

Original Article

Evaluation of the Effect of Nitrofurantoin Paste as an Intracanal Medicament on the Chemical Structure of Radicular Dentine

Mewan S. Abdulrahman^{1*}, Bestoon M. Faraj¹, Kawa F. Dizaye²

Abstract

Objective: To investigate the effects of a new intracanal medicament (nitrofurantoin paste) on radicular dentin's chemical structure compared with modified triple antibiotic paste (MTAP).

Methods: A total of 9 extracted human maxillary canine teeth were used. After cutting the crowns, the root length was standardized to 15 mm. Three 5 mm root cylinders (coronal, middle, and apical) thirds were obtained. Each cylinder was then sectioned longitudinally across the root canal's maximum diameter, resulting in six half-root specimens; two coronal halves, two middle halves, and two apical halves. Then each specimen was divided into three groups. Group 1: 25mg/mL Nit paste, Group 2: 25mg/mL MTAP, and Group 3: (Untreated). Fourier transform infrared spectrophotometer (FTIR) with a diamond attenuated total reflectance (ATR) accessory was used to obtain infrared spectra to analyze dentin specimens. The data were analyzed statistically using one-way ANOVA and Tukey test, and significant differences were used for statistical analyses.

Results: Dentin thirds treated with MTAP had a lower phosphate/amide I ratio ($p < 0.05$) than Nit paste compared with the untreated group. Moreover, the phosphate/amide I ratio was highest in the apical third and decreased to the lowest value in the coronal third. The differences among all three-thirds were statistically significant.

Conclusions: According to the current study results, at the concentration of (25 mg/mL), nitrofurantoin paste minimizes the reduction of phosphate/amide I ratio of the radicular dentine compared with the 25mg/mL MTAP.

Keywords: *Intracanal medicament, chemical structure, Modified triple antibiotic paste, Nitrofurantoin.*

Submitted: March 23, 2021, Accepted: June 6, 2021, Published: December 1, 2021.

Cite this article as: Abdulrahman MS, Faraj BM, Dizaye KF. Evaluation of the effect of nitrofurantoin paste as an intracanal medicament on the chemical structure of radicular dentine. *Sulaimani Dent J.* 2021;8(2):26-33.

<https://doi.org/10.17656/sdj.10137>

1. Department of Conservative Dentistry, College of Dentistry, University of Sulaimani, Sulaimani, Iraq.
2. Department of Basic Sciences, College of Medicine, Hawler Medical University, Erbil, Iraq.

* Corresponding author: mewan.abdulrahman@univsul.edu.iq

Introduction

Intracanal medicament involves the local application of the antibacterial agent to eliminate bacteria in each part of the root canal system, which conventional root canal treatment procedures such as instrumentation and irrigation⁽¹⁾. *Enterococcus faecalis* is the most predominant microorganism, leading to persistent periradicular lesions and endodontic failure^(2,3). *Enterococcus faecalis* is found in root canal failures in nearly about (67–77%)⁽⁴⁻⁶⁾. Modified triple antibiotic paste (MTAP), as an intracanal medicament used in endodontics, comprises three antibiotics (metronidazole, ciprofloxacin, and minocycline). MTAP is a modification of TAP by replacing minocycline with clindamycin to prevent crown discoloration⁽⁷⁻⁹⁾. MTAP was as effective as TAP in reducing *Enterococcus faecalis* in the root canal system⁽¹⁰⁾; however, there is a controversy between the studies supporting its efficacy to eradicate *Enterococcus faecalis* in the root canal system⁽¹¹⁻¹³⁾, which may be due to emerging bacterial resistance^(14,15).

On the other hand, Nitrofurantoin (Nit) is a synthetic nitrofurantoin compound⁽⁶⁾ and is widely utilized as an oral antibiotic therapy for urinary tract infections (UTIs)⁽⁷⁾. Nit is the drug of choice for treating infections caused by multidrug-resistant pathogens^(18,19). Several utilized studies confirmed that Nit is highly effective against *Enterococcus faecalis*⁽²⁰⁻²²⁾. As an intracanal medicament at a 25 mg/mL concentration, Nitrofurantoin was found to be highly effective in eliminating *Enterococcus Faecalis* from the root canal system than MTAP⁽²³⁾. Any intracanal medicament used in endodontics may negatively affect radicular dentin's physical and mechanical properties⁽²⁴⁾. Several studies demonstrated that TAP and MTAP resulted in significant reductions in microhardness of radicular dentin, resulting in demineralization of dentine surface and changes in the chemical structure of dentine⁽²⁵⁻²⁷⁾. No previous study was found to assess the effect of Nitrofurantoin paste (Nit) on the chemical composition of the radicular dentin; therefore, this study was designed to evaluate the effect of 25 mg/mL Nit paste as an antibiotic intracanal medicament on the chemical composition of superficial radicular dentin and compare the results with that of 25 mg/mL MTAP, using Attenuated Total Reflection Fourier Transform Infrared Spectroscopy (ATR-FTIR). To investigate the effects of 25 mg/mL Nit paste on the chemical structure of radicular dentin in comparison with 25 mg/mL modified triple antibiotic paste (MTAP).

Materials and methods

Specimens preparation

A total of 9 extracted human maxillary canine teeth with the following criteria: roots were completely formed, without calcifications or resorption internally, and no exaggerated root curvatures. They were extracted for periodontal reasons. The canine teeth selected were from patients aged 40 to 50 years. The selection of those teeth was, according to morphological and dimensional similarity.

A periodontal curette was used to remove soft tissue remnants, debris, and calculus on the root surfaces. The teeth were stored in normal saline at room temperature; radiographic confirmation of a patent single canal is done in a proximal view. The crowns were cut at the cement-enamel junction perpendicularly to the long axis of the teeth with a rotary double-faced diamond disc (15LC Diamond Wafering Blade, Buehler, Illinois, USA) under continuous water cooling. The root length was cut and standardized to 15 mm. Three 5 mm root cylinders (coronal, middle, and apical) thirds were obtained using a water-cooled rotary double-faced diamond disc. Then, each cylinder was sectioned longitudinally across the root canal's maximum diameter resulting in six half-root specimens; two coronal halves, two middle halves, and two apical halves as shown in Figure 1, A; thus, six specimens were obtained from each root.

To flatten the surfaces, each specimen's pulpal and convex sides were leveled under continuous water-cooling using 1200-grit silicon carbide grinding paper. For smear layer removal from the dentine surface of the specimens, rinsing with 17% EDTA (ethylenediaminetetraacetic acid) (Root canal preparation solution, D Line. Estonia. Europe) and 5.25% NaOCl (Sultan Healthcare, Pennsylvania, USA) had been done. As a final step, each surface was rinsed with 10 mL of sterile normal saline to remove the remnants of EDTA and NaOCl.

Preparation of intracanal medicaments Four pure antibacterial powders were used; Nitrofurantoin (Nit) (Procter & Gamble Company, Cincinnati, Ohio, USA), ciprofloxacin, metronidazole, and clindamycin (Skywalk Pharmacy, Wauwatosa, Wisconsin, USA) to calculate the required amount of the antibacterial powder, an analytical balance Sartorius Entris 64-1S (Analytical Balance 60x0.1mg, Sartorius Lab

Instruments GmbH & Co.KG. Goettingen. Germany), was used.

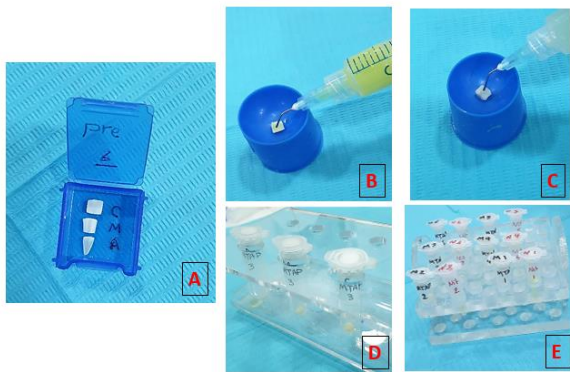


Figure 1: A. Three sectioned thirds of one side of the root, (C=coronal, M=middle and A=apical) thirds, B. Application of nitrofurantoin paste on the specimen. C. Application of modified triple antibiotic paste (MTAP) on the specimen. Specimens in a small conical sample container. E. The medicament groups.

Group Nit (Nit paste): nitrofurantoin solution was prepared by mixing pure powder of Nitrofurantoin with distilled water (AstraZeneca, Boston, Massachusetts, USA), Methylcellulose powder (MC) (Sigma-Aldrich, Chemie GmbH Schnellendorf, Germany) was added to the nitrofurantoin solution to get a thick paste-like consistency mixture⁽²⁸⁾.

Preparation of 25 mg/mL Nit paste: 25 mg (nitrofurantoin) + 1 mL (distilled water) + 80 mg (MC) to prepare 25 mg/mL Nit paste⁽²³⁾.

Group (MTAP): MTAP was prepared by mixing equal proportions of pure powder of metronidazole, ciprofloxacin, and clindamycin with distilled water to prepare the MTAP solution. (MC) the powder was added to this MTAP solution to get a thick, paste-like consistency.

Preparation of 25 mg/mL MTAP paste: 25 mg (metronidazole) + 25 mg (ciprofloxacin) + 25 mg (clindamycin) + 1 mL (distilled water) + 80 mg (MC) were mixed to prepare 25 mg/mL MTAP paste⁽⁷⁾.

Group Untreated: 80 mg of (MC) + 1 mL of distilled water to form MC paste.

Sample distribution: The specimens were randomly divided into three groups: Group (Nit) 25 mg/mL nitrofurantoin (n = 18), Group (MTAP) 25 mg/mL MTAP (n = 18), and Untreated group (no medicament) (n = 18).

Medicament Application For both treated groups, an amount of 0.15 ml of the medicament paste or sterile water was used⁽²⁴⁾, which was sufficient to cover the pulpal surface of each specimen using a disposable syringe with an angled flat tip endodontic needle, as illustrated in Figure 1, B-C. The specimens were then placed in a small conical sample container as shown in Figure 1, D-E, and a wet gauze was embedded in the cap's pocket of the container to keep the specimens wet without touching the specimens or the paste. The containers were stored in an incubator (Memmert, Schwabach, Germany) at approximately 100% relative humidity at 37°C for one week. After that, specimens were removed from the containers and rinsed with sterile distilled water until no visible paste was observed and were air-dried entirely.

Attenuated total reflectance Fourier transform infrared spectroscopy measurement (ATR-FTIR)

A 4100 ATR-FTIS device. (Bruker, Tensor 27, Germany) Fourier transform infrared spectrophotometer (FTIR) with a diamond attenuated total reflectance (ATR) accessory was used to obtain infrared spectra for analysis of dentin specimens, as shown in Figure 2. First, two spots on the specimens' pulpal surface were randomly chosen, then they were placed on a standard FTIR sample holder with a 3-mm diameter opening; after that, the spectra were measured.



Figure 2: ATR-FTIS device.

The ATR-FTIR spectra of air were recorded and then automatically subtracted by the Spectra Manager CFR software. Next, the spectrum underwent processing by dedicated Spectra manager CFR software through smoothing, baseline correction, and normalization to the amide I peak.

The mineral matrix ratio, which is the ratio of the phosphate ν_1 , ν_3 to the amide I peak, was used to assess

the effect of applying the nitrofurantoin paste and MTAP on apatite and collagen composition of the surface of radicular dentin. This can be achieved through the calculation of the relative contents of these inorganic and organic components. Phosphate/amide I ratios were measured, and their mean was calculated depending on the spectra recorded in each group; consequently, it was used for quantitative and statistical analysis⁽²⁴⁾.

The higher the (phosphate/amide I) ratio compared to the control (no medicament) dentin samples represented a higher collagen deproteinization process, while the lower the (phosphate/amide I) ratio compared to the control (no medicament) dentin samples indicated a higher dentin demineralization⁽²⁴⁾.

Eventually, polishing was performed using fine grades of composite polishing kit, then to obtain a smooth glossy mirror-like surface, 0.1 mm alumina suspension on a rotary felt disc was used.

Statistical analyses

All data were checked for normality using the ANOVA test, and the normality assumptions were satisfied. The differences in phosphate/amide I ratios among experimental groups were examined using one-way ANOVA followed by Tukey's HSD tests. A $p < 0.05$ level of statistical significance was applied for all analyses.

Results

One- The effect of Nit paste and MTAP on the phosphate / Amide I ratio on the (corona, middle, and apical) third of the radicular dentin is shown in table 1, which demonstrated that, compared to the untreated group, the (phosphate / Amide I ratio) was less affected with Nit group. At the same time, it was more affected in the MTAP group. There was a non-significant difference ($p > 0.05$) between the three groups in the (coronal and apical). Simultaneously, there was a significant difference between MTAP and the untreated group ($p < 0.05$) in the middle third, as shown in Table 2. Furthermore, the mean of the effects of intracanal medicaments in all three-thirds of the root was compared to the untreated group, and the differences were highly significant in all groups ($p < 0.001$), as shown in Table 2. In all groups, it is noted that the effect of the intracanal medicament was more potent in the coronal third, less in the middle third, and least in the apical third, as demonstrated in tables1. Furthermore,

the difference between the root thirds was highly significant ($p < 0.001$) in each group, except for the difference between root thirds (coronal and middle thirds) in the MTAP group, which showed a significant difference ($p < 0.05$), as shown in Table 3.

Attenuated total reflectance (ATR) spectra of Nit group, MTAP group, and the Untreated group are shown in Figure 3.

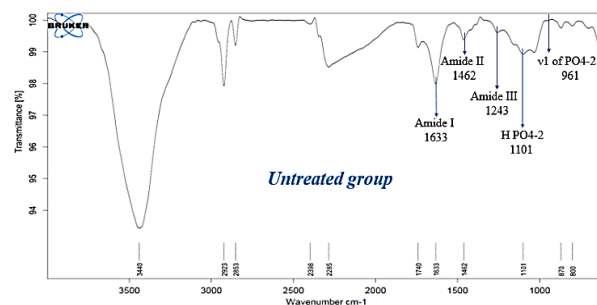


Figure 3: Attenuated total reflectance (ATR) spectra of Nit paste, MTAP, and untreated group.

Discussion

Intracanal medicaments may negatively affect radicular dentine's chemical, physical, and mechanical properties^(24,29). Therefore, to further understand the effects of antibiotic medicaments on radicular dentine, the changes in the chemical integrity of superficial radicular dentine after Nit paste and MTAP applications were explored in this study, using ATR-FTIR. In this study, we used one concentration of Nit paste as a testing group (25 mg/mL) with one concentration of MTAP (25 mg/mL). According to a recent study that used nitrofurantoin paste as an experimental intracanal medicament, this concentration has been shown to be highly effective against *Enterococcus Faecalis* in endodontic therapy, according to a recent study that used nitrofurantoin paste as an experimental intracanal medicament and compared it to MTAP with the same concentration⁽²³⁾. Besides, we evaluated the effect of the two medicament pastes on the three-thirds of the root (coronal, middle, and apical thirds) and compared the results with the untreated group. The results showed that in all the root thirds, the 25mg/ml MTAP group showed more reduction in the Phosphate/Amide I ratio than 25mg/ml of Nit paste group. Simultaneously, there was a non-significant reduction in phosphate/amide I ratio for both medicaments compared with the untreated group. After treatment, this reduction in phosphate/amide I ratio indicates a demineralization effect by these pastes⁽²⁴⁾.

Table 1: Mean of (phosphate / Amide I ratio) of the three groups (Nit paste, MTAP, and untreated group), in the (coronal, middle and apical) third of the radicular dentin.

Coronal								
Untreated			Nit			MTAP		
PHOSPHATE	AMIDE I	P/A ratio	PHOSPHATE	AMIDE I	P/A ratio	PHOSPHATE	AMIDE I	P/A ratio
96100	160034	0.60049739	96003.9	160655.2	0.59757729	95619.5	161769.3	0.59108557
96098	160037	0.60047364	96003.3	160654.5	0.59757616	95618.5	161769.1	0.59108025
96101	160032	0.60051114	96002.8	160655.8	0.59756821	95619.2	161769.6	0.59108262
96105	160039	0.60050987	96002.4	160653.9	0.59757279	95619.7	161768.9	0.59108827
96103	160029	0.60053490	96001.8	160653.4	0.59757091	95618.9	161769.2	0.59108223
96097	160030	0.60049365	96004.3	160654.6	0.59758201	95619.1	161768.8	0.59108493
Mean		0.60050343	Mean		0.59757456	Mean		0.59108398
Middle								
Untreated			Nit			MTAP		
PHOSPHATE	AMIDE I	P/A ratio	PHOSPHATE	AMIDE I	P/A ratio	PHOSPHATE	AMIDE I	P/A ratio
96113	160027	0.60060489	96012.3	160643.2	0.59767422	95617.3	161733.9	0.59120135
96121	160030	0.60064363	96013.9	160642.7	0.59768678	95616.2	161745.6	0.59115178
96127	160024	0.60070364	96011.5	160641.8	0.59767445	95618.9	161755.3	0.59113303
96116	160021	0.60064616	96012.3	160639.1	0.59768947	95617.3	161739.2	0.59118198
96134	160035	0.60070609	96011.6	160637.3	0.59769182	95616.1	161745.7	0.59115080
96129	160023	0.60071989	96014.8	160640.5	0.59769983	95618.9	161771.3	0.59107209
Mean		0.60067072	Mean		0.5976861	Mean		0.59114851
Apical								
Untreated			Nit			MTAP		
PHOSPHATE	AMIDE I	P/A ratio	PHOSPHATE	AMIDE I	P/A ratio	PHOSPHATE	AMIDE I	P/A ratio
96159	160003	0.60098248	96023.1	160621.2	0.59782332	95620.5	161721.4	0.59126683
96163	160011	0.60097743	96032.5	160620.4	0.59788482	95621.3	161723.2	0.59126519
96169	160019	0.60098488	96020.7	160631.5	0.59777005	95622.1	161733.2	0.59123359
96155	160013	0.60091992	96026.9	160622.6	0.59784177	95620.3	161722.1	0.59126393
96160	160010	0.60096244	96031.2	160621.5	0.59787263	95620.2	161732.5	0.59122439
96158	160014	0.60093492	96025.4	160630.1	0.59780452	95621.5	161741.7	0.59119881
Mean		0.60096035	Mean		0.59783285	Mean		0.59124212

Nit: Nitrofurantoin, MTAP: Modified triple antibiotic paste, P/A ratio: PHOSPHATE/ AMIDE I ratio, N=18

Table 2: The differences between the groups (Nit paste, MTAP, and untreated group) in each root thirds (coronal, middle, and apical thirds) and the mean of the three thirds in each group.

Site of FTIR	Group	p-value
Coronal	Nit - MTAP	0.729
	Untreated - Nit	0.194
	Untreated - MTAP	0.725
Middle	Nit - MTAP	0.202
	Untreated - Nit	0.302
	Untreated - MTAP	0.037
Apical	Nit - MTAP	0.469
	Untreated - Nit	0.892
	Untreated - MTAP	0.644
Mean of Coronal-Middle-Apical	Nit - MTAP	<0.001
	Untreated - Nit	<0.001
	Untreated - MTAP	<0.001

Table 3: The differences between root thirds (coronal, middle, and apical thirds) in each group (Nit paste, MTAP, and untreated group).

Group	Site of FTIR	p-value
Nitrofurantoin	Coronal-Middle	<0.001
	Coronal-Apical	<0.001
	Middle-Apical	<0.001
MTAP	Coronal-Middle	0.006
	Coronal-Apical	<0.001
	Middle-Apical	<0.001
Untreated	Coronal-Middle	<0.001
	Coronal-Apical	<0.001
	Middle-Apical	<0.001

This reduction in phosphate/amide I ratio with MTAP is attributed to the acidity of the antibiotic components of MTAP, being acidic (pH =4.9) compared to the Nit paste, which is neutral (pH =7.1), (30) moreover, any acidic agent causes dentin demineralization⁽³¹⁻³⁴⁾. A study showed a significant reduction in the Phosphate/Amide I ratio when using MTAP in two concentrations (1mg/mL and 1g/mL). They showed that the lower concentration (1mg/mL) with a pH of 6.6

caused less Phosphate/Amide I ratio reduction than the higher concentration (1 g/mL) with a pH of 4.0 that caused strong demineralization of radicular dentine; moreover, they confirmed by the SEM (Scanning electron microscope) images the presence of an appetite-depleted collagen layer in the root canals treated with 1 g/ml MTAP⁽³⁵⁾. To date, there are no reports on the effect of nitrofurantoin paste as an intracanal medicament on the phosphate/amide I ratio of radicular dentine. The reduction in phosphate/amide I ratio when using nitrofurantoin paste was less than that of MTAP, which can be attributed to the pH of Nitrofurantoin, which is neutral (7.1)⁽³⁰⁾. Furthermore, nitrofurantoin paste has a single antibiotic agent in contrast to the MTAP, which is composed of three antibiotics with 25mg of each agent in the mixture, which increases the total quantitative concentrations of those antibiotics, which will further affect the phosphate/amide I ratio of radicular dentine, since the pH and concentrations of the material used to affect the microhardness as well as the chemical structure of the radicular dentin^(32,33).

In all groups, the effect of the intracanal medicament was more potent in the coronal third, less in the middle third, and least in the apical third, with a highly significant difference among them. This can be apprehended by the tubular density, which varies from one area to another, on the root dentin surface, minimizing an orientation from cervical to apical dentin⁽³⁶⁾. Furthermore, there is an inverse correlation between dentine microhardness and tubular density, as Pashley et al. stated that the dentin microhardness decreases with the increase in tubular density⁽³⁷⁾. Consequently, there is an overall highly positive correlation between phosphate/amide I ratios and dentin's microhardness value⁽²⁴⁾. Moreover, this is due to the reduction in the amount of calcified matrix between tubules due to an increase in the tubule density, which is also associated with a decrease in the amount of inter-tubular dentin resulting in less phosphate/amide I ratios⁽³⁷⁾. As a result, the reduction in microhardness may increase the solubility and permeability of the root canal dentine, which minimizes the sealing ability and adhesion of dental materials to dentine, resulting in suppressing resistance to bacterial invasion and facilitating facilitates a bacterial invasion coronal leakage⁽³⁸⁾.

The limitation in the present study is that we studied the effects of Nitrofurantoin on the radicular dentine's chemical composition and was compared with an antibiotic-based medicament and did not involve other non-antibiotic intracanal medicaments like

chlorohexidine. Additionally, the current study investigated only one concentration for each paste. Therefore, further studies are needed to enhance the knowledge about other aspects of nitrofurantoin paste in endodontics.

References:

- Segura-Egea J, Gould K, Şen BH, Jonasson P, Cotti E, Mazzoni A, et al. Antibiotics in Endodontics: a review. *Int Endod J*. 2017;50(12):1169-84.
- Rôças I, Siqueira J. Root canal microbiota of teeth with chronic apical periodontitis. *J Clin Microbiol*. 2008;46(11):3599-606.
- Wang Q-Q, Zhang C-F, Chu C-H, Zhu X-F. Prevalence of *Enterococcus faecalis* in saliva and filled root canals of teeth associated with apical periodontitis. *Int J Oral Sci*. 2012;4(1):19-23.
- Siqueira Jr JF, Rôças IN. Polymerase chain reaction-based analysis of microorganisms associated with failed endodontic treatment. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 2004;97(1):85-94.
- Sedgley C, Nagel A, Dahlén G, Reit C, Molander A. Real-time quantitative polymerase chain reaction and culture analyses of *Enterococcus faecalis* in root canals. *J Endod*. 2006;32(3):173-7.
- Gomes BP, Pinheiro ET, Jacinto RC, Zaia AA, Ferraz CC, Souza-Filho FJ. Microbial analysis of canals of root-filled teeth with periapical lesions using polymerase chain reaction. *J Endod*. 2008;34(5):537-40.
- Karczewski A, Feitosa SA, Hamer EI, Pankajakshan D, Gregory RL, Spolnik KJ, et al. Clindamycin-modified triple antibiotic nanofibers: a stain-free antimicrobial intracanal drug delivery system. *J Endod*. 2018;44(1):155-62.
- Kirchhoff AL, Raldi DP, Salles AC, Cunha RS, Mello I. Tooth discoloration and internal bleaching after the use of triple antibiotic paste. *Int Endod J*. 2015;48(12):1181-7.
- Jagdale S, Bhargava K, Bhosale S, Kumar T, Chawla M, Jagtap P. Comparative evaluation of coronal discoloration induced by two triple antibiotic revascularization protocols when used at varying depths of temporary sealing material at the end of varying time periods. *J Conserv Dent*. 2018;21(4):388-393.
- Algarni AA, Yassen GH, Gregory RL. Inhibitory effect of gels loaded with a low concentration of antibiotics against biofilm formation by *Enterococcus faecalis* and *Porphyromonas gingivalis*. *J Oral Sci*. 2015;57(3):213-8.
- Ravi K. Antimicrobial efficacy of various intracanal medicaments against *Enterococcus faecalis*. *Journal of Pharmaceutical Sciences and Research*. 2017;9(10):1861-3.
- Ghabraei S, Marvi M, Bolhari B, Bagheri P. Minimum intracanal dressing time of triple antibiotic paste to eliminate *Enterococcus faecalis* (ATCC 29212) and determination of minimum inhibitory concentration and minimum bactericidal concentration: an ex vivo study. *J Dent (Tehran)*. 2018;15(1):1-9.
- Sabarathinam J, Muralidharan N. Antimicrobial efficacy of four different intracanal medicaments on contaminated extracted teeth: In vitro study. *Drug Invent Today*. 2018;10(2): 3026- 29.
- Slots J. Selection of antimicrobial agents in periodontal therapy. *J Periodontal Res*. 2002;37(5):389-98.
- Huang GT-J. A paradigm shift in endodontic management of immature teeth: conservation of stem cells for regeneration. *J Dent*. 2008;36(6):379-86.
- Cunha BA. Nitrofurantoin: an update. *Obstet Gynecol Surv*. 1989;44(5):399-406.
- Karam MRA, Habibi M, Bouzari S. Urinary tract infection: Pathogenicity, antibiotic resistance and development of effective vaccines against Uropathogenic *Escherichia coli*. *Mol Immunol*. 2019;108(1):56-67.
- Munoz-Davila MJ. Role of old antibiotics in the era of antibiotic resistance. Highlighted Nitrofurantoin for the treatment of lower urinary tract infections. *Antibiotics (Basel)*. 2014;3(1):39-48.
- Gardiner BJ, Stewardson AJ, Abbott IJ, Peleg AY. Nitrofurantoin and fosfomycin for resistant urinary tract infections: old drugs for emerging problems. *Aust Prescr*. 2019;42(1):14-19.
- Abdulla FE, Abdulla EM. Antibiotic options for *Enterococcus faecalis* infection. *Pak J Med Sci*. 2006;22(3):286-90.
- Rahbar M, Hajia M, Farzanehkah M. (2007). Activity of Nitrofurantoin Against Urinary Tract Infection (UTI) Isolates of Vancomycin-Resistant Enterococci (VRE): A Three-Year Survey in an Iranian Hospital. *Iran J Pathol*. 2007;2(4):171-4.
- Toner L, Papa N, Aliyu SH, Dev H, Lawrentschuk N, Al-Hayek S. Vancomycin resistant enterococci in urine cultures: Antibiotic susceptibility trends over a decade at a tertiary hospital in the United Kingdom. *Investig Clin Urol*. 2016;57(2):129-34.
- Alrahman MSA, Faraj BM, Dizaye KF. Assessment of Nitrofurantoin as an Experimental Intracanal Medicament in Endodontics. *Biomed Res Int*. 2020; 2020:2128473.
- Yassen GH, Eckert GJ, Platt JA. Effect of intracanal medicaments used in endodontic regeneration procedures on microhardness and chemical structure of dentin. *Restor Dent Endod*. 2015;40(2):104-12.
- Eliades G. Clinical relevance of the formulation and testing of dentine bonding systems. *J Dent*. 1994;22(2):73-81.
- Prather BT, Ehrlich Y, Spolnik K, Platt JA, Yassen GH. Effects of two combinations of triple antibiotic paste used in endodontic regeneration on root microhardness and chemical structure of radicular dentine. *J Oral Sci*. 2014;56(4):245-51.
- Nerness A, Ehrlich Y, Spolnik K, Platt J, Yassen G. Effect of triple antibiotic paste with or without ethylenediaminetetraacetic acid on surface loss

- and surface roughness of radicular dentine. *Odontology*. 2016;104(2):170-5.
28. Alyas SM, Fischer BI, Ehrlich Y, Spolnik K, Gregory RL, Yassen GH. Direct and indirect antibacterial effects of various concentrations of triple antibiotic pastes loaded in a methylcellulose system. *J Oral Sci*. 2016;58(4):575-82.
 29. Elgendy AA, Nagy MM. The effect of different intracanal medications on fracture resistance of root canal dentin. *Tanta Dent J*. 2015;12(3):163-7.
 30. Mewan A, Alrahman, Bestoon Muhammed Faraj, Dizaye KF. Evaluation of the biocompatibility of Nitrofurantoin as an experimental intracanal medicament in endodontics therapy. *Int J Pharm Res*. 2020;12(03):259-74.
 31. Rosihan A, Widodo W, Bayu IS, Eko S. Effect pH on demineralization dental erosion. *Int J Chem. Eng*. 2015;6(2):138-41.
 32. Zhang K, Kim YK, Cadenaro M, Bryan TE, Sidow SJ, Loushine RJ, et al. Effects of different exposure times and concentrations of sodium hypochlorite/ethylenediaminetetraacetic acid on the structural integrity of mineralized dentin. *J Endod*. 2010;36(1):105-9.
 33. Kara Tuncer A, Tuncer S, Siso SH. Effect of QMix irrigant on the microhardness of root canal dentine. *Australian Dental Journal*. 2015;60(2):163-8.
 34. Parhizkar A, Nojehdehian H, Asgary S. Triple antibiotic paste: momentous roles and applications in endodontics: a review. *Restor Dent Endod*. 2018;43(3): e28.
 35. Prather BT, Ehrlich Y, Spolnik K, Platt JA, Yassen GH. Effects of two combinations of triple antibiotic paste used in endodontic regeneration on root microhardness and chemical structure of radicular dentine. *J Oral Sci*. 2014;56(4):245-51.
 36. Komabayashi T, Nonomura G, Watanabe LG, Marshall Jr GW, Marshall SJ. Dentin tubule numerical density variations below the CEJ. *J Dent*. 2008;36(11):953-8.
 37. Pashley D, Okabe A, Parham P. The relationship between dentin microhardness and tubule density. *Endod Dent Traumatol*. 1985;1(5):176-9.
 38. Nikhil V, Jaiswal S, Bansal P, Arora R, Raj S, Malhotra P. Effect of phytic acid, ethylenediaminetetraacetic acid, and chitosan solutions on microhardness of the human radicular dentin. *J Conserv Dent* 2016(2):179-83.