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# Effect of allicin-incorporated graphene oxide hydrogel on dentin microhardness

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## Abstract

**Objective** The success of root canal treatment and regenerative endodontics relies on thorough disinfection and dentin integrity preservation to ensure long-term tooth survival. This study evaluates the pH stability, material characteristics, microhardness and antimicrobial effects of an allicin-incorporated GO-AgNP hydrogel compared to conventional intracanal medicaments.

**Methods** An allicin-incorporated GO-AgNP hydrogel was synthesized using allicin extract, GO-AgNPs, and sodium alginate. Characterization was performed via FTIR, SEM, and EDX. pH stability of AllGOAgNP, CaOH, CHX, and TAP was assessed at 5 min, 24 h, and 7 days using a digital pH meter. A total of 120 extracted human premolars were randomly assigned to four groups: (1) Control, (2) CaOH, (3) TAP, and (4) AllGOAgNP. Medicaments were applied and incubated at 37 °C with 100% humidity for 1 week, 1 month, and 3 months. Dentin microhardness was evaluated using a Vickers microhardness tester before and after treatment across the coronal, middle, and apical thirds. Additionally, antimicrobial efficacy against *E. faecalis* and *C. albicans* was assessed using the disc diffusion method, with inhibition zones measured for each medicament. Statistical analysis was performed using one-way ANOVA and Tukey's post-hoc test ( $p < 0.05$ ).

**Results** FTIR analysis confirmed the successful incorporation of allicin, GO, AgNPs, and sodium alginate. SEM images showed a uniform nanoparticle distribution in the hydrogel, and EDX confirmed the presence of key elements, including silver and sulfur. The Allicin-GO-AgNP hydrogel maintained a near-neutral pH (mean 7.083), while CaOH (mean 12.297) and TAP (mean 12.683) exhibited highly alkaline pH levels. ANOVA results demonstrated significant differences in microhardness across groups and regions ( $p < 0.05$ ). The Allicin-GO-AgNP hydrogel exhibited significantly higher microhardness than CaOH and TAP across all regions ( $p < 0.001$ ), with no significant difference from the control in the coronal and middle thirds ( $p > 0.05$ ). SEM-EDX analysis of treated dentin confirmed minimal structural alterations in the Allicin-GO-AgNP hydrogel group compared to the control. In antimicrobial testing, the hydrogel demonstrated moderate efficacy with inhibition zones of 20 mm against *E. faecalis* and 13 mm against *C. albicans*, outperforming calcium hydroxide after 24 h.

**Conclusion** The Allicin-GO-AgNP hydrogel demonstrated superior dentin preservation compared to conventional intracanal medicaments. Its near-neutral pH, structural stability, and microhardness retention make it a promising alternative for endodontic applications, particularly in regenerative endodontics. Future studies should focus on its long-term biocompatibility and antimicrobial effectiveness in clinical settings.

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**Clinical significance** The Allicin-GO-AgNP hydrogel preserves dentin integrity better than conventional medicaments, reducing the risk of structural weakening. Its near-neutral pH minimizes collagen degradation, making it a promising option for regenerative endodontics. This novel hydrogel offers a biocompatible alternative with potential long-term clinical benefits.

**Keywords** Allicin, Dentin microhardness, Graphene oxide, Intracanal medicament, Regenerative endodontics, Silver nanoparticles

## Introduction

The success of endodontic treatment depends on thorough disinfection and the preservation of dentin integrity to ensure long-term tooth survival. Intracanal medicaments are crucial in enhancing root canal disinfection by complementing mechanical debridement [1, 2]. Among conventional medicaments, calcium hydroxide (CaOH) and triple antibiotic paste (TAP) are widely used due to their antimicrobial efficacy [3–5]. However, their prolonged application has been associated with adverse effects on dentin, including demineralization, reduced fracture resistance, and decreased microhardness, particularly in immature teeth with fragile roots [6].

The preservation of dentin structure is critical in procedures such as endodontic regeneration (ER), where maintaining root strength is essential for long-term clinical success. Despite their efficacy in microbial control, conventional medicaments have been reported to compromise dentin properties such as flexural strength, microhardness, and mineral content, making teeth more susceptible to fractures [6–8]. These concerns highlight the need for novel intracanal medicaments that support disinfection and preserve dentin integrity.

Advancements in nanotechnology have led to the exploration of materials like graphene oxide (GO) and silver nanoparticles (AgNPs) in endodontics [9]. With its high surface area and ability to be functionalized, GO offers potential benefits for drug delivery and material reinforcement, while AgNPs exhibit strong broad-spectrum antimicrobial properties [10]. Additionally, allicin, a bioactive compound derived from garlic, has gained attention for its antioxidant and biological effects, which may further enhance the properties of intracanal medicaments [11].

Incorporating allicin, GO, and AgNPs into a hydrogel-based formulation represents a novel strategy to optimize dentin preservation while maintaining a favourable pH environment for endodontic applications. This study evaluates the pH stability, material characteristics, and microhardness effects of an allicin-incorporated GO-AgNP hydrogel compared to conventional intracanal medicaments such as CaOH and TAP. We hypothesize that the allicin-incorporated GO-AgNP hydrogel will exhibit comparable or superior dentin-preserving properties to conventional intracanal medicaments.

## Methodology

The study was conducted following ethical approval from the Institutional Review Board of Saveetha Dental College and Hospitals, Chennai, India, under approval number SRB/SDC/PhD/ENDO-2364/24/TH-014. Ethical guidelines outlined by the institution were strictly adhered to throughout the study.

The preparation of graphene oxide silver nanoparticles (GO-AgNPs) involved a series of steps adapted from the method described by Annapoorani et al. [9]. For sodium alginate hydrogel preparation, sodium alginate powder (Sigma-Aldrich) was dissolved in deionized water at a concentration of 2% (w/v) and stirred continuously until a clear solution was obtained. For allicin extraction, native garlic cloves were procured, peeled and crushed. The garlic mash, weighing 1000 mg, was mixed with 25 ml of cold water, shaken vigorously, and diluted with an additional 25 ml of cold water. Care was taken to avoid exposure of the garlic extract to heat, which could cause allicin degradation. The mixture was filtered through a 0.45  $\mu\text{m}$  glass filter into an HPLC vial for quality analysis to confirm allicin. GO-AgNPs and allicin extract were added to the sodium alginate solution at a 1:1 ratio (v/v) and stirred thoroughly to ensure a uniform distribution.

The hydrogel was characterized using Fourier Transform Infrared (FTIR) spectroscopy (JASCO Global, Model name FP-8300) analysis to identify functional groups and confirm the incorporation of GO-AgNPs, sodium alginate, and allicin.

The surface morphology of the hydrogels was examined using a scanning electron microscope (SEM, Model: JSM-7100 F, JEOL Ltd.), and energy-dispersive X-ray spectroscopy (EDX) was performed to determine the elemental composition of the hydrogels and confirm the presence of allicin GO-AgNPs.

The pH of four intracanal medicaments—Allicin-incorporated graphene oxide-silver nanoparticle hydrogel (AllGOAgNP), calcium hydroxide (CaOH), chlorhexidine gel (CHX), and triple antibiotic paste (TAP)—was evaluated at 5 min, 24 h, and 7 days post-preparation. Measurements were performed using a calibrated digital pH meter (ELICO Table Top, Model LI120). Before each measurement session, the pH meter was standardized with buffer solutions of known pH (4.0, 7.0, and 10.0) to ensure accuracy. Three independent replicates were tested for each medicament at all time points.

Freshly extracted human single-rooted lower premolar teeth ( $n = 120$ ), extracted for orthodontic reasons, were selected for this in vitro study. The sample size was determined using a power analysis to ensure statistical validity. A pilot study used 10 samples per group to estimate the expected effect size. Based on these preliminary results, a power calculation was performed using G\*Power software (version 3.1.9.7) with an alpha error probability of 0.05 and a power of 0.8. All soft tissues, debris, and calculus were thoroughly removed from the teeth after extraction through hand and ultrasonic scalers. The teeth were stored in distilled water at room temperature for a week before the experiment. The teeth were only considered for inclusion if they showed complete root formation and no previous endodontic treatment or restoration. Teeth showing signs of internal or external root resorption, a root fracture, a visible crack, incomplete root formation, internal calcification, or any canal obliteration identifiable on radiographs were excluded.

The roots were decorated at the cemento-enamel junction (CEJ) using a low-speed diamond saw under constant water coolant. These were then prepared into standardized segments of about 14 mm in length. Working length was determined by inserting a size 10 K-file (Dentsply Sirona Endodontics, Ballaigues, Switzerland) into the root canal until the tip was visible at the apical foramen, after which 1 mm was reduced from this length. The root canal was prepared with the ProTaper NiTi rotary system (Dentsply Sirona Endodontics) following the manufacturer's guidelines up to size F3. For instrumentation, canals were irrigated with 2 ml of 2.5% sodium hypochlorite (NaOCl) after every file use, then irrigated with 2 ml of saline for dislodgment of debris. Prepared root canals were dried with paper points.

The specimens were then randomly assigned to four groups:

- **Group 1:** No treatment was applied. ( $n = 30$ )
- **Group 2:** Calcium hydroxide (RC Cal). ( $n = 30$ )
- **Group 3:** Triple antibiotic paste. ( $n = 30$ )
- **Group 4:** Allicin-incorporated graphene oxide silver nanoparticle hydrogel. ( $n = 30$ )

The intracanal medicaments were introduced into the root canals using a lentulo spiral (Dentsply Maillefer) at low speed to ensure uniform distribution throughout the canal. The medicaments were delivered until visible at the apex to confirm complete canal filling. The specimens were then incubated at 37 °C with 100% humidity for 1 week, 1 month, and 3 months before analysis.

After applying the respective medicaments, the teeth were incubated at 37 °C with 100% humidity for 1 week, 1 month and 3 months. The samples were vertically sectioned under the coolant. Dentin microhardness was

measured at the coronal, middle, and apical thirds of each root using a Vickers microhardness tester (Shimadzu HVM-G, Kyoto, Japan), both before and after treatment.

One extra sample each from Group 1 (No Medicament) and Group 4 (Allicin GO-AgNP Hydrogel) was analyzed using SEM-EDX to assess surface morphology and compositional changes of root dentin surface, comparing the untreated baseline with the experimental group.

The antimicrobial activity of Group 1 (Calcium hydroxide), Group 2 (double antibiotic paste), Group 3 (Plain hydrogel), G4 (No medicament), and G5 (Freshly prepared Allicin GO-AgNP gel) was evaluated using the standard disc-diffusion method. Cultures of *Candida albicans* (ATCC 10231) and *Enterococcus faecalis* (ATCC 29212) were prepared and standardized to a concentration of  $1.5 \times 10^8$  CFU/mL, corresponding to the 0.5 McFarland standard. Sterile swabs were saturated with the bacterial suspensions and evenly spread over Mueller-Hinton Agar (MHA) plates (HiMedia, Mumbai, India) for *E. faecalis* and Sabouraud Dextrose Agar (SDA) plates for *C. albicans*. Discs were impregnated with 40  $\mu$ L of G1-syringe, G2-powder, G3, G4, and G5 extracts, each at a concentration of 20 mg/mL. The plates were then incubated aerobically at 37 °C for 24 h. After incubation, the diameter of the inhibition zones (in mm) was measured using a digital Vernier caliper to assess the antimicrobial efficacy of the tested samples. Each sample was tested in triplicate, and the mean inhibition zone diameter was recorded for statistical analysis.

#### Statistical analysis

Statistical analysis was conducted using SPSS software 23.1. The normality of the data was assessed using the Shapiro-Wilk test. A one-way analysis of variance (ANOVA) was conducted to compare differences in pH stability and dentin microhardness among the four groups. Post-hoc pairwise comparisons were performed using Tukey's test to determine significant intergroup differences. A p-value of  $< 0.05$  was considered statistically significant. All results were reported as mean  $\pm$  standard deviation (SD).

#### Results

The FTIR spectrum of the allicin-incorporated graphene oxide silver nanoparticle sodium alginate hydrogel exhibits key peaks indicating the functional groups and interactions within the material. The broad peak at  $3353 \text{ cm}^{-1}$  corresponds to O–H stretching from alginate and graphene oxide hydroxyl groups. Peaks at  $1641 \text{ cm}^{-1}$  and  $1378 \text{ cm}^{-1}$  are attributed to C=O and C–O stretching of carboxylate groups, while the band at  $1083 \text{ cm}^{-1}$  represents C–O–C vibrations from the alginate backbone. Allicin's presence is confirmed by the  $\text{SO}_2$  stretching at  $1130\text{--}1170 \text{ cm}^{-1}$  and C–H bending at  $1375 \text{ cm}^{-1}$ , with

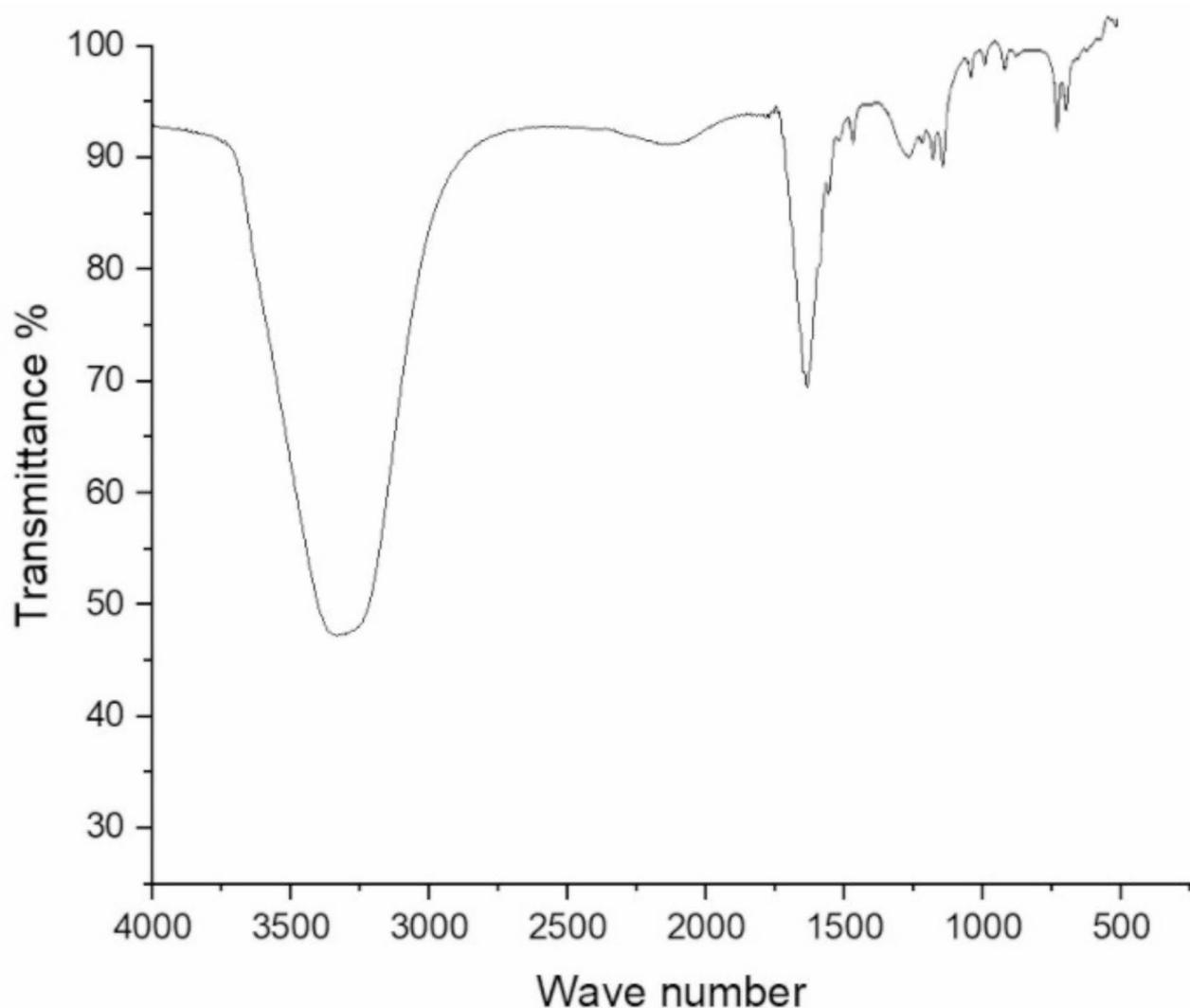
Ag–S interactions appearing at  $\sim 490\text{--}510\text{ cm}^{-1}$ . Additionally, graphene oxide contributes peaks for aromatic C=C stretching near  $1560\text{ cm}^{-1}$ , and Ag–O stretching is evident at  $\sim 520\text{--}600\text{ cm}^{-1}$ . Together, these features confirm the successful incorporation of allicin, graphene oxide, silver nanoparticles, and sodium alginate into the hydrogel. (Fig. 1)

Scanning Electron Microscopy (SEM) image of the GO-AgNP-loaded sodium alginate hydrogel demonstrated a surface with embedded nanoparticles. At the same time, Energy-Dispersive X-ray Spectroscopy (EDX) confirmed the presence of Carbon 45.81%, Oxygen 18.77%, Calcium 15.94%, Sodium 12.24%, Silver 4.33% and sulphur 2.91%, further supporting the successful incorporation of allicin (allyl thiosulfinate) as Sulphur, GO and AgNPs into the hydrogel matrix. (Fig. 2)

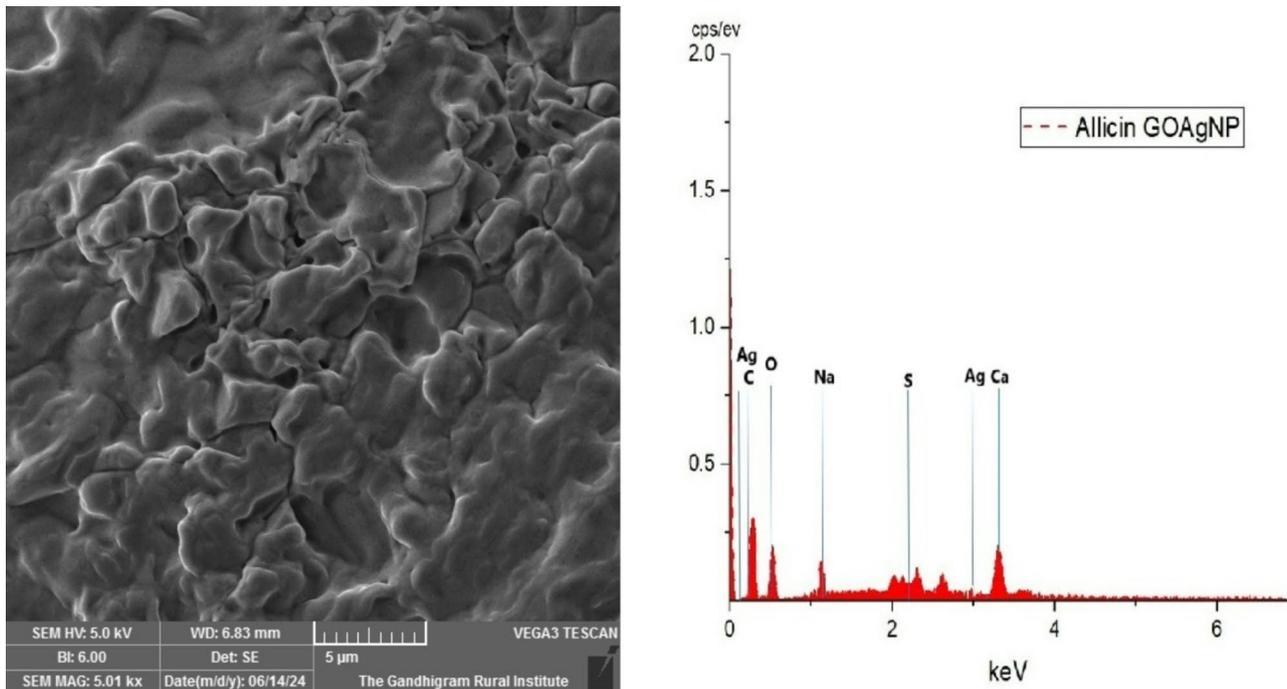
The ANOVA results revealed significant differences in pH values across the different medicament

groups (Allicin GO-AgNP hydrogel, Calcium Hydroxide, Chlorhexidine Gel, and Triple Antibiotic Paste) at the three-time points (Initial, 24 h, and 7 Days) with an F-statistic of 180.74 and  $p < 0.001$ . Post-hoc Tukey's test showed significant differences in Allicin GO-AgNP Hydrogel, Calcium Hydroxide, and Triple Antibiotic Paste at all time points. (Table 1)

The one-way ANOVA revealed significant differences in microhardness between the groups and across different regions ( $p < 0.05$ ). The Tukey HSD post-hoc test showed that the Allicin GO-AgNP hydrogel exhibited significantly higher microhardness than the CaOH and TAP groups across all regions ( $p < 0.001$ ). Additionally, the Control group demonstrated significantly higher microhardness than the CaOH and TAP groups in all regions ( $p < 0.05$ ). However, there was no significant difference between the Control and Allicin GO-AgNP Hydrogel groups in the coronal and middle regions ( $p > 0.05$ ),



**Fig. 1** FTIR graph of allicin incorporated graphene oxide silver nanoparticle hydrogel



**Fig. 2** SEM image and EDX of Allicin incorporated graphene oxide silver nanoparticle hydrogel

**Table 1** Mean pH table for each medicament at different time points

Medications	Initial (pH)	24 h (pH)	7 Days (pH)	Mean pH
Allicin-GO-AgNP hydrogel	7.05	7.12	7.08	7.083
Calcium Hydroxide	12.0	12.45	12.44	12.297
Chlorhexidine Gel	7.0	7.0	7.0	7.0
Triple Antibiotic Paste	12.6	12.7	12.75	12.683

#### Post hoc analysis

Comparison	Mean Difference ( $\Delta$ )	p-Value	Significance
Sodium Alginate vs. Calcium Hydroxide	5.214	< 0.001	Highly Significant
Sodium Alginate vs. Chlorhexidine Gel	0.083	0.91	Not Significant
Sodium Alginate vs. Triple Antibiotic Paste	5.6	< 0.001	Highly Significant
Calcium Hydroxide vs. Chlorhexidine Gel	5.297	< 0.001	Highly Significant
Calcium Hydroxide vs. Triple Antibiotic Paste	-0.386	0.062	Not Significant
Chlorhexidine Gel vs. Triple Antibiotic Paste	5.6	< 0.001	Highly Significant

although the Control group had a slightly higher microhardness in the apical region ( $p < 0.05$ ). (Table 2)

As shown in Fig. 3, the Control and Allicin GO-AgNP Hydrogel groups maintained higher microhardness, especially in the coronal and middle regions, with minimal reduction over 3 months. (Fig. 3) Outliers, especially in the TAP and CaOH groups, suggest variability in dentin response, with some samples experiencing more severe softening.

In the SEM analysis of the root canal surface (supplementary figures S1–S3), the treated group (Allicin GO-AgNP Hydrogel) showed a surface with uniform nanoparticle distribution, maintaining the dentin's structural integrity. EDX analysis confirmed graphene oxide

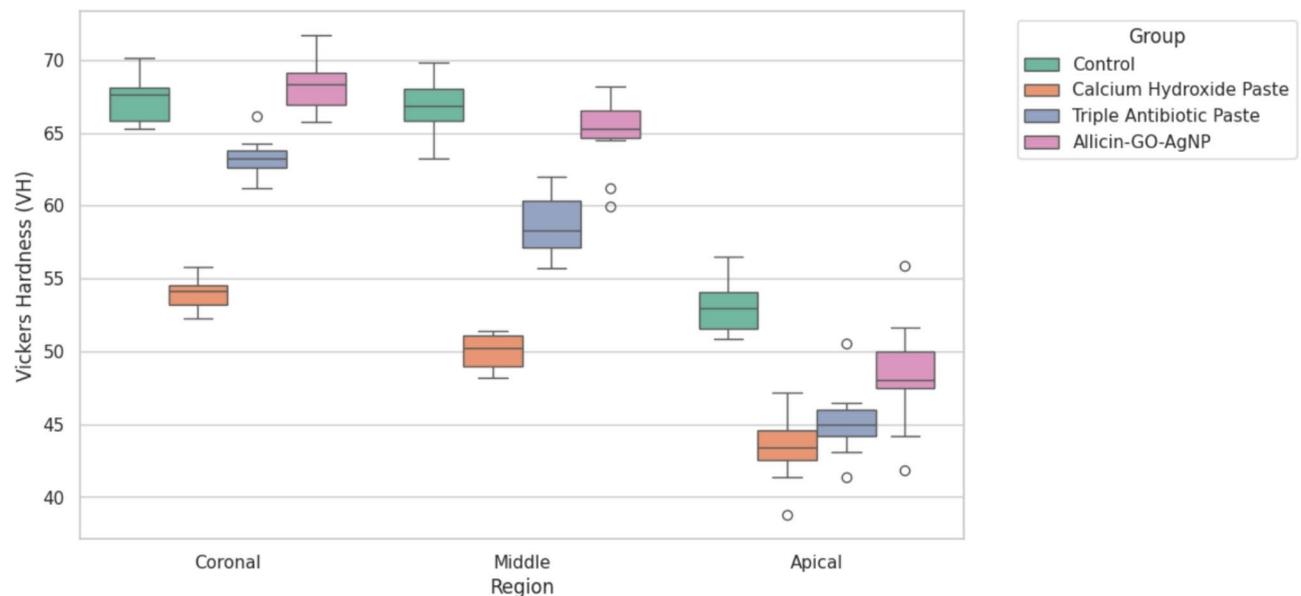
incorporation by slightly higher concentrations of Carbon (C) and Oxygen (O), traces of silver (Ag) and sulfur (S) in the treated dentin, indicating successful incorporation of the hydrogel being deposited on the dentin surface. (Figs. 4 and 5)

The antimicrobial efficacy testing revealed that against *E. faecalis*, Group 2 (Double antibiotic paste) exhibited the largest inhibition zone (37 mm), followed by Group 5 (Allicin GO-AgNP gel) at 20 mm, and Group 1 (Calcium hydroxide) at 10 mm. For *C. albicans*, Group 1 showed the highest inhibition (18 mm), while Group 5 demonstrated 13 mm. These findings indicate that the double antibiotic paste was most effective against *E. faecalis*, while calcium hydroxide exhibited the greatest inhibition

**Table 2** Mean (SD) of microhardness for roots treated with endodontic regeneration medicaments and a control group for 1 week, 1 month, and 3 months

Region	Group	1 Week (VH)	1 Month (VH)	3 Months (VH)	p-value
		Mean ( $\pm$ SD)	Mean ( $\pm$ SD)	Mean ( $\pm$ SD)	
Coronal	Control	66.335 ( $\pm$ 2.38) Ab	65.478 ( $\pm$ 2.385) Aa	66.36 ( $\pm$ 2.385) Aa	0.0174
	Calcium Hydroxide Paste	63.046 ( $\pm$ 1.76) Ab	57.460 ( $\pm$ 1.476) Aa	55.278 ( $\pm$ 1.56) Ba	0.0046
	Triple Antibiotic Paste	67.462 ( $\pm$ 0.46) Aa	63.600 ( $\pm$ 0.982) Aa	63.650 ( $\pm$ 1.71) Aa	0.000096
	Allicin-GO-AgNP	68.607 ( $\pm$ 1.46) Aa	65.480 ( $\pm$ 2.953) Aa	68.795 ( $\pm$ 1.56) Aa	
Middle	Control	67.173 ( $\pm$ 2.33) Ab	64.280 ( $\pm$ 2.331) Aa	67.33 ( $\pm$ 2.331) Aa	0.00019
	Calcium Hydroxide Paste	65.272 ( $\pm$ 0.76) Ab	56.720 ( $\pm$ 0.879) Aa	49.630 ( $\pm$ 1.74) Ba	0.0019
	Triple Antibiotic Paste	58.081 ( $\pm$ 1.76) Bb	55.580 ( $\pm$ 2.385) Aa	58.633 ( $\pm$ 2.46) Aa	0.000063
	Allicin-GO-AgNP	71.792 ( $\pm$ 2.47) Ab	65.700 ( $\pm$ 3.568) Aa	65.113 ( $\pm$ 1.97) Aa	0.359
Apical	Control	57.036 ( $\pm$ 4.54) Bb	54.580 ( $\pm$ 4.547) Ba	52.86 ( $\pm$ 2.47) Cc	0.6504
	Calcium Hydroxide Paste	54.782 ( $\pm$ 3.74) Bb	48.940 ( $\pm$ 4.202) Ca	43.823 ( $\pm$ 3.46) Cc	0.2551
	Triple Antibiotic Paste	54.110 ( $\pm$ 2.36) Bb	51.780 ( $\pm$ 2.852) Ca	45.308 ( $\pm$ 2.76) Cc	0.0269
	Allicin-GO-AgNP	57.266 ( $\pm$ 3.6) Bb	54.800 ( $\pm$ 3.009) Ca	47.950 ( $\pm$ 3.20) Cc	

At each time point, different upper-case letters indicate significant differences between different groups within the same time point p-values indicate the significance of differences among groups at each time point

**Fig. 3** Microhardness of different intracanal medicament after 3 months

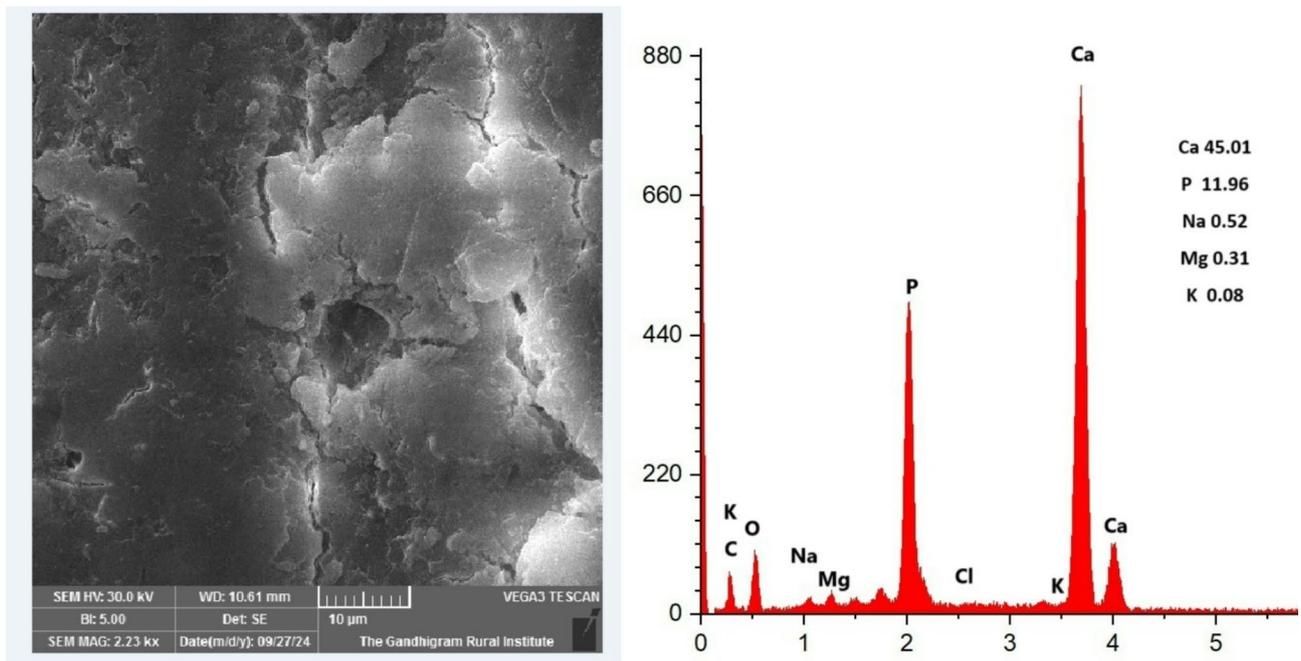
against *C. albicans*. The Allicin GO-AgNP gel displayed moderate antimicrobial activity against both strains. (Fig. 6)

## Discussion

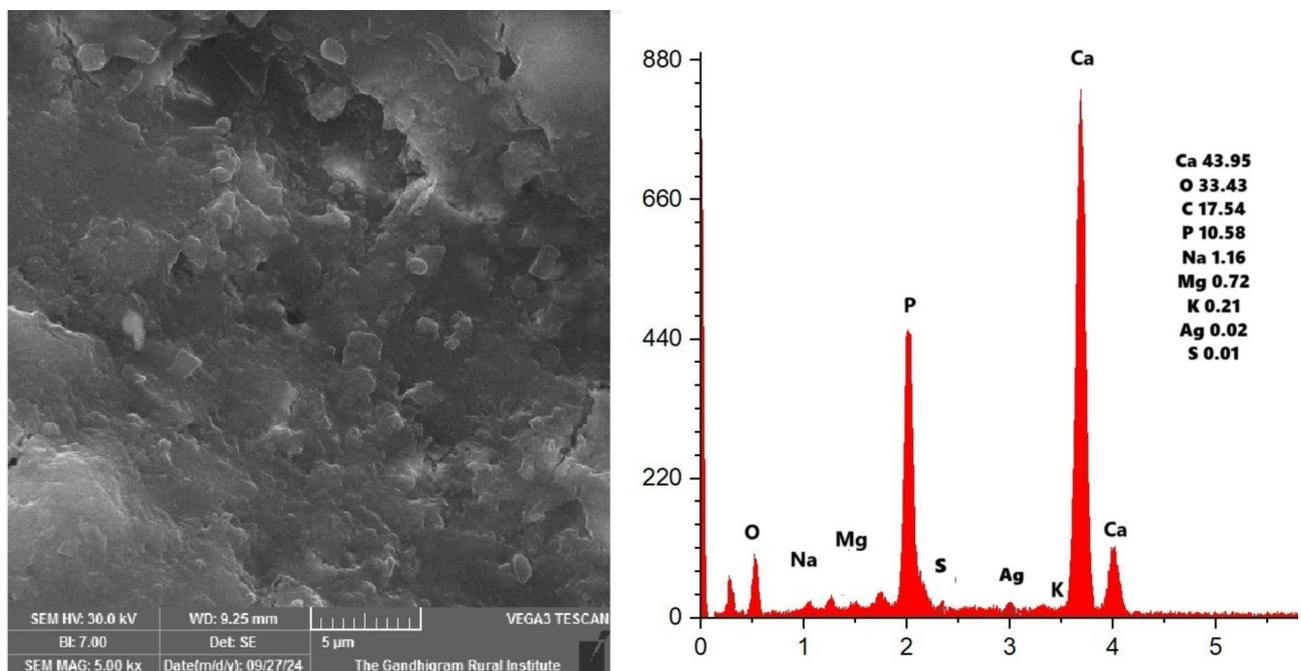
Intracanal medicaments are critical in endodontic therapy, especially in regenerative approaches requiring extended placement periods [12]. Prolonged use of intracanal medicaments is essential to ensure adequate root canal disinfection, prevent recontamination, and combat resistant microorganisms that may have survived the cleaning and shaping processes and promote healing [3, 13]. The application time for these medicaments can vary significantly, often lasting up to 11 weeks in regenerative

endodontics, necessitating the exploration of their impact on the dentin structure [14]. Given the delicate nature of immature teeth, preserving dentin integrity is vital for successful outcomes [15].

Recent advancements in nanotechnology have redefined the possibilities within dentistry, providing innovative solutions that address longstanding challenges in endodontic treatments. Specifically, materials in the form of nanoparticles and hydrogels have gained attention for their superior antimicrobial properties, enhanced bioactivity, and targeted drug delivery capabilities [16]. Graphene, a carbon allotrope, is the thinnest material that can form a crystal lattice without structural dislocations [17]. The antibacterial property of silver nanoparticles



**Fig. 4** SEM image and EDX of dentin without intracanal medicament



**Fig. 5** SEM image and EDX of dentin after Allicin-GO-AgNP hydrogel intracanal medicament

was enhanced by adding graphene, and there were few cytotoxic effects [18].

Hydrogel-based medicaments represent an innovative solution in regenerative therapy, offering a sustained release of bioactive compounds while maintaining the structural integrity of dentin [19]. Sodium alginate in regenerative endodontics is highly promising because it is a sustained release mechanism for the bioactive agents.

At the same time, it creates an environment conducive to cell attachment, proliferation, and differentiation. Specifically, it acts as a scaffold that supports stem cell growth and encourages pulp-dentin complex regeneration [20]. Because such a material is biodegradable, it may permit gradual degradation of a material as new tissue forms and thus is highly suited to endodontic regenerative procedures. Such formulations can effectively deliver



**Fig. 6** Antimicrobial activity of intracanal medicaments against *Enterococcus faecalis* and *Candida albicans* using the disc diffusion method

antimicrobial agents over prolonged periods without the detrimental effects typically associated with traditional medicaments [21, 22].

The use of allicin in this study is particularly noteworthy due to its potent antimicrobial properties, which are essential for effective endodontic treatments. As a natural compound derived from garlic, allicin or allyl thiosulfinate has demonstrated broad-spectrum antibacterial activity against pathogens commonly found in root canal infections. [23] By integrating allicin into the formulation, we have developed a novel biocomposite nanomaterial, Allicin-incorporated GOAgNp (graphene oxide silver nanoparticles). AgNP integration into GO results in enhanced antibacterial capabilities that work synergistically. The AgNP is stabilized by the GO matrix, and this results in enhanced antibacterial activity [9].

The pH of intracanal medicaments significantly impacts dentin's properties. The Allicin-GO-AgNP hydrogel maintained a near-neutral pH (mean 7.083), unlike the highly alkaline Calcium Hydroxide (mean 12.297) and Triple Antibiotic Paste (mean 12.683). This neutral pH minimizes collagen degradation and preserves dentin microhardness, whereas high pH levels can weaken dentin [6]. The stable pH of Allicin-GO-AgNP hydrogel supports its potential as a biocompatible medicament, balancing antimicrobial action with dentin preservation, making it suitable for regenerative endodontics.

Microhardness analysis is a vital method for evaluating the effects of various intracanal medicaments on dentin integrity [24]. It provides insights into the structural

preservation of dentin, which is crucial for the long-term success of endodontic therapies [25]. The results demonstrated a marked reduction in dentin hardness, particularly in the apical region of the calcium hydroxide (CaOH) group. This finding is consistent with White et al. (2002), who reported that prolonged exposure to calcium hydroxide can soften dentin due to its highly alkaline pH [26]. The gradual dissolution of dentin mineral content, specifically calcium phosphate, significantly contributed to this reduction in microhardness [27].

Similarly, the triple antibiotic paste (TAP) group showed a notable reduction in dentin hardness, especially in the middle and apical regions. This observation aligns with the findings of Mozayeni et al. (2014), where TAP, due to its acidic nature and the presence of minocycline, caused significant demineralization of dentin, weakening its structure [28]. The increase in dentine microhardness observed in the samples treated with the graphene-combined silver nanoparticles group might be due to the deposition of silver nanoparticles on the dentine surface and inside the dentinal tubules [10].

In contrast, the Allicin-GO-AgNP hydrogel group demonstrated a relatively consistent microhardness profile across all regions, closely paralleling the control group with no medicament. This finding suggests that the novel hydrogel formulation has a less aggressive impact on dentin than traditional intracanal medicaments. The antimicrobial properties of graphene oxide and silver nanoparticles are effective without significantly altering dentin structure [9]. Additionally, allicin's antioxidant

properties may help reduce oxidative stress on dentin, preserving its mineral content [11].

The SEM analysis confirmed that the Allicin GO-AgNP hydrogel preserved dentin structural integrity. EDX analysis further validated the deposition of the hydrogel by detecting silver (Ag) and sulfur (S) in the treated dentin. This deposition likely contributed to the maintenance of dentin microhardness, as the hydrogel formed a protective layer that reduced mineral loss. Silver nanoparticles in the graphene oxide matrix enhanced this effect by stabilizing the dentin structure [10].

The antimicrobial efficacy of intracanal medicaments is pivotal for eliminating residual pathogens, yet achieving this without compromising dentin integrity remains a critical challenge. In this study, the Allicin-GO-AgNP hydrogel exhibited moderate antimicrobial activity against *E. faecalis* (20 mm) and *C. albicans* (13 mm), lower than DAP or CaOH but beneficial for controlled disinfection. This moderate efficacy is attributed to its composite formulation, which ensures sustained antimicrobial release while minimizing dentin degradation [29, 30]. Unlike DAP or CaOH, which act through rapid antibiotic or pH shifts, the hydrogel's controlled release prevents premature depletion of antimicrobial agents [4, 6, 19]. The hydrogel's antimicrobial action stems from GO-induced membrane disruption, AgNP-mediated enzyme inhibition, and allicin's anti-quorum sensing properties. [18, 23, 31]

These findings reject the null hypothesis and say that incorporating nanotechnology-based materials such as Allicin-GO-AgNP hydrogels into endodontic protocols could improve treatment outcomes by providing effective disinfection while preserving dentin integrity. This innovative biocomposite material minimizes potential adverse effects on dentin structure while promoting healing, making it a promising candidate for regenerative endodontics.

While this study offers valuable insights, the sample size and the laboratory conditions may limit the generalizability of the results. Further research should explore the long-term effects of these nanotechnology-based medicaments under clinical conditions, including their impact on biocompatibility and patient outcomes. Additionally, future studies could investigate the synergistic effects of combining nanomaterials with other bioactive agents to enhance both antimicrobial and regenerative potential.

## Conclusion

This study highlights the potential of the novel allicin-incorporated graphene oxide-silver nanoparticle (Allicin-GO-AgNP) hydrogel in preserving dentin microhardness compared to conventional intracanal medicaments like calcium hydroxide and triple antibiotic paste. While the

hydrogel maintained near-neutral pH and demonstrated superior structural preservation of dentin, future studies are required to validate its long term antimicrobial properties.

## Supplementary information

The online version contains supplementary material available at <https://doi.org/10.1186/s12903-025-05870-1>.

Supplementary Material 1

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## Author contributions

R.P, M.R and M.I.K made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation. R.P, A.S, M.F and M.I.K took part in drafting, revising, or critically reviewing the article; all the authors gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

## Funding

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## Data availability

The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

## Ethics declarations

### Ethics approval and consent to participate

The study was initiated after getting ethical approval from the Institutional Review Board of Saveetha Dental College and Hospital, SIMATS, Chennai, Tamil Nadu, India, with an approval number SRB/SDC/PhD/ENDO-2364/24/TH-014. The study was carried out following the Declaration of Helsinki. Consent to participate is not applicable as no human participants were involved in the study.

### Consent for publication

Not applicable.

### Competing interests

The authors declare no competing interests.

### Clinical trial number

Not applicable.

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