Original Article

DOI: 10.7860/JCDR/2026/85961.24254

Dentistry Section

# Effect of Sonic versus Ultrasonic Irrigant Activation on Dentinal Tubule Penetration of AH Plus Bioceramic Sealer and ReGen Bioactive Endosealer: An In-vitro Confocal Laser Scanning Microscopy Study

SAI MEGHANA BAPATU YERUGUTI[1], ROOPADEVI GARLAPATI[2], NAGESH BOLLA[3], SAYESH VEMURI[4], RAM SUNIL CHUKKA[5], ANILA BANDLAPALLY SREENIVASA GUPTHA[6]



## ABSTRACT

**Introduction:** Successful root canal therapy relies on thorough canal disinfection and the ability of sealers to penetrate the dentinal tubules effectively. Although mechanical shaping and irrigation lower bacterial presence, irrigant activation, especially using sonic or ultrasonic devices, boosts smear layer elimination and overall cleaning. The clinical success of Bioceramic Sealers (BCS) depends on their deep penetration into dentinal tubules for a fluid-tight seal. However, there is limited comparative evidence on whether sonic or ultrasonic activation better enhances intratubular adaptation of newer sealers like AH Plus Bioceramic and ReGen Bioactive Endosealer. This study uses Confocal Laser Scanning Microscopy (CLSM) to evaluate and compare these methods and aims to provide a standardised basis for selecting optimal irrigation activation protocols.

**Aim:** To evaluate and compare the maximum depth of penetration of AH Plus BCS (AHPB) and ReGen Bioactive Endosealer sealer after sonic and ultrasonic activation

**Materials and Methods:** The present in-vitro study was conducted in the Department of Conservative Dentistry and Endodontics at Sibar Institute of Dental Sciences in Guntur, Andhra Pradesh, India, from July 2023 to September 2024. A total of 60 extracted human mandibular premolars with single roots were extracted for reasons related to periodontal or orthodontic reasons. Standard access cavities were prepared, followed by Working Length (WL) determination, and shaping was done with ProTaper Gold files up to size F3. Samples were randomly assigned to four experimental groups (n=15) depending on the combination of sealer and activation method: Group-1 (SLP-EA + AHPB), Group-2 (SLP-EA + ReGen Bioactive Endosealer), Group-3 (Ultra X + AHPB), and Group-4 (Ultra X + ReGen Bioactive Endosealer). Post-irrigation and activation, root canals were filled using Rhodamine B-stained sealers along with gutta-percha. Samples were then sliced at the middle and apical thirds and examined using Confocal Laser Scanning Microscopy (CLSM). Statistical analysis was performed using One-way Analysis of Variance (ANOVA), significant value was set at $p<0.05$, followed by Tukey's post-hoc test.

**Results:** A statistically significant difference ($p<0.05$) in maximum sealer penetration was identified in the middle third, with Group 3 (Ultra X + AHPB) showing the greatest depth, followed by Groups 1, 4 and 2. No significant differences were found at the apical third, though AHPB-based groups showed marginally deeper penetration.

**Conclusion:** AH Plus Bioceramic Sealer demonstrated greater dentinal tubule penetration than ReGen Bioactive Endosealer, particularly when used with ultrasonic activation. These findings indicate that both the sealer type and the irrigant activation technique influence dentinal tubule penetration under the experimental conditions of this in-vitro study.

**Keywords:** Endoactivator, Irrigation, Smear layer

## INTRODUCTION

The core objective of root canal treatment is to eradicate microorganisms from the root canal system and to prevent any chances of reinfection [1]. Though mechanical instrumentation eliminates most of the pulp tissue, some canal surfaces remain untouched and are coated by a smear layer [2]. This smear layer, typically measuring 1-2 µm in thickness [3], inhibits the penetration of sealers and is commonly removed using Ethylenediaminetetraacetic Acid (EDTA) following Sodium hypochlorite (NaOCl) irrigation. Sealers are then able to enter the dentinal tubules, with reported penetration depths ranging from 98-1490 µm, most notably in cervical and middle thirds [4].

Because of the intricate anatomy of the apical portion, achieving full debridement through instrumentation alone remains difficult. The efficiency of irrigants is influenced by factors like the size of the apical preparation, the presence of a vapour lock, and the irrigation technique employed [1]. Traditional needle irrigation has limitations, prompting the adoption of irrigant activation methods to improve cleaning and debris removal [5]. Sonic and ultrasonic activations enhance the flow and energy of irrigants, aiding in smear layer elimination and canal cleanliness [6]. Sonic systems, such as the EndoActivator, function at lower frequencies generates fluid movement and acoustic streaming [7]. According to Kanter V et al., their flexible polymer tips help reduce iatrogenic damage during use [8]. They are also effective in increasing sealer penetration into lateral canals and dentinal tubules [9].

The Smart Lite Pro EndoActivator (SLP-EA) uses elliptical motion through its polymer tips to improve irrigant movement. With a cordless design, ergonomic build, and a sonic agitation speed of 18,000 rpm, it is an advanced sonic activation tool [10]. Ultrasonic irrigation devices operate "usually within the 25-30 kHz range", producing cavitation and acoustic microstreaming due to oscillation of the ultrasonic file [11].

Endodontic sealers are classified into zinc oxide-eugenol, resin-based, and bioceramic categories. BCS, composed of calcium silicates, phosphates, and zirconium oxide, are hydrophilic, chemically bonds to dentin, and is bioactive [12]. Their enhanced sealing capability aids in bacterial inhibition and long-term obturation success [13]. Products like MTA Fillapex, BioRoot RCS, and EndoSequence BC Sealer exhibit greater tubule infiltration than AH Plus due to smaller particle size and hydrophilic nature. AHPB and ReGen Bioactive Endosealer Sealer are recent bioceramic formulations with improved biocompatibility, alkaline pH, and adaptability to tubules [14].

The CLSM enables evaluation of sealer penetration with fluorescent labelling, without altering the setting characteristics [15]. Greater depth of penetration is associated with better sealing and antimicrobial efficacy [16]. However, no prior research has compared the penetration of AHPB and ReGen Bioactive Endosealer sealers activated by both SLP-EA and Ultra X systems. Hence, this in-vitro study aimed to evaluate and compare the maximum penetration depth of these two sealers in the middle and apical thirds when subjected to sonic and ultrasonic activation. The comparison helps determine the most effective sealer-activation combination for improved tubular sealing and better endodontic outcomes. The primary objective of the study was to evaluate and compare the maximum depth of penetration of AHPB and ReGen Bioactive Endosealer following activation with SLP EndoActivator and Ultra X at the middle and apical third regions of the root canal.

**Null hypothesis:** There is no significant difference in the depth of dentinal tubule penetration between AHPB and ReGen Bioactive Endosealer, irrespective of the activation method (sonic or ultrasonic) or the root canal level (middle and apical thirds).

**Alternate hypothesis:** There is a significant difference in the depth of dentinal tubule penetration between APB and ReGen Bioactive Endosealer, influenced by the activation method (sonic or ultrasonic) and/or the root canal level (middle and apical thirds).

## MATERIALS AND METHODS

The present in-vitro study was conducted in the Department of Conservative Dentistry and Endodontics at Sibar Institute of Dental Sciences in Guntur, Andhra Pradesh, India, from July 2023 to September 2024. The study received ethical clearance from the Institutional Ethics Committee (Ref No: Pr.197/IEC/SIBAR/2023).

**Sample size calculation:** Sample size estimation was calculated with an effect size of 0.6, an alpha level of 0.05, and a desired statistical power of 95%. The minimum required sample size was calculated to be 60 specimens using G*Power software (version 3.1.9.2) [17]. These teeth were then randomly divided into four experimental groups, with each group containing 15 specimens. Grouping was based on the combination of irrigation activation method and the type of BCS used.

**Inclusion criteria:** Teeth with complete crown structure, fully formed roots, and patent root canals were selected for the study.

**Exclusion criteria:** Carious teeth; teeth with fractures or visible cracks; teeth exhibiting internal or external resorption; calcified teeth; teeth with previous restorations or prior root canal treatment; and teeth with developmental anomalies were excluded.

### Study Procedure

Sixty recently extracted human mandibular premolars with single canals (n=60) were selected for the study. The external surfaces were cleaned using an ultrasonic scaler to eliminate any calculus or remaining soft tissue, and the teeth were stored in a 0.1% thymol solution until required. Intraoral Periapical (IOPA) radiographs were taken from both mesiodistal and buccolingual angles to confirm the presence of a single canal.

Standard access cavities were prepared, and canal patency was established using ISO #10 stainless steel K-files, advancing until the file tip was visible at the apical foramen. The WL was defined as 1 mm short of the apex and verified radiographically. Root canal instrumentation was completed using the ProTaper Gold rotary file system (Sx to F3) along with RC Help as a lubricant. Final apical preparation was standardised to size 35 using a K-file, as K-files are commonly used for apical enlargement because they allow better control of WL and facilitate effective debridement and irrigant penetration in the apical third [18]. Throughout the instrumentation, 2 mL of 5.25% sodium hypochlorite was used for irrigation after each file, followed by saline.

**Irrigation protocol and group distribution:** A standardised final irrigation regimen was followed for all samples: 5 mL of 5.25% NaOCl for 60 seconds, 5 mL of 17% EDTA for 60 seconds, and a final rinse with 10 mL of distilled water. Sixty samples were randomly divided into four groups of n=15:

**Group 1:** SLP-EA (sonic) + AH Plus Bioceramic Sealer (AHPB)

**Group 2:** SLP-EA (sonic) + ReGen Bioactive Endosealer

**Group 3:** Ultra X (ultrasonic) + AH Plus Bioceramic Sealer (AHPB)

**Group 4:** Ultra X (ultrasonic) + ReGen Bioactive Endosealer

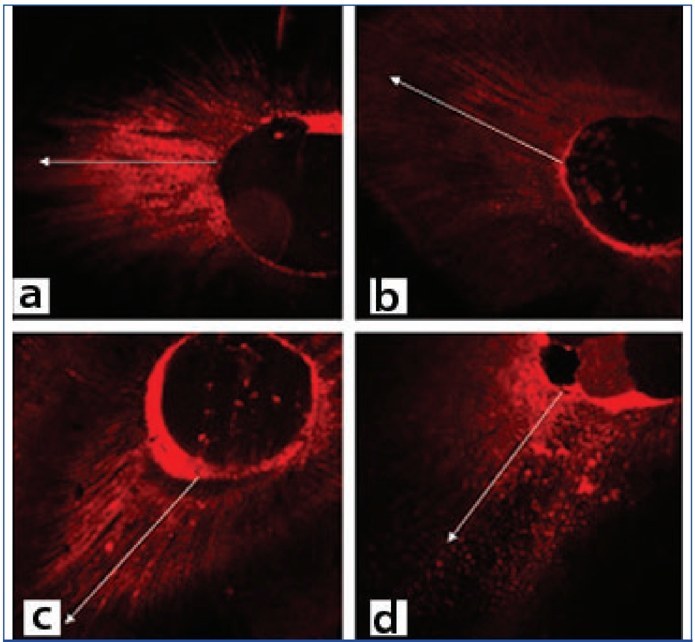
The sonic device, SLP-EA, was used with a red polymer tip (25/04), positioned 2 mm short of WL, and activated for 60 seconds at 18,000 cycles per minute in 3D mode [7,10]. Ultra X was used for ultrasonic activation with identical parameters. Additionally, EDTA was activated for 30 seconds in both approaches [19,20].

Each sealer was mixed with Rhodamine B dye. A lentulospiral was used to deliver the sealer into canals, followed by lateral condensation with gutta-percha. Cavit was used to seal the access cavities. Specimens were then stored at 37°C and 95% relative humidity for one week to simulate clinical conditions [Table/Fig-1] [19].

**[Table/Fig-1]:** Specimens stored in a humidifier at 37°C.

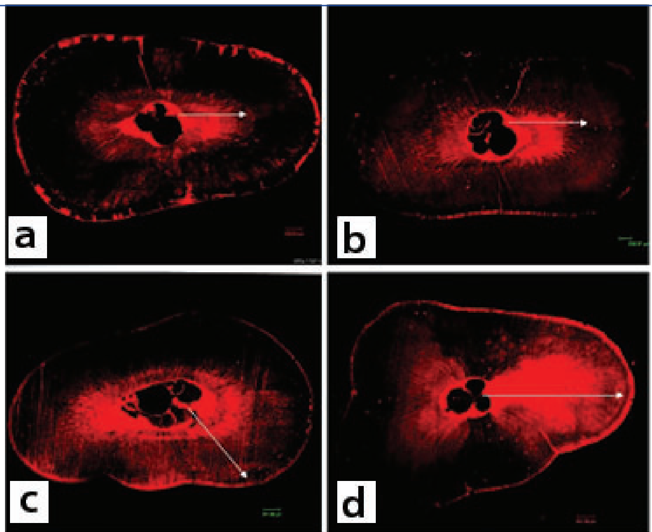
### Sectioning and CLSM Evaluation

Each root was embedded in acrylic resin and sectioned horizontally using a hard tissue microtome to obtain 2 mm slices at two levels: 5 mm (middle third) and 3 mm (apical third) from the apex. These slices were evaluated under CLSM to assess the maximum depth of sealer penetration (µm). Penetration depth was measured from the canal wall to the deepest point of the fluorescent signal under CLSM. The maximum depth of root canal sealer penetration was measured in micrometres (µm) from the root canal wall to the point of deepest sealer penetration. No image analysis software or scoring criteria were used; measurements were recorded directly as quantitative values. Penetration was assessed at the middle third [Table/Fig-2]




**[Table/Fig-2]:** CLSM images of sealers penetration after sonic and ultrasonic activation in the middle third: a) Ultra-X with AH Plus Bioceramic sealer (AHPB); b) SLP-EA with AH Plus Bioceramic sealer (AHPB); c) Ultra-X with ReGen Bioactive Endosealer bioactive endosealer; d) SLP-EA with ReGen Bioactive Endosealer bioactive endosealer.

and apical third [Table/Fig-3] by measuring the maximum linear depth (µm) from the canal wall to the deepest fluorescent signal under CLSM using the microscope's calibrated scale.




**[Table/Fig-3]:** CLSM images of sealers penetration after sonic and ultrasonic activation in the apical third: a) Ultra-X with ReGen Bioactive Endosealer bioactive endosealer; b) SLP-EA with ReGen Bioactive Endosealer bioactive endosealer; c) SLP-EA with AH Plus Bioceramic sealer (AHPB); d) Ultra-X with AH Plus Bioceramic sealer (AHPB).

Blinding was not performed; CLSM evaluation was conducted by a single trained examiner under standardised and calibrated conditions to ensure measurement consistency and reduce bias. The measurements were performed by a single examiner to avoid intra-examiner variability. Repeated measurements were taken for each section, and the mean value was calculated to enhance reliability and minimise observational error. EDTA was activated for 30 seconds in accordance with smear layer removal protocols to minimise dentinal erosion [20]. Sealers were activated for 60 seconds to enhance flow and maximise dentinal tubule penetration through improved agitation and distribution [21]. Specimens were stored at 37°C and 95% humidity to simulate intraoral conditions and ensure complete sealer setting before analysis [17]. A one-week storage period is commonly adopted in in-vitro endodontic studies to allow full material maturation and stabilisation before testing [17].

## STATISTICAL ANALYSIS

Data were analysed using Statistical Package for the Social Science (SPSS), IBM Corporation Ltd., Armonk, New York, USA, version 26.0. Data normality was evaluated using the Shapiro-Wilk test. The mean and Standard Deviation (SD) were used to depict descriptive statistics. One-way ANOVA test was used for intergroup comparison ($p<0.05$ considered significant). Comparison of maximum depth of sealer penetration (µm) between the four groups in the middle third region using Tukey's post-hoc test.

## RESULTS

The comparative data on maximum sealer penetration (µm) among the four subgroups in the middle third of the canal are presented in [Table/Fig-4]. Statistically significant differences were found, with AHPB achieving greater depth than ReGen Bioactive Endosealer, and ultrasonic activation producing better results than sonic activation.

In the apical third, all groups exhibited comparable mean penetration values, and the differences observed were not statistically significant [Table/Fig-5]. The apical third ($p=0.068$) showed a trend toward significance but did not reach the 0.05 threshold. This may be due to anatomical complexities such as lower tubule density and narrower canals, which limit penetration and increase variability. A comparison of the maximum depth of sealer penetration (µm) between the four groups in the middle third region using Tukey's post-hoc test are represented in [Table/Fig-6].

## DISCUSSION

The primary goal of endodontic therapy is to create a hermetic seal within the root canal system using a combination of core material and endodontic sealer. An ideal seal cannot be achieved with solid core material alone; sealers are essential for three-dimensional obturation. The smear layer hinders intracanal medicaments and sealers from penetrating dentinal tubules [22]. Its removal allows sealers to form a tight barrier and mechanically interlock with root dentin.

Sealer penetration depends on factors like canal size, tubule density, and the sealer's physicochemical properties especially flow [23]. Pseudoplastic sealers decrease in viscosity with increased shear rate, enhancing penetration. This study evaluated the penetration depths of AHPB and ReGen Bioactive Endosealer sealers activated using SLP-EA (sonic) and Ultra X (ultrasonic) in the middle and apical thirds. Group 3 (Ultra X + AHPB) showed the greatest penetration, followed by Group 1 (SLP-EA + AHPB), Group 4 (Ultra X + ReGen

| | | | 95% confidence interval for mean | | | |
|---|---|---|---|---|---|---|
| Group middle third | Mean±SD | Std. error | Lower bound | Upper bound | F value | p-value |
| Group 1: AH Plus + Sonic (SLP-EA) | 1122.0673±100.48146 | 25.94420 | 1066.4226 | 1177.7121 | 9.286 | <0.001* |
| Group 3: AH Plus + Ultrasonic (Ultra X) | 1128.0793±175.44850 | 45.30061 | 1030.9192 | 1225.2395 | | |
| Group 2: ReGen Bioactive Endosealer + Sonic (SLP-EA) | 878.6720±159.94631 | 41.29796 | 790.0967 | 967.2473 | | |
| Group 4: ReGen Bioactive Endosealer + Ultrasonic (Ultra X) | 993.3727±157.24526 | 40.60055 | 906.2931 | 1080.4522 | | |

**[Table/Fig-4]:** Comparison of maximum depth of sealer penetration (µm) between the four groups in the middle third region using ANOVA (N=15).
Test: One-way Analysis of Variance; p≤0.05 considered statistically significant; *denotes significance; p-value: 0.001*, F-value: 9.286

| Group apical third | Mean±SD | Std. error | 95% Confidence interval for mean | | F value | p-value |
|---|---|---|---|---|---|---|
| | | | Lower bound | Upper bound | | |
| Group 1: AH Plus + Sonic (SLP-EA) | 786.4113±217.47291 | 56.15126 | 665.9789 | 906.8438 | 2.507 | 0.068 |
| Group 3: AH Plus + Ultrasonic (Ultra X) | 875.1620±139.71149 | 36.07335 | 797.7924 | 952.5316 | | |
| Group 2: ReGen Bioactive Endosealer + Sonic (SLP-EA) | 721.4693±114.54276 | 29.57481 | 658.0377 | 784.9010 | | |
| Group 4: ReGen Bioactive Endosealer + Ultrasonic (Ultra X) | 759.9787±148.69838 | 38.39376 | 677.6322 | 842.3251 | | |

**[Table/Fig-5]:** Comparison of maximum depth of sealer penetration (µm) between the four groups in the apical third region using ANOVA (N=15).
Test: One-way Analysis of Variance; p≤0.05 considered statistically significant; * denotes no significance; p-value: 0.068, F-value: 2.507

| Groups | | Mean Difference | SE | p-value |
|---|---|---|---|---|
| Group 1: AH Plus + Sonic (SLP-EA) | Group 2: (ReGen sealer + Sonic) | 243.39533* | 55.13244 | 0.000* |
| | Group 3: (AHPB + Ultrasonic) | -6.01200 | 55.13244 | 1.000# |
| | Group 4: (ReGen sealer + Ultrasonic) | 128.69467 | 55.13244 | 0.139 |
| Group 2: (ReGen sealer + Sonic) | Group 1: AH Plus + Sonic (SLP-EA) | -243.39533* | 55.13244 | 0.000* |
| | Group 3: (AHPB + Ultrasonic) | -249.40733* | 55.13244 | 0.000* |
| | Group 4: (ReGen Bioactive Endosealer sealer + Ultrasonic) | -114.70067 | 55.13244 | 0.252 |
| Group 3: (AHPB + Ultrasonic) | Group 1: AH Plus + Sonic (SLP-EA) | 6.01200 | 55.13244 | 1.000 |
| | Group 2: (ReGen Bioactive Endosealer sealer + Sonic) | 249.40733* | 55.13244 | 0.000* |
| | Group 4: (ReGen Bioactive Endosealer sealer + Ultrasonic) | 134.70667 | 55.13244 | 0.106 |
| Group 4: (ReGen Bioactive Endosealer sealer + Ultrasonic) | Group 1: AH Plus + Sonic (SLP-EA) | -128.69467 | 55.13244 | 0.139 |
| | Group 2: (ReGen Bioactive Endosealer sealer + Sonic) | 114.70067 | 55.13244 | 0.252 |
| | Group 3: (AHPB + Ultrasonic) | -134.70667 | 55.13244 | 0.106 |

**[Table/Fig-6]:** Comparison of maximum depth of sealer penetration (µm) between the four groups in the middle third region using post-hoc test.
#The p-value of 1.000 is the Bonferroni-adjusted value from the post-hoc test; when the adjusted p-value exceeds 1, statistical software reports it as 1.000. Hence, it reflects a non significant difference rather than identical mean and SD between the groups.

Bioactive Endosealer) and Group 2 (SLP-EA + ReGen Bioactive Endosealer) in the middle and apical third regions. Greater middle third penetration was due to wider tubules and improved irrigant flow. The null hypothesis was rejected in terms of the middle third region, as statistically significant differences were observed between the groups, with AHPB demonstrating greater depth of penetration than ReGen Bioactive Endosealer, and ultrasonic activation producing superior results compared to sonic activation. The null hypothesis was accepted, indicating that there was no statistically significant difference in the maximum depth of sealer penetration (µm) between the four groups in the apical third region.

Ultrasonic groups generally showed deeper penetration than sonic groups. These findings align with Lee SJ et al., who reported ultrasonic irrigation's superiority over needle irrigation [24]. Enhanced irrigant flow and acoustic streaming from ultrasonic oscillation explain this efficacy. Burleson A et al., and Ahmad M et al., also reported better apical cleaning via ultrasonic activation [25,26]. Siu C and Van der Sluis JC described how ultrasonic devices create acoustic nodes, increasing streaming and cavitation effects [27].

Ferreira RB et al., study confirmed ultrasonic superiority by ultrasonic-induced turbulence, cavitation, and thermal enhancement, which improved smear layer removal and tubule penetration [28]. This may be attributed to poor irrigant flow in the apical canal and better smear layer clearance in the coronal and middle thirds. Both the number and diameter of dentinal tubules decrease from the coronal toward the apical third, which limits sealer penetration [29].

Higher concentrations of sodium hypochlorite (such as 5.25%) are commonly used in endodontic irrigation because of their superior antimicrobial activity and tissue dissolution capacity; however, high concentrations may adversely affect dentin structure and mechanical properties. This effect has been reported to reduce dentin micro-hardness and fracture strength and to cause degradation of the intratubular dentin walls [30].

Sealer diffusion was maximum in the coronal section, followed by the middle and apical sections due to the higher density and larger diameter of dentinal tubules coronally and the presence of sclerotic dentin and occluded tubules toward the apical region, which reduces penetration [31]. Studies by Akcay M et al., confirm decreasing penetration toward the apex [32]. Sunny G et al., and Jordan et al., reported significantly higher penetration at 5 mm compared to 1 mm from the apex [33]. CLSM and SEM findings by Oliveira JS and Ugur Aydin Z et al., also reflect this trend [34,35]. Ultrasonic activation promotes deeper sealer penetration than manual or sonic methods due to acoustic streaming and cavitation [4,35-37]. De Gregorio C et al., confirmed improved smear layer removal with higher ultrasonic intensity [38]. Jiang LM et al., demonstrated better irrigant flow and dye penetration in ultrasonic groups [39]. Ateş AA et al., also observed increased sealer depth at 5 mm over 2 mm [40].

Dentinal tubules taper from the pulpal surface toward the outer dentin, with diameters of approximately 1-2 µm near the pulp and less than 1 µm near the DEJ/CEJ, resulting in reduced dentin permeability toward the outer dentin and apical regions [41]. Clinically, improved sealer penetration boosts antimicrobial action and obturation quality, especially with antibacterial sealers [42,43].

In the current study, the four-wall access cavity retained more irrigant, enhancing activation, unlike crown-removed samples. Activation caused apical fluid movement and irrigant displacement, improving solution exchange. CLSM enabled precise, non-destructive visualisation of dye-labeled sealers in dentinal tubules [44]. Greater sealer penetration, crucial for minimising microleakage and reinfection, was observed in AHPB groups (Group-1 and 3) compared to ReGen Bioactive Endosealer groups (Group-2 and 4).

AHPB's better flow, wettability, and smaller particle size contributed to this [45]. Hamdy TM et al., reported that AHPB exhibited a film thickness of 20±1 μm, demonstrating its ability to form a thin sealer layer that may improve adaptation to canal walls [45].

Consistent canal preparation and smear layer removal minimised bias, strengthening validity. CLSM enabled accurate 3D penetration measurement. Better penetration was seen in the middle third due to larger, denser tubules; apical thirds posed challenges due to sclerosis and narrowing [46,47]. Ultrasonic activation improved penetration via acoustic streaming and smear layer removal [48,49].

### Limitation(s)

The primary drawback of the current study was its inability to produce a dynamic oral environment that in-vitro circumstances could not precisely and completely replace with limited materials/devices, and short-term evaluation. Still, results suggest that AHPB with ultrasonic activation offers enhanced sealing. Future in-vivo studies should explore long-term clinical outcomes and sealer-irrigant interactions.

**Clinical implications and future perspectives:** Ultrasonic activation, particularly with AHPB, may enhance dentinal tubule penetration and potentially improve the quality of obturation. Future in vivo studies with larger sample sizes and long-term clinical evaluation are recommended to determine the true clinical significance of these findings.

## CONCLUSION(S)

Within the limitations of this CLSM study, AHPB showed the greatest depth of sealer penetration compared to ReGen Bioactive Endosealer. Overall, AHPB with ultrasonic activation achieved the highest sealer penetration when compared to sonic activation, notably in the middle and apical thirds.

## REFERENCES

[1] Ismail PM, Ahamed S, Sabiha PB, Sekhar MC, Moosani G, Reddy SN, et al. Comparison of sealer penetration by using different irrigation techniques–an In-vitro study. J Clin Diagn Res. 2016;10(12):ZC50.
[2] Paqué F, Ganahl D, Peters OA. Effects of root canal preparation on apical geometry assessed by micro–computed tomography. J Endod. 2009;35(7):1056-59.
[3] Likhitkar MS, Kulkarni SV, Burande A, Solanke V, Kumar CS, Kamble B. To evaluate the influence of smear layer with different instruments and obturation methods on microleakage of root canal filled teeth: In vitro study. J Int Soc Prev Community Dent. 2016;6(3):240-44.
[4] Mamootil K, Messer HH. Penetration of dentinal tubules by endodontic sealer cements in extracted teeth and in vivo. Int Endod J. 2007;40(11):873-81.
[5] Haapasalo M, Shen Y, Wang Z, Gao Y. Irrigation in endodontics. Br Dent J. 2014;216(6):299-303.
[6] Hachem CE, Nehme W, Kaloustian MK, Ghosn N, Daou M, Zogheib C, et al. The effectiveness of different irrigation techniques on debris and smear layer removal in primary mandibular second molars: an in vitro study. J Contemp Dent Pract. 2022;23(11):1173-79.
[7] SmartLite Pro EndoActivator [Internet]. Available from: https://www.dentsplysirona.com/en-in/discover/discover-by-brand/smartlite-pro-endoactivator.html.
[8] Kanter V, Weldon E, Nair U, Varella C, Kanter K, Anusavice K, et al. A quantitative and qualitative analysis of ultrasonic versus sonic endodontic systems on canal cleanliness and obturation. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2011;112(6):809-13.
[9] Guimarães BM, Amoroso-Silva PA, Alcalde MP, Marciano MA, de Andrade FB, Duarte MA. Influence of ultrasonic activation of 4 root canal sealers on the filling quality. J Endod. 2014;40(7):964-68.
[10] Basam RC, Bolla N, Cross R, Vemuri S, Bolla L, Vajrala NR. Evaluation of efficacy of SmartLite Pro EndoActivator sonic activation using adenosine triphosphate bioluminescence meter: A randomized clinical trial. J Conserv Dent Endod. 2025;28(7):619-25.
[11] John S. Comparative evaluation of ultrasonic and sonic activation techniques on irrigant penetration and debris removal in complex root canal systems. IJPBR [Internet]. 31Dec.2020 [cited 13Mar.2026];8(04):17-0.
[12] Al-Haddad A, Che Ab Aziz ZA. Bioceramic-based root canal sealers: A review. Int J Biomater. 2016;2016:9753210.
[13] Sjogren U, Hagglund B, Sundqvist G, Wing K. Factors affecting the long-term results of endodontic treatment. J Endod. 1990;16(10):498-504.
[14] Dentsply Sirona. AH Plus Bioceramic Sealer – International Safety Sheet [Internet]. [cited 2025 May 8]. Available from: https://assets.dentsplysirona.com/master/regions-countries/north-america/usa/product-categories/endodontics/END-EN-US-Document-international-safety-sheet-AH-Plus-Bioceramic-Sealer-NAM.pdf.
[15] Watson TF. Fact and artefact in confocal microscopy. Adv Dent Res. 1997;11(4):433-41.
[16] 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;108(3):450-57.
[17] Örs SA, Yılmaz GA, Şengül N, Keleş A, Eren SK. Effect of final irrigation protocols on the bond strength and dentinal tubule penetration of root canal sealers: an ex vivo laboratory study. BMC Oral Health. 2026;26(1):216.
[18] Schäfer E, Schlingemann R. Efficiency of rotary nickel-titanium K3 instruments compared with stainless steel hand K-Flexofile. Part 2. Cleaning effectiveness and shaping ability in severely curved root canals of extracted teeth. Int Endod J. 2003;36(3):208-17.
[19] Machado R, Cruz ATG, de Araujo BMM, Klemz AA, Klug HP, da Silva Neto UX. Tubular dentin sealer penetration after different final irrigation protocols: A confocal laser scanning microscopy study. Microsc Res Tech. 2018;81(6):649-54.
[20] Perez De Arce CVI, Rodríguez OPA, Echeverri CD. Sonic versus ultrasonic activation of 10% EDTA for smear layer removal in the root canal apical third. Int J Odontostomat. 2014;8(1):153-59.
[21] van der Sluis LWM, Versluis M, Wu MK, Wesselink PR. Passive ultrasonic irrigation of the root canal: a review of the literature. Int Endod J. 2007;40(6):415-26.
[22] Violich DR, Chandler NP. The smear layer in endodontics - a review. Int Endod J. 2010;43(1):2-15.
[23] Ch T, Shaik I, Khan MM, Parvekar P, Mirza MB, Mustafa M, et al. The sealer penetration into the dentinal tubules: An appraisal of different irrigation systems: Original research. J Pharm Bioallied Sci. 2021;13(Suppl 2):S1280-S1285.
[24] Lee SJ, Wu MK, Wesselink PR. The effectiveness of syringe irrigation and ultrasonics to remove debris from simulated irregularities within prepared root canal walls. Int Endod J. 2004;37(10):672-78.
[25] Burleson A, Nusstein J, Reader A, Beck M. The in vivo evaluation of hand/rotary/ultrasound instrumentation in necrotic, human mandibular molars. J Endod. 2007;33(7):782-87.
[26] Ahmad M, Pitt Ford TR, Crum LA. Ultrasonic debridement of root canals: acoustic streaming and its possible role. J Endod. 1987;13(10):490-99.
[27] Siu C, Baumgartner JC. Comparison of the debridement efficacy of the EndoActivator system and passive ultrasonic irrigation in curved root canals. J Endod. 2010;36(11):1785-88.
[28] Ferreira RB, Alfredo E, Porto de Arruda M, Silva Sousa YT, Sousa-Neto MD. Histological analysis of the cleaning capacity of nickel-titanium rotary instrumentation with ultrasonic irrigation in root canals. Aust Endod J. 2004;30(2):56-58.
[29] Mjör IA, Smith MR, Ferrari M, Mannocci F. The structure of dentine in the apical region of human teeth. Int Endod J. 2001;34(5):346-53.
[30] Xu H, Ye Z, Zhang A, Lin F, Fu J, Fok ASL. Effects of concentration of sodium hypochlorite as an endodontic irrigant on the mechanical and structural properties of root dentine: A laboratory study. Int Endod J. 2022;55(10):1091-102.
[31] D'Costa VF, Rodrigues AL, Bangera MK, Bhat PA, Rai RU. A confocal microscopic study on percentage penetration of different sealers into dentin. J Pharm Bioallied Sci. 2021;13(Suppl 1):S725-S730.
[32] Akcay M, Arslan H, Durmus N, Mese M, Capar ID. Dentinal tubule penetration of AH Plus, iRoot SP, MTA Fillapex, and GuttaFlow Bioseal root canal sealers after different final irrigation procedures: A confocal microscopic study. Lasers Surg Med. 2016;48:70-76.
[33] Sunny G, Doddwad PK, Shenvi S. Comparative evaluation of effect of N-acetyl cysteine, maleic acid, and ethylenediaminetetraacetic acid on the depth of dentinal tubule penetration of an epoxy resin-based root canal sealer: A confocal laser scanning microscopy study. J Conserv Dent Endod. 2025;28:309-13.
[34] Oliveira JS, Raucci Neto W, Faria NS, Fernandes FS, Miranda CE, Abi Rached-Junior FJ. Quantitative assessment of root canal roughness with calcium-based hypochlorite irrigants by 3D CLSM. Braz Dent J. 2014;25(5):409-15.
[35] Uğur Aydin Z, Koşumcu S, Meşeci B. Effect of different irrigation activation techniques on sealer penetration: A confocal laser microscopy study. Chin J Dent Res. 2021;24(2):113-18.
[36] Galler KM, Grubmüller V, Schlichting R, Widbiller M, Eidt A, Schuller C, et al. Penetration depth of irrigants into root dentine after sonic, ultrasonic and photoacoustic activation. Int Endod J. 2019;52(8):1210-17.
[37] Saber Sel-D, Hashem AA. Efficacy of different final irrigation activation techniques on smear layer removal. J Endod. 2011;37(9):1272-75.
[38] De Gregorio C, Estevez R, Cisneros R, Heilborn C, Cohenca N. Effect of EDTA, sonic, and ultrasonic activation on the penetration of sodium hypochlorite into simulated lateral canals: an in vitro study. J Endod. 2009;35:891-95.
[39] Jiang LM, Verhaagen B, Versluis M, Langedijk J, Wesselink P, van der Sluis LW. The influence of the ultrasonic intensity on the cleaning efficacy of passive ultrasonic irrigation. J Endod. 2011;37(5):688-92.
[40] Ateş AA, Arıcan B, Çiftçioğlu E, Küçükay ES. Influence of different irrigation regimens on the dentinal tubule penetration of a bioceramic-based root canal sealer: a confocal analysis study. Lasers Med Sci. 2021;36(8):1771-77.
[41] Arends J, Stokroos I, Jongebloed WG, Ruben J. The diameter of dentinal tubules in human coronal dentine after demineralization and air-drying: a combined light microscopy and SEM study. Caries Res. 1995;29(2):118-21.
[42] Gu Y, Perinpanayagam H, Kum DJ, Yoo YJ, Jeong JS, Lim SM, et al. Effect of different agitation techniques on the penetration of irrigant and sealer into dentinal tubules. Photomed Laser Surg. 2017;35(2):71-77.

[43] De Deus G, Gurgel-Filho ED, Maniglia-Ferreira C, Coutinho Filho T. The influence of filling technique on depth of tubule penetration by root canal sealer: a study using light microscopy and digital image processing. Aust Endod J. 2004;30:23-28.
[44] Kokkas AB, Boutsioukis AC, Vassiliadis LP, Stavrianos CK. The influence of the smear layer on dentinal tubule penetration depth by three different root canal sealers: an in vitro study. J Endod. 2004;30(2):100-02.
[45] Hamdy TM, Galal MM, Ismail AG, Saber S. Physicochemical properties of AH plus bioceramic sealer, Bio-C Sealer, and ADseal root canal sealer. Head Face Med. 2024;20(1):2.
[46] El Hachem R, Khalil I, Le Brun G, Pellen F, Le Jeune B, Daou M, et al. Dentinal tubule penetration of AH Plus, BC Sealer and a novel tricalcium silicate sealer: a confocal laser scanning microscopy study. Clin Oral Investig. 2019;23(4):1871-76.
[47] El Hachem R, Le Brun G, Le Jeune B, Pellen F, Khalil I, Abboud M. Influence of the EndoActivator irrigation system on dentinal tubule penetration of a novel tricalcium silicate-based sealer. Dent J (Basel). 2018;6(3):45.
[48] Raducka M, Piszko A, Piszko PJ, Jawor N, Dobrzyński M, Grzebieluch W, et al. Narrative review on methods of activating irrigation liquids for root canal treatment. Appl Sci. 2023;13:7733.
[49] Ansar A, Afeef, Harishs Shetty K. Uses of ultrasonics in endodontics, a review. Int J Adv Res. 2018;6:1448-59.

**PARTICULARS OF CONTRIBUTORS:**
1. Postgraduate Student, Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur, Andhra Pradesh, India.
2. Professor, Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur, Andhra Pradesh, India.
3. Professor, Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur, Andhra Pradesh, India.
4. Professor and Head, Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur, Andhra Pradesh, India.
5. Professor, Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur, Andhra Pradesh, India.
6. Professor, Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur, Andhra Pradesh, India.

**NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:**
Dr. Sai Meghana Bapatu Yeruguti,
Postgraduate Student, Department of Conservative Dentistry and Endodontics,
Sibar Institute of Dental Sciences, Takkellapadu, Guntur-522509,
Andhra Pradesh, India.
E-mail: saimeghanareddy14@gmail.com

**PLAGIARISM CHECKING METHODS:** [Jain H et al.]
- Plagiarism X-checker: Dec 20, 2025
- Manual Googling: May 21, 2026
- iThenticate Software: May 23, 2026 (19%)

**ETYMOLOGY:** Author Origin

**EMENDATIONS:** 8

**AUTHOR DECLARATION:**
- Financial or Other Competing Interests: None
- Was Ethics Committee Approval obtained for this study? Yes
- Was informed consent obtained from the subjects involved in the study? No
- For any images presented appropriate consent has been obtained from the subjects. Yes

Date of Submission: **Dec 18, 2025**
Date of Peer Review: **Feb 02, 2026**
Date of Acceptance: **May 26, 2026**
Date of Publishing: **Sep 01, 2026**