

Effect of 2% Chlorhexidine Gel Medicament on Push-out Bond Strength of Epoxy Resin and Bioceramic Sealers in teeth extracted from Diabetics: An In-vitro Study

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ABSTRACT

Introduction: Diabetes Mellitus (DM) impairs healing, immune function and dentin metabolism, potentially affecting adhesion. Chlorhexidine (CHX) gel is widely used as an intracanal medicament due to its sustained antimicrobial action; however, its influence on sealer bonding in diabetes-altered dentin is unclear. AH Plus® and BioRoot Root Canal Sealer (RCS) sealers are commonly used, yet their adhesion following 2% CHX application requires evaluation.

Aim: To evaluate the effect of 2% CHX gel on the Push-Out Bond Strength (POBS) of AH Plus® and BioRoot RCS in teeth from diabetics.

Materials and Methods: This in-vitro study was conducted at the Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur, Andhra Pradesh, India. The study duration was three months, from June 2024 to August 2024. A total of 52 single-rooted teeth were prepared up to size 35/06 using Sodium Hypochlorite (NaOCl), sonic activation and Ethylenediaminetetraacetic Acid (EDTA). Specimens were

divided into two groups: without 2% CHX (Group-1) and with 2% CHX (Group-2) gel applied for two weeks, followed by obturation using AH Plus® and BioRoot RCS. After two weeks, 2-mm mid-root slices were subjected to POBS testing and bond failure mode analysis.

Results: One-way Analysis of Variance (ANOVA) showed a significant difference in POBS among the groups (F value 9.36, p-value <0.001). AH Plus® with 2% CHX showed the highest mean POBS (2.44 ± 0.59 MPa), followed by AH Plus® without CHX (1.81 ± 0.80 MPa). BioRoot RCS without CHX showed 1.27 ± 0.59 MPa, while BioRoot RCS with 2% CHX showed 1.41 ± 0.44 MPa. CHX increased Bond Strength (BS) for AH Plus®, while only a minimal increase was observed for BioRoot RCS. Post-hoc test illustrated that AH Plus® with 2% CHX (Group-2A: 2.44 ± 0.58) demonstrated a significantly greater POBS compared to BioRoot RCS with 2% CHX (Group-2B: 1.41 ± 0.44) (p-value <0.05).

Conclusion: The AH Plus® showed higher POBS than BioRoot RCS, with 2% CHX significantly enhancing AH Plus® adhesion but not BioRoot RCS.

Keywords: Adhesion, Matrix metalloproteinase inhibitors, Substantivity, Systemic health influence

INTRODUCTION

The DM is a metabolic disorder characterised by impaired insulin secretion or reduced insulin action [1]. It induces structural and functional alterations in the dental pulp by disrupting circadian rhythms, inhibiting dentin bridge formation and compromising vascularity, thereby adversely affecting tissue nourishment, healing and regeneration [2]. DM also interferes with calcification, enamel-dentin development and collagen integrity. Advanced glycation leads to increased dentin brittleness, altered mechanical properties and enlargement of dentinal tubule diameter and density, particularly in areas adjacent to the pulp [1]. Since peritubular dentin is highly mineralised, diabetes-related changes in calcification may significantly influence its structural characteristics [1].

The success of endodontic treatment relies on effective debridement and disinfection of the root canal system [3]. Mechanical instrumentation alone is insufficient, as it leaves residual debris and microorganisms, necessitating chemomechanical preparation [4]. Sodium Hypochlorite (NaOCl), Ethylenediaminetetraacetic Acid (EDTA), and CHX are commonly employed irrigants [3], although their antimicrobial efficacy depends on microbial resistance patterns. CHX exhibits substantivity [5], broad-spectrum antimicrobial activity and low toxicity [6]; however, it lacks tissue-dissolving ability [6]. Intracanal medicaments enhance antimicrobial efficacy, especially in cases of trauma, abscesses, perforations, resorptive defects and immature teeth [7]. Among commonly used medicaments,

CHX is effective against *Enterococcus faecalis* [8] and 2% CHX is recommended for persistent endodontic infections [9]. CHX gel also facilitates instrumentation and reduces smear layer formation [10]. As DM accelerates collagen degradation through increased Matrix Metalloproteinase (MMP) activity [1], CHX, an MMP inhibitor, may enhance dentin-sealer BS [11,12]. Conversely, inadequate removal of intracanal medicaments may hinder sealer penetration and compromise BS [9].

Bioceramic sealers have gained popularity due to their bioactivity and osteoinductive potential [13]. BioRoot RCS, composed of tricalcium silicate, zirconium oxide and mineral fillers [14], demonstrates low cytotoxicity, antibacterial properties and sustained calcium ion release that supports tissue regeneration [15]. Its hydrophilic nature improves sealing through apatite formation along dentinal walls [15]. Epoxy resin-based sealers such as AH Plus® are widely used owing to their low solubility, favourable sealing ability, strong adhesion to dentin and biocompatibility [3]. Effective adhesion between the sealer, gutta-percha and dentin is essential to prevent microleakage and ensure long-term treatment success [7].

The POBS testing is a reliable method for assessing interfacial adhesion, although stress distribution may influence results. POBS testing is commonly used to evaluate the adhesion between root canal sealers and dentinal walls. It measures the resistance of the obturation material to dislodgement under compressive forces, thereby reflecting the integrity and stability of the sealer-dentin

interface. Adequate BS is essential for maintaining a hermetic seal, preventing microleakage and improving the long-term success of endodontic treatment. Although variations in stress distribution may affect the results, the push-out test is considered a reliable and widely accepted method for assessing the bonding performance of endodontic sealers [16]. The use of 2-mm-thick root slices minimises these limitations [17,18]. Cold Lateral Compaction (CLC) was selected due to its safety, cost-effectiveness and ability to promote sealer penetration, despite inherent technique sensitivity and the possibility of void formation [19].

Although CHX has been shown to enhance the BS of resin-based sealers [7], diabetic dentin exhibits altered physicochemical properties that may affect sealer adhesion [1]. Furthermore, evidence regarding the influence of CHX as an intracanal medicament on sealer-dentin bonding in diabetic teeth is limited [1,2].

Therefore, the present study aimed to evaluate the effect of 2% CHX gel on the POBS of root canal sealers in teeth obtained from diabetic patients.

MATERIALS AND METHODS

The present in-vitro investigation was carried out in the Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur, Andhra Pradesh, India, over a period of three months from June 2024 to August 2024. Ethical approval for the study was obtained from the Institutional Ethics Committee (Approval No.: Pr.196/IEC/SIBAR/2023). Informed consent was obtained from patients.

Sample size calculation: Sample size was calculated using G*power 3.1.9.2 software with effect size of 0.8, Type 1 (α error): 0.05, power of the study (β error-80%). A total of 52 teeth were required for the study, which were allocated into two primary groups of 26 specimens each. These groups were further divided into four subgroups consisting of 13 samples per subgroup. Allocation concealment was ensured using sealed opaque envelopes prepared by an independent investigator.

A total of 52 freshly extracted permanent single-rooted teeth with a single canal which were extracted due to periodontal reasons were obtained from patients with a documented history of DM for a duration exceeding five years. Samples were collected from department of Oral and Maxillofacial surgery in SIBAR institute of dental sciences, Guntur.

Inclusion criteria: Teeth that were non carious, single-rooted, exhibited a single canal and had completely formed apices were selected.

Exclusion criteria: Teeth with multiple canals, root caries, developmental anomalies, cracks, fractures, previous endodontic treatment, or existing restorations were excluded from the study.

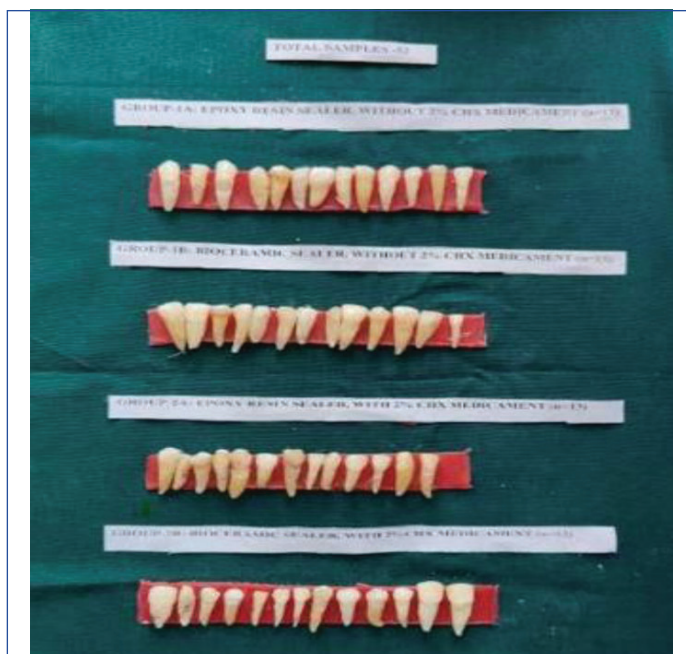
Study Procedure

Root Canal Instrumentation and Preparation: With the required armamentarium as shown in [Table/Fig-1]. Following extraction, all the samples [Table/Fig-2] were debrided using an ultrasonic scaler (Coltene Biosonic S1L Portable Scaler, Switzerland) to eliminate surface contaminants and were subsequently stored in 0.1% thymol solution until use. Radiographic examination using a dental X-ray unit (Acteon, X-Mind, England) confirmed the presence of a single canal [Table/Fig-3]. Access cavity preparation was performed using Endo Access burs (Dentsply Sirona, USA) [Table/Fig-4] [9]. Canal patency was established with size 10 and 15 K-files [Table/Fig-5] [9]. The working length was determined and standardised at 1 mm short of the apical foramen. Rotary instrumentation was completed up to size #35/.06 [Table/Fig-6] [9].

Irrigation was carried out using 2 mL of 3% NaOCl (PRIME Dental Products Pvt., Ltd., India) for one minute, followed by sonic activation using an EndoActivator (Super Endo Sonic Flush Endo



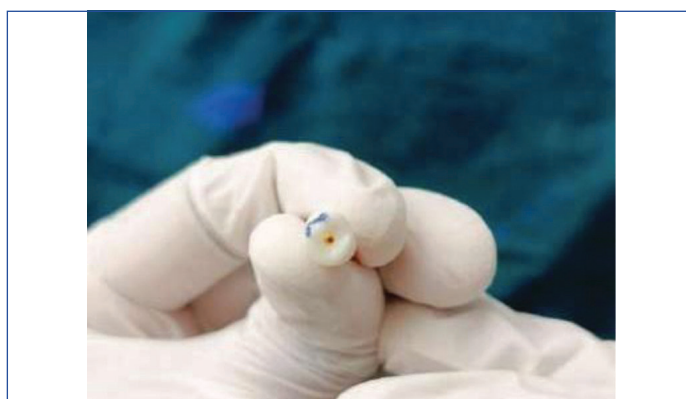
[Table/Fig-1]: Armamentarium.



[Table/Fig-2]: Total collected samples.



[Table/Fig-3]: Preoperative radiograph confirming presence of single canal.



[Table/Fig-4]: Access cavity preparation [9].



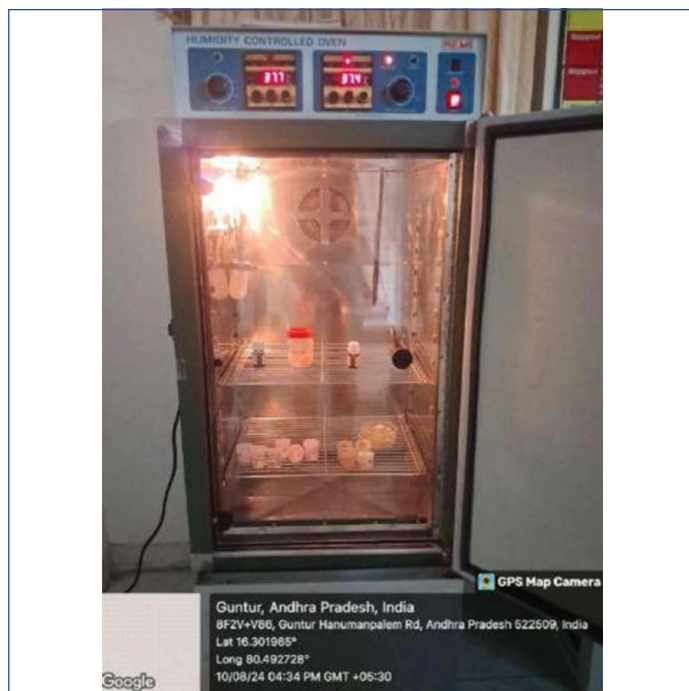
[Table/Fig-5]: Negotiation of canal [9].



[Table/Fig-8]: Application of CHX gel using lentulospiral.



[Table/Fig-6]: Biomechanical preparation [9].



[Table/Fig-9]: Samples placed in humidifier [9].

Irrigator) equipped with a 25/.04 tip for 30 seconds [19]. After each irrigation cycle, the canals were flushed with 1 mL of distilled water for one minute. Upon completion of instrumentation, the canals were irrigated with 2 mL of 17% EDTA (Waldent Alchem, India) for one minute, followed by a final rinse using 5 mL of distilled water for one minute [20].

Grouping and medicament application: Extracted human teeth specimens were randomly allocated into two groups (n=26 each) using a computer-generated randomisation sequence created in IBM SPSS Statistics (Version 21, IBM Corp., Armonk, NY, USA). Allocation concealment was maintained by an independent researcher who prepared the randomisation list and assigned specimens using sequentially numbered, opaque, sealed envelopes to ensure unbiased group distribution.

- **Group-1 (n=26):** No application of 2% CHX gel;
- **Group-2 (n=26):** Application of 2% CHX gel.

In Group-2, 2% CHX gel (Waldent Alchem, India) [Table/Fig-7] was delivered into the root canals using a size #25 lentulospiral [Table/Fig-8] (Waldent Alchem, India). The access cavities were temporarily sealed with Cavitemp {Amrit Chemicals & Minerals Agency (AMMDENT), India}, and all samples were incubated in a humidity-controlled oven {Rajendra Electric Motor Industries (REMI), India} at 37°C with 95% relative humidity for a period of two weeks [Table/Fig-9] [9].



[Table/Fig-7]: Intracanal placement of 2% CHX Gel.



[Table/Fig-10]: Removal of CHX medicament using sonic endoactivation [9].

After the incubation period, the intracanal medicament in Group-2 was removed using hand K-files in conjunction with distilled water irrigation and sonic activation [Table/Fig-10] [9]. This was followed by irrigation with 2 mL of 3% NaOCl for one minute, sonic activation for 30 seconds using a 25/.04 EndoActivator tip, rinsing with 1 mL of distilled water for one minute, irrigation with 2 mL of 17% EDTA for one minute and a final rinse with 5 mL of distilled water for one minute [20].

The specimens were further categorised into four subgroups:

- **Group-1A:** AH Plus® sealer without CHX medicament;
- **Group-1B:** BioRoot RCS sealer without CHX medicament;
- **Group-2A:** AH Plus® sealer following CHX medicament application;

- **Group-2B:** BioRoot RCS sealer following CHX medicament application.

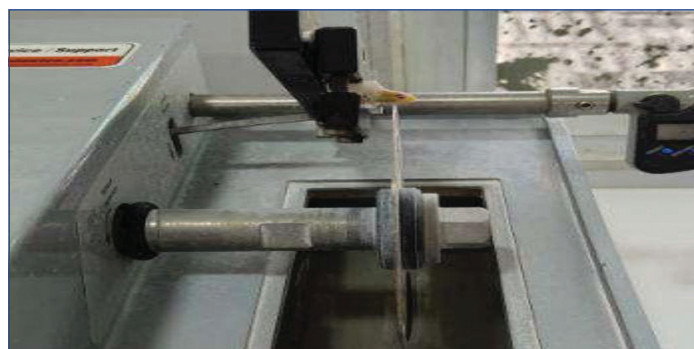
Obturation procedure: The root canals were dried using absorbent paper points prior to obturation. AH Plus® (Dentsply, Konstanz, Germany) and BioRoot RCS (Septodont, France) sealers were prepared in accordance with the manufacturers' guidelines. Obturation was performed using the CLC technique. A size 35/02 master gutta-percha cone coated with sealer was placed to the predetermined working length. Lateral compaction was achieved using a size #25 finger spreader (Mani, Tochigi, Japan) along with accessory gutta-percha cones. Excess gutta-percha was removed at the canal orifice level [21].

Subsequently, the access cavities were restored using a composite resin (Beautifil II, Shofu Inc.) and all specimens were stored under humid conditions for two weeks to ensure complete setting of the sealers.

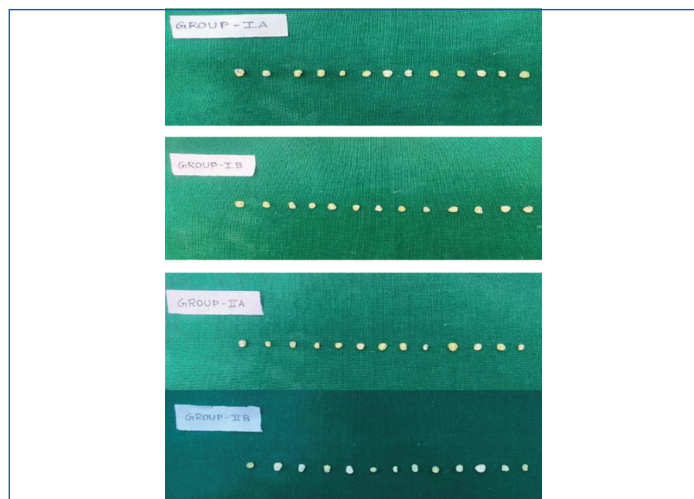
Push-Out Bond Strength (POBS) evaluation: Total tooth samples were mounted on acrylic Moulds [Table/Fig-11]. Using microtome (Baincut LSS) [Table/Fig-12], each specimen was sectioned to obtain 2-mm-thick slices from the middle third of the root using a microtome (Baincut LSS) [Table/Fig-13] [9].



[Table/Fig-11]: Total tooth samples mounted on acrylic moulds.



[Table/Fig-12]: Tooth sectioning under hard tissue microtome (Baincut LSS).



[Table/Fig-13]: Total tooth samples after sectioning with hard tissue microtome [9].

The POBS testing was conducted using a universal testing machine (Instron E3000) [Table/Fig-14]. A cylindrical plunger with a diameter of 0.5 mm was employed. The slices were positioned with the apical surface facing the plunger to facilitate loading in an apico-coronal direction. A crosshead speed of 1 mm/min was applied until debonding occurred [9].



[Table/Fig-14]: Push Out Bond Strength (POBS) test using universal testing machine (Instron).

Failure mode assessment: Following BS testing, the mode of failure was analysed under a stereomicroscope (Magnus, Olympus India Pvt., Ltd.). The observed failure patterns were documented and subjected to statistical analysis [9].

STATISTICAL ANALYSIS

Data analysis was performed using IBM SPSS Statistics for Windows, Version 21.0 (IBM Corp., Armonk, NY, USA). One-way Analysis of Variance (ANOVA) was applied to compare POBS among groups, followed by Tukey's post-hoc test for pair-wise comparisons. Chi-square test was used to evaluate differences in failure modes. A p-value<0.05 was considered statistically significant.

RESULTS

As presented in [Table/Fig-15] (One-way analysis of variance), the overall trend in BS values was observed as follows: AH Plus® with 2% CHX > AH Plus® > BioRoot RCS with 2% CHX > BioRoot RCS. One-way ANOVA revealed statistically significant difference in POBS among the four groups ($F=9.36$, p value< 0.001). The mean POBS of AH Plus® without CHX (Group-1A: 1.81 ± 0.802) was higher than that of BioRoot RCS without CHX (Group-1B: 1.27 ± 0.58); while AH Plus® with CHX application (Group-2A: 2.44 ± 0.58) demonstrated a greater POBS compared to BioRoot RCS with CHX (Group-2B: 1.41 ± 0.44).

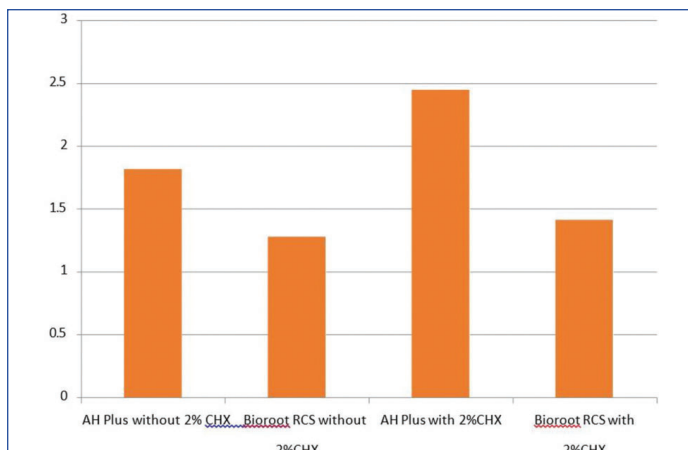
Group	N	Mean	Std. deviation	Std. error mean	F value	p-value
AH Plus® without 2% CHX (Group-1A)	13	1.8123	0.80214	0.22247	9.36	<0.001*
BioRoot RCS without 2% CHX (Group-1B)	13	1.2754	0.58586	0.16249		
AH Plus® with 2% CHX (Group-2A)	13	2.4431	0.58678	0.16274		
BioRoot RCS with 2% CHX (Group-2B)	13	1.4108	0.44101	0.12231		

[Table/Fig-15]: Comparison of mean Push-Out Bond Strength (POBS) between the four study subgroups using One way analysis of variance.

One-way analysis of variance; p-value ≤ 0.05 considered statistically significant

As presented in [Table/Fig-16] the overall trend in BS values was observed as follows: AH Plus® with 2% CHX > AH Plus® > BioRoot RCS with 2% CHX > BioRoot RCS.

The Tukey's post-hoc test to identify intergroup differences showing the mean POBS of AH Plus® without CHX (Group-1A: 1.81 ± 0.802) was higher than that of BioRoot RCS without CHX (Group-1B: 1.27 ± 0.58); however, the difference was not statistically significant (p -value ≥ 0.05). In contrast, AH Plus® used following CHX application



[Table/Fig-16]: Bar chart showing the POBS values among the four study sub-groups.

(Group-2A: 2.44 ± 0.58) demonstrated a significantly greater POBS compared to BioRoot RCS with CHX (Group-2B: 1.41 ± 0.44) (p -value ≤ 0.05) as presented in [Table/Fig-17].

Comparison	Mean Difference	p-value	Significance
AH Plus® without 2% CHX vs AH Plus® with 2% CHX (Group-1A vs Group-2A)	0.63	0.057	Borderline
BioRoot RCS without 2% CHX vs BioRoot RCS with 2% CHX (Group-1B vs Group-2B)	0.14	0.944	NS
AH Plus® without 2% CHX vs BioRoot RCS without 2% CHX (Group-1A vs Group-1B)	0.54	0.133	NS
AH Plus® with 2% CHX vs BioRoot RCS with 2% CHX (Group-2A vs Group-2B)	1.03	0.0005	Significant
AH Plus® with 2% CHX vs BioRoot RCS without 2% CHX (Group-2A vs Group-1B)	1.17	0.0001	Significant
AH Plus® without 2% CHX vs BioRoot RCS with 2% CHX (Group-1A vs Group-2B)	0.40	0.357	NS

[Table/Fig-17]: Multiple comparisons of Push-Out Bond Strength (POBS) among experimental groups using Tukey's post-hoc test.

The pair-wise comparisons to study the effect of CHX on push out BS of sealers which show that CHX tends to increase BS of Epoxy Resin sealer, but the improvement was not statistically significant (p -value > 0.05) [Table/Fig-18a]. CHX showed no statistically significant effect on bioceramic sealers (p -value > 0.05) has been presented in [Table/Fig-18b]. However, while comparing effect of CHX on both the sealers, with CHX Push out BS of AH Plus® significantly increased compared to BioRoot RCS (p -value ≤ 0.05) [Table/Fig-18c].

Groups	Mean	Comparison	p-value
Without 2% CHX (Group-1A)	1.81	vs With 2% CHX	0.057 (NS, borderline)
With 2% CHX (Group-2A)	2.44	—	—

[Table/Fig-18a]: Pair-wise comparisons showing effect of CHX on AH Plus® sealer.

Groups	Mean	Comparison	p-value
Without 2% CHX (Group-1B)	1.28	vs With CHX	0.944 (NS)
With 2% CHX (Group-2B)	1.41	—	—

[Table/Fig-18b]: Pair-wise comparisons showing effect of CHX on BioRoot RCS sealer.

Condition	Epoxy mean	Bioceramic mean	p-value
Without 2% CHX	1.81 (Group-1A)	1.28 (Group-1B)	0.133 (NS)
With 2% CHX	2.44 (Group-2A)	1.41 (Group-2B)	0.05 (significant)

[Table/Fig-18c]: Pair-wise comparisons comparing effect of CHX on Epoxy and Bioceramic sealer.

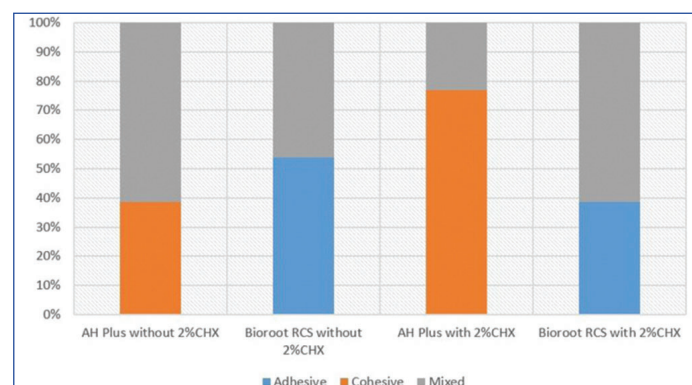
The variation in failure patterns among the experimental groups showing statistically significant difference between the experimental groups (p -value < 0.001 as represented in [Table/Fig-19]). Group-1A predominantly exhibited mixed failures, whereas adhesive failures were more frequently observed in Group-1B. Group-2A showed mainly cohesive failures, while mixed failure modes were most commonly noted in Group-2B.

The distribution of bond failure modes among the experimental groups is depicted in [Table/Fig-20]. Group-1A predominantly exhibited mixed failures, whereas adhesive failures were more frequently observed in Group-1B. Group-2A demonstrated mainly cohesive failures, while Group-2B showed a higher incidence of mixed failure patterns.

Groups	Bond Strength (BS)			Chi-square value	p-value
	Adhesive	Cohesive	Mixed		
AH Plus® without 2% CHX (Group-1A)	0	5 (38.5%)	8 (61.5%)	33.66	$< 0.001^*$
BioRoot RCS without 2% CHX (Group-1B)	7 (53.8%)	0	6 (46.2%)		
AH Plus® with 2% CHX (Group-2A)	0	10 (76.9%)	3 (23.1%)		
BioRoot RCS with 2% CHX (Group-2B)	5 (38.5%)	0	8 (61.5%)		

[Table/Fig-19]: Chi-square test illustrating the variation in failure patterns among the experimental groups.

Chi-square test; p -value ≤ 0.05 considered statistically significant



[Table/Fig-20]: Comparison of mode of failure between the four study subgroups using Chi-square test.

DISCUSSION

The POBS values observed in the present study followed the order: AH Plus® with 2% CHX $>$ AH Plus® without 2% CHX $>$ BioRoot RCS with 2% CHX $>$ BioRoot RCS without 2% CHX. In the absence of CHX, AH Plus® exhibited superior BS compared to BioRoot RCS.

One-way ANOVA revealed a statistically significant overall difference (F value 9.36, p -value < 0.001), pair-wise comparisons indicated that AH Plus® with CHX demonstrated superior BS compared to BioRoot RCS groups. This suggests a material-specific enhancement rather than a generalised effect.

In the present study, the mean POBS of AH Plus® without CHX {Group-1A (1.81 ± 0.802)} was reported to be higher than BioRoot RCS without CHX {Group-1B (1.27 ± 0.58)} without any significant difference (p value > 0.05). AH Plus® sealer with CHX {Group-2A (2.44 ± 0.58)} demonstrated significantly higher mean POBS values compared to the BioRoot RCS with CHX {Group-2B (1.41 ± 0.44)} with p -value ≤ 0.05 (Tukey post-hoc). There was a positive effect of CHX on POBS of AH Plus® {(Group-1A (1.81 ± 0.802) and Group-2A (2.44 ± 0.58)} but without significant difference with p -value > 0.05 . There was no significant effect of CHX on POBS of BioRoot RCS {Group-1B: 1.27 ± 0.58 and Group-2B: 1.41 ± 0.44 } with (p -value > 0.05).

The CLC was selected to evaluate POBS due to its safety, cost-effectiveness and ability to promote sealer penetration, despite inherent technique sensitivity and the possibility of void formation [19].

This finding was in agreement with Donnermeyer D et al., who attributed the enhanced adhesion of AH Plus® to its ability to form covalent bonds with dentinal collagen, whereas calcium silicate-based sealers rely primarily on mineral infiltration-mediated micromechanical retention [22].

Neelakantan P et al., demonstrated improved BS of AH Plus® following EDTA and NaOCl irrigation, which was attributed to effective smear layer removal facilitating deeper sealer penetration [23]. In contrast, NaOCl used alone may negatively affect resin bonding by degrading the collagen or organic matrix of dentin [23-25]. Kurup D et al., also reported enhanced bonding performance of AH Plus®, attributing it to its dimensional stability and minimal polymerisation shrinkage [26]. Conversely, Saleh IM et al., suggested that retention of the smear layer may improve calcium silicate-based sealers marginal adaptation, which could explain the reduced BS of BioRoot RCS following smear layer removal [27].

Contradictory findings have been reported in the literature. Nouroloyouni A et al. and Navjot SM et al., observed higher BS values for calcium silicate-based sealers, attributing this to compromised resin-dentin bonding after NaOCl-induced collagen degradation and to hydroxyapatite formation during the setting reaction of calcium silicate sealers, which enhances their adhesion [19,28].

In the present study, mean POBS of AH Plus® sealer has been significantly improved with CHX intracanal medicament (Group-2A). For BioRoot RCS the mean BS has increased but there was no significant effect of CHX on BS. Similar results were obtained by Maan S et al., where AH Plus® showed considerably more POBS than MTA Fillapex following administration of CHX intracanal medication [7]. They claimed that CHX might improve resin-based sealer BS which may account for increased POBS of AH Plus®. Additionally, they noted that AH Plus® superior BS might be influenced by its capacity to form covalent bond with collagen amino groups, its reduced shrinkage, intrinsic volumetric expansion and long-term dimensional stability [7].

Comparable to the results of present research, AH Plus® had better BS than MTA Fillapex with CHX as a final irrigant mentioned by Gupta PR et al., CHX strengthens resin-based sealer bond to dentine as it acts as an MMP inhibitor and prevents collagen degradation, thus allows better penetration of resin sealers into collagen network [11]. The irrigation protocol used to remove smear layer i.e., 17% EDTA results in demineralisation and exposes collagen matrix with which AH Plus® sealer bonds. This might be the reason for improved BS of Resin sealer [11]. The stable BS achieved by AH Plus® after application of 2% CHX medicament in this study might also be due to above mentioned properties of CHX.

Although BioRoot RCS demonstrated an increase in BS after CHX application, the difference was not statistically significant. Similar findings were reported by Roy D et al., who suggested that while CHX may improve dentin wettability, residual medicament may restrict sealer penetration into dentinal tubules [9].

The comparatively lower BS of BioRoot RCS in the present study may also be attributed to alterations in diabetic dentin. Although CHX has been shown to enhance the BS of resin-based sealers [7], diabetic dentin exhibits altered physicochemical properties that may affect sealer adhesion [1]. Abbassy MA et al., reported reduced mineralisation and impaired collagen maturation in teeth affected by DM [29]. Additionally, Saghir MA et al., observed reduced POBS values for mineral trioxide aggregate in diabetic dentin due to increased dentinal tubule density and reduced peritubular mineralisation, factors that adversely affect sealers dependent on mineral-based bonding mechanisms [1]. Hyperglycaemia-associated increases in MMP and Advanced Glycation End-product

(AGE) activity further compromise the organic dentinal matrix [30]. CHX may partially counteract these effects by inhibiting dentinal MMPs and cathepsins, thereby favouring improved adhesion of epoxy resin-based sealers such as AH Plus® [11].

Failure mode analysis revealed variations based on sealer type and CHX application. In the absence of CHX, AH Plus® predominantly exhibited mixed failure patterns, whereas BioRoot RCS showed mainly adhesive failures, indicating weaker sealer-dentin interaction. Following CHX application, AH Plus® demonstrated predominantly cohesive failures, which is consistent with findings reported by Gupta H et al., Vilanova WV et al. and Albino Souza M et al., reflecting improved bonding and mechanical integrity [3-5]. BioRoot RCS with CHX showed mainly mixed failures, while adhesive failures were more frequent without CHX. This may be attributed to reduced calcium availability and mineralisation in diabetic dentin, which adversely affects the inorganic-phase-dependent bonding mechanism of calcium silicate-based sealers. Although Roy D et al., reported cohesive failures for both sealers following intracanal medicament use, the altered characteristics of diabetic dentin may explain the differences observed in the present study [9].

Overall, AH Plus® demonstrated superior POBS owing to its chemical bonding potential and favourable physical properties, with CHX application further enhancing its adhesion. Although BioRoot RCS showed improvement following CHX application, its bonding effectiveness appeared limited by diabetes-induced dentinal alterations. Further investigations are warranted to substantiate these findings.

Limitation(s)

The present in-vitro study utilised teeth obtained from diabetic patients, which may limit direct extrapolation of the results to clinical scenarios. The use of single-rooted teeth with straight canals restricts the applicability of the findings to more complex root canal anatomies. POBS was evaluated after a two-week period of CHX application; longer evaluation periods may be necessary to assess long-term bond durability. Minor variations in sealer manipulation could have influenced the outcomes. Additionally, BS assessment was limited to the middle third of the root, which may not represent variations along the entire canal length. The findings may not be applicable to healthy dentin, underscoring the need for comparative studies.

CONCLUSION(S)

Within the limitations of the present study, AH Plus® exhibited significantly higher POBS than BioRoot RCS in teeth obtained from diabetic patients. The application of 2% CHX enhanced the adhesion of AH Plus®, while BioRoot RCS also demonstrated an increase in BS following CHX application, although this improvement was not statistically significant. Considering the altered bonding behaviour of sealers in diabetic dentin, the use of 2% CHX as an intracanal medicament may contribute to improved obturation quality, especially when epoxy resin-based sealers are employed. Further in-vivo and comparative studies are required to validate these findings under clinical conditions.

REFERENCES

- [1] Saghir MA, Karamifar K, Fakharzadeh A, Conte M, Morgano SM. Effect of diabetes on tubular density and push-out bond strength of mineral trioxide aggregate to dentin. *J Endod.* 2020;46(11):1584-91. Doi: 10.1016/j.joen.2020.07.025. Epub 2020 Jul 29. PMID: 32738338.
- [2] Saghir MA, Nath D, Rahmani B, Amini S, Karamifar K, Peters OA. The effect of diabetes on fracture resistance of teeth: An in vitro study. *Aust Endod J.* 2021;47(3):499-505. Doi: 10.1111/aej.12512. Epub 2021 Apr 4. PMID: 33813800.
- [3] Gupta H, Kandaswamy D, Manchanda SK, Shourie S. Evaluation of the sealing ability of two sealers after using chlorhexidine as a final irrigant: An in vitro study. *J Conserv Dent.* 2013;16(1):75-78. Doi: 10.4103/0972-0707.105304. PMID: 23349582; PMCID: PMC3548352.

- [4] Vilanova WW, Carvalho-Junior JR, Alfredo E, Sousa-Neto MD, Silva-Sousa YT. Effect of intracanal irrigants on the bond strength of epoxy resin-based and methacrylate resin-based sealers to root canal walls. *Int Endod J*. 2012;45(1):42-48. Doi: 10.1111/j.1365-2591.2011.01945.x. Epub 2011 Sep 8. PMID: 21899566.
- [5] Albino Souza M, Dalla Lana D, Gabrielli E, Barbosa Ribeiro M, Miyagaki DC, Cecchin D. Effectiveness of final decontamination protocols against *Enterococcus faecalis* and its influence on bond strength of filling material to root canal dentin. *Photodiagnosis Photodyn Ther*. 2017;17:92-97. Doi: 10.1016/j.pdpdt.2016.11.004. Epub 2016 Nov 16. PMID: 27866000.
- [6] Dinesh K, Murthy BV, Narayana IH, Hegde S, Madhu KS, Nagaraja S. The effect of 2% chlorhexidine on the bond strength of two different obturating materials. *J Contemp Dent Pract*. 2014;15(1):82-85. Doi: 10.5005/jp-journals-10024-1492. PMID: 24939270.
- [7] Maan S, Bhatt VD, Singh R, Gupta S, Noorain SA, Gill A, et al. The effect of four different intracanal medicaments on the push-out bond strength of root canal sealers. *J Med Life*. 2022;15(4):448-53. Doi: 10.25122/jml-2020-0104. PMID: 35646182; PMCID: PMC9126449.
- [8] Hashem AA, Ghoneim AG, Lutfy RA, Fouda MY. The effect of different irrigating solutions on bond strength of two root canal-filling systems. *J Endod*. 2009;35(4):537-40. Doi: 10.1016/j.joen.2009.01.003. Epub 2009 Feb 26. PMID: 19345800.
- [9] Roy D, Katakai R, Das L, Jain K. Influence of 2% chlorhexidine on the dislodgement resistance of AH Plus, BioRoot RCS, and GuttaFlow 2 sealer to dentin and sealer-dentin interface. *J Conserv Dent*. 2022;25(6):642-47. Doi: 10.4103/jcd.jcd_355_22. Epub 2022 Aug 17. PMID: 36591575; PMCID: PMC9795685.
- [10] Ferraz CC, Gomes BP, Zaia AA, Teixeira FB, Souza-Filho FJ. In vitro assessment of the antimicrobial action and the mechanical ability of chlorhexidine gel as an endodontic irrigant. *J Endod*. 2001;27(7):452-55. Doi: 10.1097/00004770-200107000-00004. PMID: 11503994.
- [11] Gupta PR, Aggarwal SD, Kshirsagar SP, Bhargava K, Rai V, Chawla M. Comparative evaluation of push-out bond strength of resin-based and mineral trioxide aggregate-based sealers after using normal saline and 2% chlorhexidine as a final irrigant: An in vitro study. *Endodontology*. 2016;28(1):32-37. Doi: 10.4103/0970-7212.184337.
- [12] Kontakiotis EG, Tsatsoulis IN, Papanakou SI, Tzanetakis GN. Effect of 2% chlorhexidine gel mixed with calcium hydroxide as an intracanal medication on sealing ability of permanent root canal filling: A 6-month follow-up. *J Endod*. 2008;34(7):866-70. Doi: 10.1016/j.joen.2008.04.004. PMID: 18570998.
- [13] Shokouhinejad N, Gorjestani H, Nasseh AA, Hoseini A, Mohammadi M, Shamshiri AR. Push-out bond strength of gutta-percha with a new bioceramic sealer in the presence or absence of smear layer. *Aust Endod J*. 2013;39(3):102-06. Doi: 10.1111/j.1747-4477.2011.00310.x. Epub 2011 May 29. PMID: 24279654.
- [14] Donnermeyer D, Vahdat-Pajouh N, Schäfer E, Dammaschke T. Influence of the final irrigation solution on the push-out bond strength of calcium silicate-based, epoxy resin-based and silicone-based endodontic sealers. *Odontology*. 2019;107(2):231-36. Doi: 10.1007/s10266-018-0392-z. Epub 2018 Oct 1. PMID: 30276580.
- [15] Atmeh AR, Chong EZ, Richard G, Festy F, Watson TF. Dentin-cement interfacial interaction: Calcium silicates and polyalkenoates. *J Dent Res*. 2012;91(5):454-59. Doi: 10.1177/0022034512443068. Epub 2012 Mar 20. PMID: 22436906; PMCID: PMC4077527.
- [16] Nagas E, Uyanik MO, Eymirli A, Cehreli ZC, Vallittu PK, Lassila LV, et al. Dentin moisture conditions affect the adhesion of root canal sealers. *J Endod*. 2012;38(2):240-44. Doi: 10.1016/j.joen.2011.09.027. Epub 2011 Nov 13. PMID: 22244645.
- [17] Nagas E, Cehreli Z, Uyanik MO, Durmaz V. Bond strength of a calcium silicate-based sealer tested in bulk or with different main core materials. *Braz Oral Res*. 2014;28:S1806-83242014000100256. Doi: 10.1590/1807-3107bor-2014.vol28.0046. Epub 2014 Sep 15. PMID: 25229786.
- [18] Saghir MA, Obeidi A, Nath D, Morgano SM. The effect of diabetes mellitus on the shear bond strength of composite resin to dentin and enamel. *Odontology*. 2022;110(1):92-98. Doi: 10.1007/s10266-021-00641-0. Epub 2021 Jul 27. PMID: 34318336.
- [19] Nourolooyouni A, Samadi V, Salem Milani A, Noorolouny S, Valizadeh-Haghi H. Single cone obturation versus cold lateral compaction techniques with bioceramic and resin sealers: Quality of obturation and push-out bond strength. *Int J Dent*. 2023;2023:3427151. Doi: 10.1155/2023/3427151. PMID: 36704662; PMCID: PMC9873427.
- [20] Wiesse PEB, Silva-Sousa YT, Pereira RD, Estrela C, Domingues LM, Pécora JD, et al. Effect of ultrasonic and sonic activation of root canal sealers on the push-out bond strength and interfacial adaptation to root canal dentine. *Int Endod J*. 2018;51(1):102-11. Doi: 10.1111/iej.12794. Epub 2017 Jun 19. PMID: 28543092.
- [21] Mounes BB, Alhashimi R. The push out bond strength of bioceramic sealer (Total Fill) after warm and cold obturation techniques: An in vitro comparative study. *Journal of Baghdad College of Dentistry*. 2022;34(3):7-16. Doi: 10.26477/jbcd.v34i3.3212.
- [22] Donnermeyer D, Dornseifer P, Schäfer E, Dammaschke T. The push-out bond strength of calcium silicate-based endodontic sealers. *Head Face Med*. 2018;14(1):13. Doi: 10.1186/s13005-018-0170-8. PMID: 30126425; PMCID: PMC6102912.
- [23] Neelakantan P, Subbarao C, Subbarao CV, De-Deus G, Zehnder M. The impact of root dentine conditioning on sealing ability and push-out bond strength of an epoxy resin root canal sealer. *Int Endod J*. 2011;44(6):491-98. Doi: 10.1111/j.1365-2591.2010.01848.x. Epub 2011 Jan 24. PMID: 21255047.
- [24] Kumar PS, Meganathan A, Shiram S, Sampath V, Sekar M. Effect of proanthocyanidin and bamboo salt on the push-out bond strength of an epoxy resin sealer to sodium hypochlorite-treated root dentin: An in vitro study. *J Conserv Dent*. 2019;22(2):144-48. Doi: 10.4103/JCD.JCD_377_18. PMID: 31142983; PMCID: PMC6519184.
- [25] Manimaran VS, Srinivasulu S, Rajesh Ebenezer A, Mahalaxmi S, Srinivasan N. Application of a proanthocyanidin agent to improve the bond strength of root dentin treated with sodium hypochlorite. *J Conserv Dent*. 2011;14(3):306-08. Doi: 10.4103/0972-0707.85822. PMID: 22025839; PMCID: PMC3198565.
- [26] Kurup D, Nagpal AK, Shetty S, Mandal TK, Anand J, Mitra R. Data on the push-out bond strength of three different root canal treatment sealers. *Bioinformation*. 2021;17(1):67-72. Doi: 10.6026/97320630017067. PMID: 34393420; PMCID: PMC8340685.
- [27] Saleh IM, Ruyter IE, Haapasalo MP, Orstavik D. Adhesion of endodontic sealers: Scanning electron microscopy and energy dispersive spectroscopy. *J Endod*. 2003;29(9):595-601. Doi: 10.1097/00004770-200309000-00013. PMID: 14503835.
- [28] Navjot SM, Ashu J, Kamalpreet K, Navneet KM, Manu R, Divya B. The effect of natural reducing agents on push-out bond strength of AH Plus and BioRoot RCS to sodium hypochlorite treated root dentin. *J Conserv Dent*. 2021;24(2):130-34. Doi: 10.4103/jcd.jcd_52_21. Epub 2021 Oct 9. PMID: 34759577; PMCID: PMC8562831.
- [29] Abbassy MA, Watari I, Bakry AS, Hamba H, Hassan AH, Tagami J, et al. Diabetes detrimental effects on enamel and dentine formation. *J Dent*. 2015;43(5):589-96. Doi: 10.1016/j.jdent.2015.01.005. Epub 2015 Feb 11. PMID: 25681642.
- [30] Ayuk SM, Abrahamse H, Houreld NN. The role of matrix metalloproteinases in diabetic wound healing in relation to photobiomodulation. *J Diabetes Res*. 2016;2016:2897656. Doi: 10.1155/2016/2897656. Epub 2016 May 23. PMID: 27314046; PMCID: PMC4893587.

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