate sealer penetration with long term treatment outcomes.

## CONCLUSION

Within the limitations of this in vitro study, the results suggest the use of Bioroot RCS as compared to AH Plus resin-based sealer or Apexit Plus CH-containing sealer after removal of CH medicament from the root canals.

## CONFLICT OF INTEREST STATEMENT

None declared. the authors have no commercial association that might pose a conflict of interest in connection with the submitted article.

## GENERATIVE AI IN SCIENTIFIC WRITING

None declared.

## REFERENCES

1. Iqbal A. Antimicrobial irrigants in the endodontic therapy. *Int J Health Sci.* 2012 Jul;6(2):186-92.
2. Ordinola-Zapata R, Bramante CM, Graeff MS, del Carpio Perochena A, Vivan RR, Camargo EJ, et al. 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.* 2009 Sep;108(3):450-7.
3. Wang Y, Guo LY, Fang HZ, Zou WL, Yang YM, Gao Y, et al. An in vitro study on the efficacy of removing calcium hydroxide from curved root canal systems in root canal therapy. *Int J Oral Sci.* 2017 Jun;9(2):110-6.
4. Pawar SS, Pujar MA, Makandar SD. Evaluation of the apical sealing ability of bioceramic sealer- AH Plus & epiphany: an in vitro study. *J Conserv Dent.* 2014 Nov-Dec;17(6):579-82.
5. Silva RV, Silveira FF, Horta MC, Duarte MA, Cavenago BC, Morais IG, et al. Filling effectiveness and dentinal penetration of endodontic sealers: a stereo and confocal laser scanning microscopy study. *Braz Dent J.* 2015 Sep-Oct;26(5):541-6.
6. Uzunoğlu-Özyürek E, Erdoğan Ö, Türker S. Effect of Calcium Hydroxide Dressing on the Dentinal Tubule Penetration of 2 Different Root Canal Sealers: A Confocal Laser Scanning Microscopic Study. *J Endod.* 2018 Jun;44(6):1030-5.
7. Haragushiku GA, Sousa-Neto MD, Silva-Sousa YT, Alfredo E, Silva SC, Silva RG. Adhesion of endodontic sealers to human root dentine submitted to different surface treatments. *Photomed Laser Surg.* 2010 Jun;28(3):405-10.
8. Siboni F, Taddei P, Zamparini F, Prati C, Gandolfi MG. Properties of BioRoot RCS, a tricalcium silicate endodontic sealer modified with povidone and polycarboxylate. *Int Endod J.* 2017 Dec;50 Suppl 2:e120-e136.
9. Kumar A, Tamanna S, Iftekhhar H. Intracanal medicaments – Their use in modern endodontics: a narrative review. *J Oral Res Rev.* 2019 Jul-Dec;11:94-9.
10. Ba-Hattab R, Al-Jamie M, Aldreib H, Alessa L, Alonazi M. Calcium hydroxide in endodontics: an overview. *Open J Stomatol.* 2016 Dec;6(12):274-89.
11. De-Deus G, Gurgel-Filho ED, Maniglia-Ferreira C, Coutinho-Filho T. Influence of the filling technique on depth of tubular penetration of root canal sealer: scanning electron microscopy study. *Braz J Oral Sci.* 2004 Jan;3(9):433-8.
12. Aayushi A, Dax R. Calcium hydroxide in endodontics. *J Appl Dent Med Sci.* 2016 Jul-Sep;2(3):78-88.
13. Khalap ND, Kokate S, Hegde V. Ultrasonic versus sonic activation of the final irrigant in root canals instrumented with rotary/reciprocating files: an in-vitro scanning electron microscopy analysis. *J Conserv Dent.* 2016 Jul-Aug;19(4):368-72.
14. Plotino G, Cortese T, Grande NM, Leonardi DP, Di Giorgio G, Testarelli L, et al. New technologies to improve root canal disinfection. *Braz Dent J.* 2016 Jan-Feb;27(1):3-8.
15. Taşdemir T, Celik D, Er K, Yildirim T, Ceyhanli KT, Yeşilyurt C. Efficacy of several techniques for the removal of calcium hydroxide medicament from root canals. *Int Endod J.* 2011 Jun;44(6):505-9.
16. Torres D, González-Rodríguez MP, Ferrer-Luque CM. Effectiveness of Endoactivator System in removing the smear layer after root canal instrumentation. *J Endod.* 2010 Feb;36(2):308-11.
17. Alturaiki S, Lamphon H, Edrees H, Ahlquist M. Efficacy of 3 different irrigation systems on removal of calcium hydroxide from the root canal: a scanning electron microscopic study. *J Endod.* 2015 Jan;41(1):97-101.
18. Parikh M, Kishan KV, Solanki NP, Parikh M, Savaliya K, Bindu VH, et al. Efficacy of removal of calcium hydroxide medicament from root canals by endoactivator and endovac irrigation techniques: a systematic review of in vitro studies. *Contemp Clin Dent.* 2019 Jan-Mar;10(1):135-42.
19. Gupta R, Sharma H, Kumari RA, Prakash AC, Rai N, Jain L. Effectiveness of two techniques in removal of calcium hydroxide medicament from root canals: an in-vitro assessment. *J Clin Diagn Res.* 2018 Jul;12(7):ZC53-5.

20. Bharti R, Tikku AP, Chandra A, Shakya VK, Yadav S. Depth and percentage of resin-based sealer penetration inside the dentinal tubules using Endovac, Endoactivator, Navi tip FX irrigation system: a confocal laser scanning microscope study. *J Conserv Dent*. 2018 Mar-Apr;21(2):216-20.
21. Komabayashi T, Colmenar D, Cvach N, Bhat A, Primus C, Imai Y. Comprehensive review of current endodontic sealers. *Dent Mater J*. 2020 Sep;39(5):703-20.
22. Rai RU, Singbal KP, Parekh V. The effect of temperature on rheological properties of endodontic sealers. *J Conserv Dent*. 2016 Mar-Apr;19(2):116-9.
23. Pius A, Mathew J, Theruvil R, George S, Paul M, Baby A, et al. In vitro study of dentinal tubule penetration and filling quality of bioceramic sealer. *Cons Dent Endod J*. 2019 Jan-Jun;4:6-13.
24. Wang Y, Liu S, Dong Y. In vitro study of dentinal tubule penetration and filling quality of bioceramic sealer. *PLoS One*. 2018 Feb;13(2):e0192248.
25. Sonu KR, Girish TN, Ponnappa KC, Kishan KV, Thameem PK. Comparative evaluation of dentinal penetration of three different endodontic sealers with and without smear layer removal – scanning electron microscopic study. *Saudi Endod J*. 2016 Jan-Apr;6(1):16-20.
26. Chen H, Zhao X, Qiu Y, Xu D, Cui L, Wu B. The tubular penetration depth and adaption of four sealers: a scanning electron microscopic study. *Biomed Res Int*. 2017;2017:2946524.
27. Arikatla SK, Chalasani U, Mandava J, Yelisela RK. Interfacial adaptation and penetration depth of bioceramic endodontic sealers. *J Conserv Dent*. 2018 Jul-Aug;21(4):373-7.
28. Shieh K, Yang J, Zhu EH, Peters OA, Hosseinpour S. Dentinal tubule penetrability and bond strength of two novel calcium silicate-based root canal sealers. *Materials*. 2023 Apr;16(9):3309.
29. Khullar S, Aggarwal A, Chhina H, Kaur T, Sharma M, Bala D. Sealer penetration in the dentinal tubules: a confocal laser scanning microscopy study. *Endodontology*. 2021 Apr;33(2):92-6.
30. Dsouza AP, Suvarna N, Shetty SH, Farhana F, Syed AA. To compare and evaluate the sealing ability of Guttaflow bioseal, Bioroot RCS and MTA fillapex with AH plus: an in vitro study. *IOSR-JDMS*. 2020 Aug;9(8):43-9.