

Effect of three different intracanal medications on canal wall cleanliness and radicular dentin microhardness

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Background/aim

Complete removal of intracanal medication from the canals before root canal obturation is an essential step. This research was conducted to comparatively assess the remaining debris after the removal of three intracanal medications: silver nanoparticles (AgNPs) paste, silver nanoparticles (AgNPs) with curcumin paste, and calcium hydroxide (Ca(OH)₂) paste on the root canal walls and their effect on microhardness of radicular dentin.

Materials and methods

Thirty human extracted single-rooted teeth were selected from the National Research Centre Dental Clinic, Egypt. The crowns of the teeth were decapitated at the cemento-enamel junction. All root canals were mechanically prepared. The specimens were randomly divided into three groups (each = 10) according to the type of medication used. Group A: AgNPs paste. Group B: AgNPs with curcumin paste. Group C: Ca(OH)₂ paste. Intracanal medications were injected into the root canals and all specimens were incubated at 37°C for 7 days, then the intracanal medications were removed from the root canals and specimens were longitudinally sectioned. One-half of each specimen was examined under a stereomicroscope to evaluate the amount of remaining debris. The other half of each specimen was used to measure dentin microhardness using the Vickers Microhardness Tester. Statistical analysis was done.

Results

AgNPs paste showed a significantly low mean value of the remaining debris than AgNPs with curcumin paste, while Ca(OH)₂ paste had a significantly highest mean value of the remaining debris than AgNPs paste alone or mixed with curcumin. AgNPs paste had the highest significant mean value of microhardness, then AgNPs with curcumin paste, while Ca(OH)₂ paste had a significantly lowest mean value of microhardness.

Conclusion

AgNPs paste and AgNPs with curcumin paste used as intracanal medications were removed from the root canals with a minimal amount of the remaining debris and less effect on dentin microhardness than the Ca(OH)₂ paste.

Keywords:

calcium hydroxide, curcumin, microhardness, remaining debris, silver nanoparticles

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Introduction

The primary cause of pulp and periradicular infection is microorganisms [1]. Root canal anatomy complexity makes achieving complete canal disinfection through instrumentation alone a challenge [2]. Irrigation and intracanal medicaments are considered adjunctive aids that are needed to completely eradicate microorganisms from the root canal allowing for periapical tissue healing [3]. Effective root canal disinfection during endodontic treatment procedures promotes its success and improves its prognosis [4].

The most routinely utilized intracanal medicaments is calcium hydroxide (Ca(OH)₂) paste because it has a bactericidal effect, high alkalinity, tissue repair power, and osteoclastic activity inhibition features [1]. There

are several disadvantages associated with Ca(OH)₂ paste as the root canal medication. Results of Andreasen *et al.* [5] stated that long-term use of Ca(OH)₂ paste as an intracanal medication causes weakening and decreases the fracture strength of hard tooth structure.

Recently, nanoparticles are introduced as a newer medicament class. They are hypothesized to have an antibacterial effect. Because of their small nano-size particles, they could cause interruption of the bacterial

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biofilm. Its nano size affords a larger outer surface area and through adsorption of other medications, it exerts stronger antimicrobial activity. Nanoparticles pose a polycationic charged structure that permits it to interact with negatively charged bacterial cell walls and stimulate antibiotic interaction with the microorganisms [6]. Silver nanoparticles (AgNPs) were reported to have antimicrobial effects against most bacterial species, fungi, and viruses. Its antibacterial effect is mediated by preventing the adhesion of microorganisms and consequently biofilm formation inhibition [7].

Although chemical medicaments have potent action, their demerits such as inflammation, hypersensitivity reactions, cytotoxicity, and development of bacterial resistance strains are very serious [8].

Taking into consideration safety and lack of or few side effects, investigators are attempting to provide more biocompatible and patient-friendly alternatives. Natural compounds are gaining great significance because they have anti-inflammatory, antimicrobial properties, biocompatibility of their active ingredients, in addition to simplicity of availability, and cost-effectiveness [9]. Research studies on herbal compounds that are advocated for dental applications demonstrated their valuable pharmacological features [3].

Curcumin is a polyphenolic material extracted from *Curcuma longa*, a dietary spice commonly called turmeric. It is the main yellow bioactive component that has anti-inflammatory, antiviral, and antibacterial activities [10]. It was found that AgNPs paste and AgNPs with curcumin paste have a more potent antibacterial effect than $\text{Ca}(\text{OH})_2$ paste against *Enterococcus faecalis*, while AgNPs with curcumin paste had the best antibacterial effect [11].

Complete removal of intracanal medication from the canals before root canal obturation is an essential step. The presence of medication remnants on root canal walls compromises the sealing of the canal filling as it could prevent the penetration of sealer deeply through dentinal tubules. Also, the interaction between root canal medications and sealer might occur, which could affect the setting of the sealer and compromise its sealing ability [12].

Before the use of any newly introduced products, adequate determination of the material's properties is obligatory. The application of root canal medication during the treatment procedure was found to affect the physical properties of the root dentin and consequently

its fracture resistance [13,14]. Investigatory studies are mandatory to evaluate the validation of the use of natural medical compounds on the hard tooth structure [15].

Composition and surface changes in radicular dentin by commonly used chemicals like intracanal medications and irrigants have been researched [16]. Changes in the mineral content of dentin could be estimated by measurement of the microhardness of dentin. It is rated as the simplest nondestructive mechanical characterization method [17].

Therefore, our research was conducted aiming to comparatively evaluate the amount of debris that remained after the removal of three intracanal medications: AgNPs paste, AgNPs with curcumin paste, and $\text{Ca}(\text{OH})_2$ paste on root canal walls and their effect on the microhardness of radicular dentin.

Materials and methods

Materials

This research was conducted on AgNPs paste (NanoTech, Giza, Egypt), AgNPs with curcumin paste (NanoTech, Giza, Egypt), and $\text{Ca}(\text{OH})_2$ paste (Metapaste, Meta Biomed) used as intracanal medication.

Calculation of sample size

The calculation of the sample size was done using the study of Aly *et al.* [3] as a reference. Ten per group was the minimally accepted sample size according to the previously mentioned study, when the response within each subject group was normally distributed with a standard deviation 8.64. The estimated mean difference was 11.5, when the power was 80% and the type I error probability was 0.05. The sample size was calculated by using the PS Power version 3.1.6 using an independent t test.

Study design

Thirty extracted human teeth with a single root canal and closed apex were chosen. They were carefully examined; as teeth with cracks, fractures, resorption, caries, or obstructed canals were discarded. Teeth were obtained from the National Research Centre Dental Clinic, Egypt. They were randomly classified into three main groups (each=10) depending on the type of medication as follows:

- (1) Group A: Ten teeth were treated with AgNPs paste.
- (2) Group B: Ten teeth were treated with AgNPs with curcumin paste.

- (3) Group C: Ten teeth were treated with $\text{Ca}(\text{OH})_2$ paste.

Intracanal medications were inserted into root canals for 7 days.

Ethical approval

The present study was conducted with the Code of Ethics of the World Medical Association, according to the principles expressed in the Declaration of Helsinki. This study has been approved by the local Ethics Committee of the National Research Center under approval number 4439102021.

Methods

Method of AgNPs paste and AgNPs with curcumin paste preparation

AgNPs paste was prepared by the chemical reduction technique by Pal *et al.* [18]. This includes microwave irradiation of silver nitrate (AgNO_3) solution in ethanolic climate in the presence of polyvinylpyrrolidone (PVP) as a stabilizing agent, while ethanol acts as a reducing agent in the existence of microwave. The solvent was evaporated at low temperatures to obtain silver in powder form. Then the suspension was diluted with carboxymethyl cellulose (CMC) aqueous paste 5% w/v (1 ml silver suspension + 4 ml CMC) to get 100 ppm paste. AgNPs with curcumin paste were prepared by dispersion of 5 mg curcumin in 1 ml silver emulsion before adding the CMC paste.

Preparation of teeth specimens

Calculus and soft tissue remnants were removed from the teeth and then stored in saline until used. All teeth's crowns were decapitated at the cemento-enamel junction by a tapered fissure bur operated by high-speed motor under coolant adjusting root lengths to be 15 mm ($\pm$) 1 mm.

Preparation of root canals

The root canals were mechanically prepared using rotary system REVO-S (RS; Micro-Mega SA, Besançon, France) till size AS35. Irrigation was done with 1 ml of 2% sodium hypochlorite (NaOCl) subsequent to every file. Finally, root canals were rinsed with 3 ml ethylenediaminetetraacetic acid (EDTA) 17%, 3 ml 2% NaOCl , and then final irrigation with 3 ml saline solution. Root canals were dried with paper points.

Injection of medications in root canals

A plastic syringe was used to inject AgNPs paste and AgNPs with curcumin paste into the canals, while the $\text{Ca}(\text{OH})_2$ paste was injected by the nozzle of its

injectable tube. The nozzle was inserted inside canals till it reached 2 mm short of the working length, and the medication was slowly injected into the canals till the plane of the cemento-enamel junction. The orifices of all specimens were sealed by Cavit (3MESPE, USA). Each specimen was placed in Eppendorf tube, labeled and incubated at 37 °C for 7 days.

Removal of medications from root canals

After 7 days all temporary restorations were removed, and the intracanal medication pastes were washed away by irrigation of the canal space with five ml of saline using a disposable syringe.

Then roots were sectioned longitudinally by making a groove along the sides of the root using a diamond fine stone mounted on a high-speed handpiece without penetrating the root canal space. All roots were sectioned into two halves with a chisel and mallet to split the root.

Evaluation of the remaining debris after removal of intracanal medications

One-half of each root was examined under a stereomicroscope at a magnification of 25X. The remaining debris of intracanal medication was calculated as the percentage of root canal area covered by the remaining intracanal medication using the image analysis software (ImageJ NHS, USA) at the apical, middle, and coronal thirds.

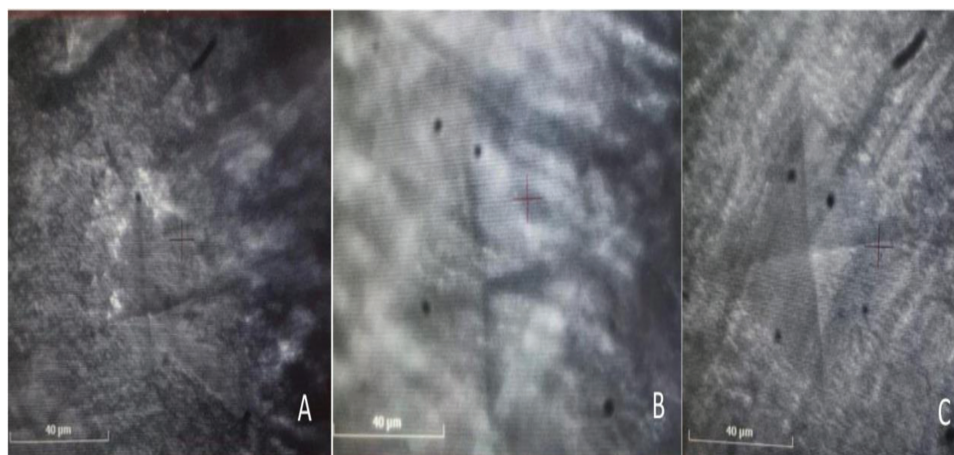
Evaluation of the effect of intracanal medications on radicular dentin microhardness

The other half of every root was mounted horizontally on an acrylic resin block (Acrostone, Dent Product, Egypt) so that the dentinal wall of the root canal is facing upward. Microhardness assessment was performed by the Vickers Microhardness Tester (Model LM-100, FM 1159 LECO Corporation Michigan, USA). Microhardness was measured at the apical, middle, and coronal thirds at 40X magnifications with 100 gm load and 15 sec dwell time as shown in Fig. 1. Three measurements were recorded for each third, and their average value was listed as the representative hardness value for data analyses.

Statistical analysis

Statistical analysis was performed using SPSS 20®, GraphPad Prism®, and Microsoft Excel 2016. All data were tested for normality by using the Shapiro–Wilk and Kolmogorov–normality test and presented as the mean difference and standard deviation (SD) records. All data were presented in one table and one graph for each test. Comparison

Figure 1



Vicker's indenter photographs on coronal sections of the root after application of the AgNPs paste (Group A), AgNPs with curcumin paste (Group B), and $\text{Ca}(\text{OH})_2$ paste (Group C) intracanal medications.

between different groups was performed by using one-way ANOVA test followed by Tukey's post hoc test for multiple comparisons.

Results

Evaluation of the remaining debris after removal of the intracanal medications

Comparison between different groups

Comparison between different groups A, B, and C was performed using one-way ANOVA test, which revealed a significant difference between all groups, at all sections where $P < 0.0001$, as represented in Table 1 and shown by the stereomicroscope image in Fig. 2 and bar chart in Fig. 3.

Coronal third: AgNPs paste showed significantly the lowest mean value of the remaining debris (0.40 ± 0.08), and then AgNPs with curcumin paste (1.02 ± 0.20), while the $\text{Ca}(\text{OH})_2$ paste had significantly the uppermost mean value (4.9 ± 0.52).

Middle third: AgNPs paste and AgNPs with curcumin paste had a significantly minimal mean value of the remaining debris (9.33 ± 0.54) and (9.03 ± 0.63), respectively, with insignificant differences among them, while the $\text{Ca}(\text{OH})_2$ paste had significantly the uppermost mean value (14.09 ± 0.75).

Apical third: AgNPs paste showed the significantly lowest mean value of the remaining debris (11.37 ± 0.72), and then AgNPs with curcumin paste (14.46 ± 0.75), while the $\text{Ca}(\text{OH})_2$ paste had significantly the uppermost mean value (17.43 ± 0.67).

Comparison between different sections

Comparison between different sections was performed by using one-way ANOVA test, which revealed a significant difference between them in all groups at $P < 0.0001$. In all groups, the coronal section had significantly the minimal mean value of the remaining debris in the root canals, and then the middle section, while the apical section had the highest mean value of debris, as represented in Table 1 and shown by the stereomicroscope image in Fig. 2 and bar chart in Fig. 3.

Evaluation of the effect of intracanal medications on dentin microhardness

Comparison between different groups

Comparison between different groups was performed using the one-way ANOVA test, which revealed a significant difference between all groups at all sections as $P < 0.0001$, as shown in Table 2 and Fig. 4.

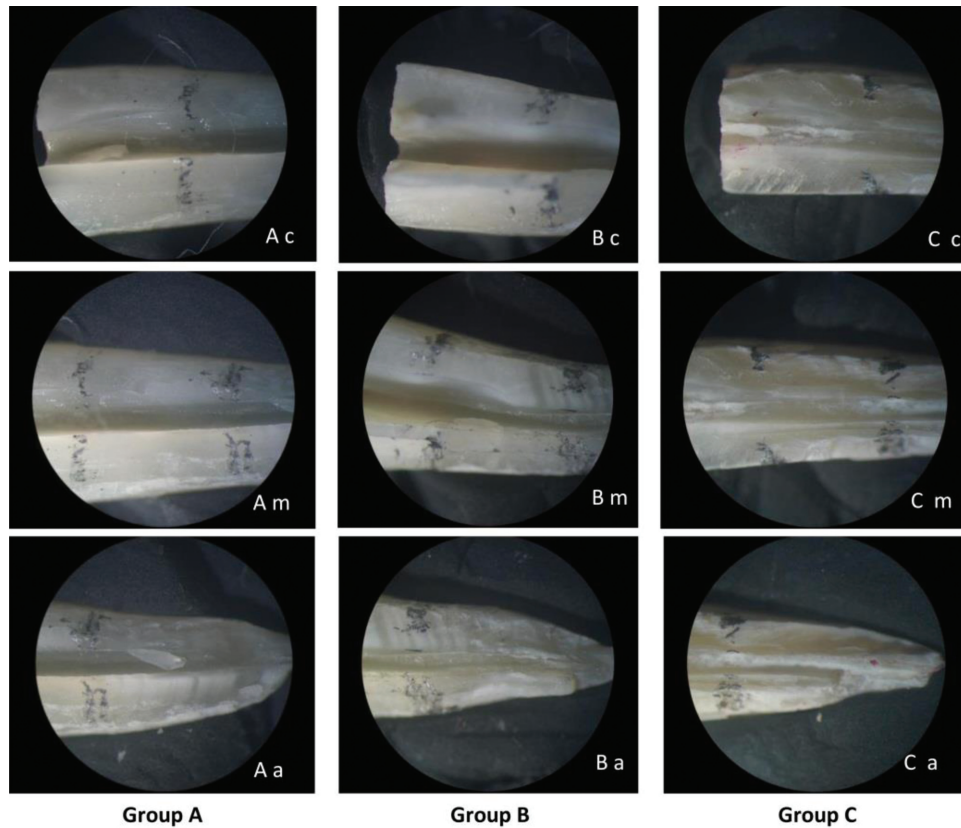
Coronal third: AgNPs paste had the significantly highest mean value of microhardness (63.83 ± 2.39),

Table 1 Mean and standard deviation of the remaining debris in all groups and sections

Remaining debris	Group A	Group B	Group C	P value
Coronal	0.40 ± 0.08^{ad}	1.02 ± 0.20^{bd}	4.90 ± 0.52^{cd}	$< 0.0001^*$
Middle	9.33 ± 0.54^{ae}	9.03 ± 0.63^{ae}	14.90 ± 0.75^{be}	$< 0.0001^*$
Apical	11.37 ± 0.72^{af}	14.46 ± 0.75^{bf}	17.43 ± 0.67^{cf}	$< 0.0001^*$
P-value	$< 0.0001^{**}$	$< 0.0001^{**}$	$< 0.0001^{**}$	

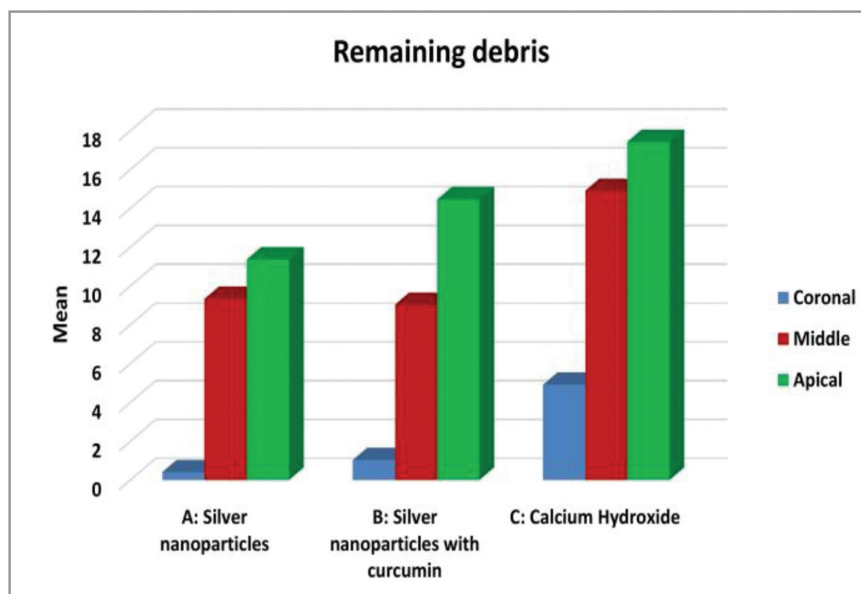
All data are expressed as mean $\pm$ SD. *: All data with different superscript letters in the same row (a, b, c) were significantly different as P -value < 0.05 , using the ANOVA test. **: All data with different superscript letters in the same column (d, e, f) were significantly different as P -value < 0.05 , using the ANOVA test.

Figure 2



Stereomicroscope image of coronal (c), middle (m), and apical (a) sections of the root canal after removal of AgNPs paste (Group A), AgNPs with curcumin paste (Group B), and Ca(OH)₂ paste (Group C).

Figure 3



Bar chart showing a comparison of intracanal medications remaining debris between different groups and sections.

and then AgNPs with curcumin paste (53.02 ± 2.72), while the Ca(OH)₂ paste had the least mean value significantly (44.01 ± 2.19).

Middle third: AgNPs paste showed significantly the highest mean value of microhardness (66.01 ± 2.49), and then AgNPs with curcumin paste (54.75 ± 1.92), while

the $\text{Ca}(\text{OH})_2$ paste had significantly minimal mean value (46.76 ± 1.95).

Apical third: AgNPs paste and AgNPs with curcumin paste had significantly the uppermost mean value of microhardness (74.64 ± 2.98) and (60.25 ± 3.81), respectively, with an insignificant difference among them, while the $\text{Ca}(\text{OH})_2$ paste had significantly the lowest mean value (45.04 ± 3.64).

Comparison between different sections

Comparison between different sections was performed using the one-way ANOVA test, which revealed a significant difference between them in all groups, as shown in Table 2 and Fig. 4.

AgNPs paste (Group A) and AgNPs with curcumin (Group B): in both groups, there was a significant difference between sections as $P < 0.0001$. The coronal and middle sections had significantly the minimal mean value with insignificant differences among them, while the apical section was significantly the highest.

$\text{Ca}(\text{OH})_2$ paste (Group C): There was an insignificant difference between all sections as $P = 0.09$.

Discussion

The target of this research was to assess the remaining debris after removal of three intracanal medications: AgNPs paste, AgNPs with curcumin paste, and $\text{Ca}(\text{OH})_2$ paste from root canal walls and their effect on root canal dentin microhardness.

Successful endodontic treatment intends to get rid of all types of intracanal microorganisms, which are the prime reason for root canal treatment failures [19]. This aim was achieved using multiple modalities of root canal decontaminants. For example; root canal irrigants such as NaOCl and medications such as the $\text{Ca}(\text{OH})_2$ paste [20].

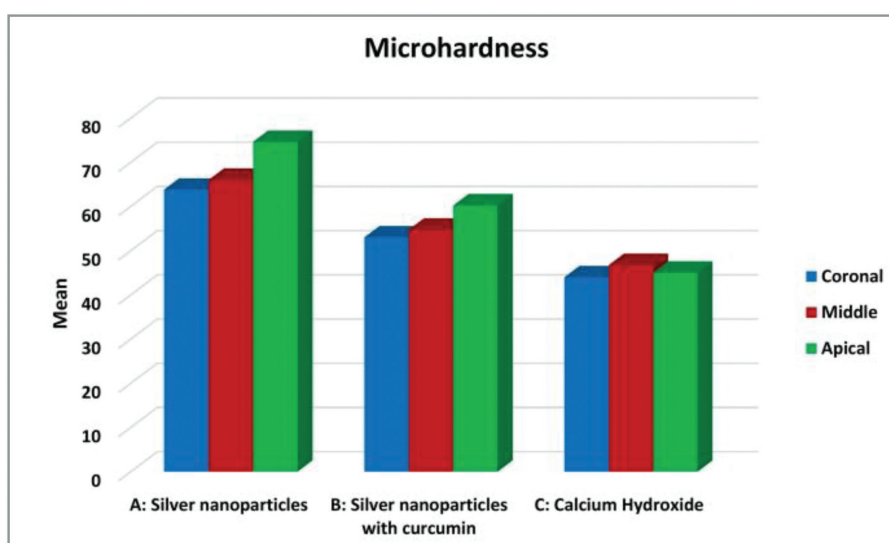
Nanoparticles and herbal natural products stormed the world of dentistry owing to their great capability in the elimination of the bacterial biofilm [21]. Many researches have shown the antibacterial effect of the

Table 2 Comparison between microhardness in all groups and sections

Microhardness	Group A	Group B	Group C	P value
Coronal	$63.83 \pm 2.39^{\text{ad}}$	$53.02 \pm 2.72^{\text{bd}}$	$44.01 \pm 2.19^{\text{cd}}$	$< 0.0001^*$
Middle	$66.01 \pm 2.49^{\text{ae}}$	$54.75 \pm 1.92^{\text{be}}$	$46.76 \pm 1.95^{\text{ce}}$	$< 0.0001^*$
Apical	$74.64 \pm 2.98^{\text{af}}$	$60.25 \pm 3.81^{\text{af}}$	$45.04 \pm 3.64^{\text{bf}}$	$< 0.0001^*$
P-value	$< 0.0001^{**}$	$< 0.0001^{**}$	$< 0.0001^{**}$	

All data are expressed as mean $\pm$ SD. *: All data with different superscript letters in the same row (a, b, c) were significantly different as P -value < 0.05 using the ANOVA test. **: All data with different superscript letters in the same column (d, e, f) were significantly different as P -value < 0.05 , using the ANOVA test.

Figure 4



Bar chart showing microhardness comparison between different groups and sections.

AgNPs paste, as it targeted many areas in the microbial cell such as plasmids, enzymes, and cell envelope [7]. It also decreased bacterial adhesion to the root wall dentin [22]. Curcumin is a kinetic ingredient of turmeric [23]. It has anti-inflammatory and antibacterial leverage by inhibiting the proliferation and growth of several types of microorganisms [24]. $\text{Ca}(\text{OH})_2$ paste was selected as the baseline for the sake of its popular clinical application and its antibacterial efficacy [25].

Thorough elimination of root canal medication before the obturation process is necessary because remnants of medicaments on the canal wall will prevent the optimum sealing and hence renders root canal therapy success [26].

The findings of our research showed that in coronal and apical sections the significantly least amount of the remaining intracanal medication debris was presented in AgNPs paste (Group A), followed by AgNPs with curcumin paste (Group B), then $\text{Ca}(\text{OH})_2$ paste (Group C), while in middle sections of groups treated with AgNPs paste alone or mixed with curcumin were significantly lower than the group treated with $\text{Ca}(\text{OH})_2$ paste with an insignificant difference between them. To date, no studies have evaluated the debris remaining in root canals subsequent to the removal of curcumin and AgNPs intracanal medications. However, several studies have evaluated their antimicrobial effect [7,22–24].

The highest amount of intracanal medication remaining debris was presented in group C treated with $\text{Ca}(\text{OH})_2$ paste in the three root canal sections. This result coincided with the results of Contardo *et al.* [27], who reported that $\text{Ca}(\text{OH})_2$ intracanal medicament had a lot of remaining debris penetrating through dentinal tubules after its removal, which lead to the reduction of the sealing capability between the canal wall and the obturation material. Also, with the findings of Kim and Kim [28], who concluded that root canals medicated with $\text{Ca}(\text{OH})_2$ paste presented high apical leakage records when filled with zinc oxide eugenol sealer.

Vickers microhardness testing was chosen in this research as it measures small samples with high accuracy [29]. The outcome of the study proved that the microhardness of the group treated with AgNPs paste (Group A) was greater than AgNPs with curcumin paste (Group B) with significant differences in two segments of the root, coronal and middle, while with insignificant difference in the apical

portion. Although to this date, there is no record of the effect of intracanal medication containing curcumin on dentin microhardness, these results could be because of the precipitation of nanoparticles of silver on the root canal wall and its permeation deeply through dentinal tubules. This led to increasing the records of microhardness of dentin as reported by Hiraishi *et al.* [30], who proved that 3.8% silver diamine fluoride was sediment on the surface of the root dentin and permeated up to 40 mm deeply into dentinal tubules. Furthermore, our findings may be related to the variation in charge allocation on the cationic portion among the canal surface and particle surface of nanosilver [31,32]. Our findings were in agreement with the research of Hassan and Khallaf [33], who demonstrated that AgNPs intracanal medicament showed higher microhardness values than the $\text{Ca}(\text{OH})_2$ paste.

The present results indicated that the microhardness of $\text{Ca}(\text{OH})_2$ paste showed significantly lowest values than the group of AgNPs paste and the group of AgNPs with curcumin paste in coronal, middle, and apical sections. These results may be owing to the proteolytic action of the $\text{Ca}(\text{OH})_2$ paste as it causes disturbance in links found among crystals of hydroxyapatite and collagen fibrils, which might be the reason of the reduction in dentin microhardness [34]. Our finding matched the finding of Seyed *et al.* [35] who demonstrated that $\text{Ca}(\text{OH})_2$ medication produced structural variations in the dentin of the root canal, which led to a decrease in its microhardness. Our finding was in contrast with the findings of Doyon *et al.* [36], who demonstrated that there were no significant differences in fracture resistance values recorded between roots treated with $\text{Ca}(\text{OH})_2$ paste and roots treated with saline after 30 days of exposure. This might be probably related to using dissimilar testing methods.

Conclusion

AgNPs paste and AgNPs mixed with curcumin paste could be considered as a promising intracanal medications as they were removed from root canals with a minimal amount of debris and with less effect on dentin microhardness compared with the $\text{Ca}(\text{OH})_2$ paste.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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