



## THE EFFICACY OF CHLORHEXIDINE AS AN INTRACANAL MEDICATION IN ROOT CANAL TREATMENT: A LITERATURE REVIEW

Khalid Sultan AlFarm<sup>1</sup>, Ali Muidh Mashub AlQahtani<sup>1</sup>, Abdulrahman Saleh AlAloula<sup>1</sup>, Aseel Raafat AlHakeem<sup>1</sup>, Mohammed Maqbol Alshamrani<sup>1</sup>, Abdulaziz Mohammed AlQahtani<sup>1</sup>, Shahzeb Hasan Ansari<sup>2\*</sup>

1. *Dental Intern, College of Dentistry, Riyadh Elm University, Riyadh 11681, Saudi Arabia.*
2. *Lecturer, Department of Preventive Dentistry, College of Dentistry, Riyadh Elm University, Riyadh 11681, Saudi Arabia.*

### ARTICLE INFO

#### Received:

03 Jul 2020

#### Received in revised form:

18 Oct 2020

#### Accepted:

23 Oct 2020

#### Available online:

28 Oct 2020

**Keywords:** Chlorhexidine, Intracanal medication, Root canal treatment.

### ABSTRACT

One of the main goals of intracanal treatments is eliminating bacteria from the root canal with increased success. Due to its bactericidal characteristics, Chlorhexidine has been traditionally recommended as a root canal medication. Other intracanal medications used alongside Chlorhexidine, such as sodium hydroxide, enhance pulpal soft tissue dissolution by using sodium hypochlorite. However, sodium hydroxide is also known to be ineffective against *Enterococcus faecalis* and its tubular infection. *Enterococcus faecalis* is an organism that is commonly associated with failure in root canal therapy during its isolation. On the other hand, besides some drawbacks, Chlorhexidine has been reported to kill significant levels of *Enterococcus faecalis* in the root canal. Consequently, the advantages and disadvantages of Chlorhexidine in root canal treatment put its efficacy into question. Employing a systematic review, in this paper, we respond to the PICO question, "What is the efficacy of chlorhexidine as an intracanal treatment in root canal medication?" The findings of the review have implications that Chlorhexidine is an effective root canal treatment. The study concluded that intracanal medication is an efficient form of therapy in intracanal medication with Chlorhexidine as a potent root canal treatment agent.

Copyright © 2013 - All Rights Reserved - Pharmacophore

**To Cite This Article:** Khalid Sultan AlFarm, Ali Muidh Mashub AlQahtani, Abdulrahman Saleh AlAloula, Aseel Raafat AlHakeem, Mohammed Maqbol Alshamrani, Abdulaziz Mohammed AlQahtani, Shahzeb Hasan Ansari, (2020), "The Efficacy of Chlorhexidine as an Intracanal Medication in Root Canal Treatment: A Literature Review", *Pharmacophore*, 11(5), 156-162.

### Introduction

In modern dentistry, endodontic therapy has grown to become one of the most progressive aspects of treatment. Root canal treatment has been performed since 1928, and the effective rate has improved significantly over the years owing to numerous developments in the area. [1]

However, most procedures are not always successful, thereby creating the need for retreatment depending on the patient's treatment history. Over the years, there has been an increase in patients who need to have their teeth maintained. However, procedures that lead to a failed endodontic therapy lead to high bacterial diversity levels and the need for retreatment. These levels of bacteria in the root canal play a crucial role in the growth of periapical lesions. In unsuccessful surgical procedures and endodontic therapy, tooth extraction or apical therapy should be considered if endodontic retreatment is not an option. Retreatment options are analyzed based on patient cooperation, clinical symptoms, and radiographic signs and symptoms. [2, 3]

Chlorhexidine is a synthetic cationic bis-guanide made up of two symmetric 4-chlorophenyl rings and two biguanide groups connected by a central hexamethylene chain.[4] Chlorhexidine is a positively charged hydrophobic and lipophilic molecule that interacts with phospholipids and lipopolysaccharides on the bacterial cell membrane and reaches the cell via some form of the active or passive transport mechanism.[5] Its effectiveness is attributed to the association between the positive charge of the molecule and the negatively charged phosphate groups on the microbial cell walls [6], thus altering the cells' osmotic balance, which enhances the permeability of the cell wall that enables the molecule of Chlorhexidine to enter the bacteria. Chlorhexidine is stable as a salt. The most popular oral preparation, chlorhexidine gluconate, is water-soluble and simple to retain at physiological pH, disassociates, and releases the positively charged Chlorhexidine portion.[4] Low molecular

**Corresponding Author:** Shahzeb Hasan Ansari; Lecturer, Department of Preventive Dentistry, College of Dentistry, Riyadh Elm University, Riyadh 11681.

weight compounds, especially potassium and phosphorus, can leak at a low concentration (0.2 percent). On the other side, Chlorhexidine is bactericidal at a higher concentration (2 percent); cytoplasmic material precipitation results in cell death. [6] Regardless of its inability to dissolve tissue and its difficulty in removing the space, chlorhexidine irrigation prevents reinfection for very long periods. [7, 8] In a research experiment conducted on substantively, Chlorhexidine demonstrated a substantive duration of 7 days in bovine incisors. Due to these and other characteristics, experts have strongly considered using Chlorhexidine as an intracanal medication. [9]

When conducting an intracanal medication, essential considerations to take into place are possible leakage and its level in the root canal system. Singh *et al.* (2016) found high levels when calcium hydroxide is used alongside zinc oxide-eugenol sealer. [10] There is no literature specifying whether Chlorhexidine will enhance leakage in the root canal system or not. The main reasons for using intracanal medication are eliminating bacterial residing in the root canal, preventing bacterial proliferation, and using it as a physiochemical barrier. [11] Physiochemical barriers prevent reinfection in the root canal and supply nutrients to the remaining bacteria. [12] Hence, this study aims to review the efficacy of Chlorhexidine as an intracanal medication in endodontics.

## Methods

### Inclusion Criteria

In this study, we included most papers that assess intracanal medication using Chlorhexidine, using other intracanal medications such as calcium hydroxide and the papers that analyze the efficacy of using both. We included research papers and articles published during the last 5 years. Additionally, we included studies experimentally assessing placing Chlorhexidine as an intracanal medication, particularly in root canal treatment. Among the included studies, some were comparing our preferred treatment with other treatments.

### Exclusion Criteria

Firstly, we excluded studies that failed to address the effect of using Chlorhexidine as an intracanal medication. Secondly, we excluded research papers in languages other than English from the systematic review. Thirdly, we disregarded all non-peer-reviewed literature such as blogs and reports due to insufficient information to answer our PICO question. Thirdly, for a study to be included in our inclusion criteria, it focused on Chlorhexidine and not other intracanal medications such as calcium hydroxide. Finally, reviews, editorials, and letters were excluded from the systematic review.

### Search Strategy and Information Sources

We conducted an exhaustive search from all electronic databases focusing on social and biomedical sciences. We included databases such as CAB Abstract, MEDLINE, African Index Medicus (AIM), Cochrane Register of Controlled Trials (CENTRAL), ERIC, WEB OF SCIENCE, World Health Organization Library Information System (WHOLIS), Public Affairs Information Services (PAIS International), Latin American and Caribbean Health Sciences Literature (LILACS). The keywords used in the study with their definitions are shown in Table 1.

**Table 1:** Keywords used in the Study.

Keywords	Definition
<b>Chlorhexidine</b>	According to the National Cancer Institute (2020), Chlorhexidine is a biguanide compound commonly in tropical antibacterial activity as an antiseptic agent. [13]
<b>Intracanal Medication</b>	According to Kumar <i>et al.</i> (2019), intracanal medication is a form of treatment used to completely remove bacteria and its byproducts, pulpal remains of infected root canals, and sealing disinfected root canals completely. [14]
<b>Root Canal Treatment</b>	Root canal treatment is a procedure applied among patients with dental infections to relieve pain and save their teeth. [3]
<b>Endodontic Therapy</b>	Also known as root canal therapy, endodontic therapy is a dental procedure used to treat infections and prevent future infections in the tooth. [3]
<b>Bacterial Growth</b>	Also known as bacterial growth, bacterial proliferation increases bacterial growth in a given place. [1]

All titles and abstracts identified through the electronic search we independently screened. Also, we retrieved and screened the full texts of all articles and studies that met our inclusion criteria. In cases where we disagreed on the paper's standard, we consulted with other authors to establish the particular paper's relevance.

### Data Extraction

We applied data extraction forms specifically designed for this research study to extract the relevant data for intracanal medication using Chlorhexidine and its efficacy. In the extraction forms, we included the study's duration and the methods used, secondly, information on the interventions and comparisons, thirdly, the given outcomes, fourthly, the publication type

and comments, and finally, the types of samples included in the studies. The extracted data were recorded and analyzed to come up with a discussion of the results.

### Critical Appraisal

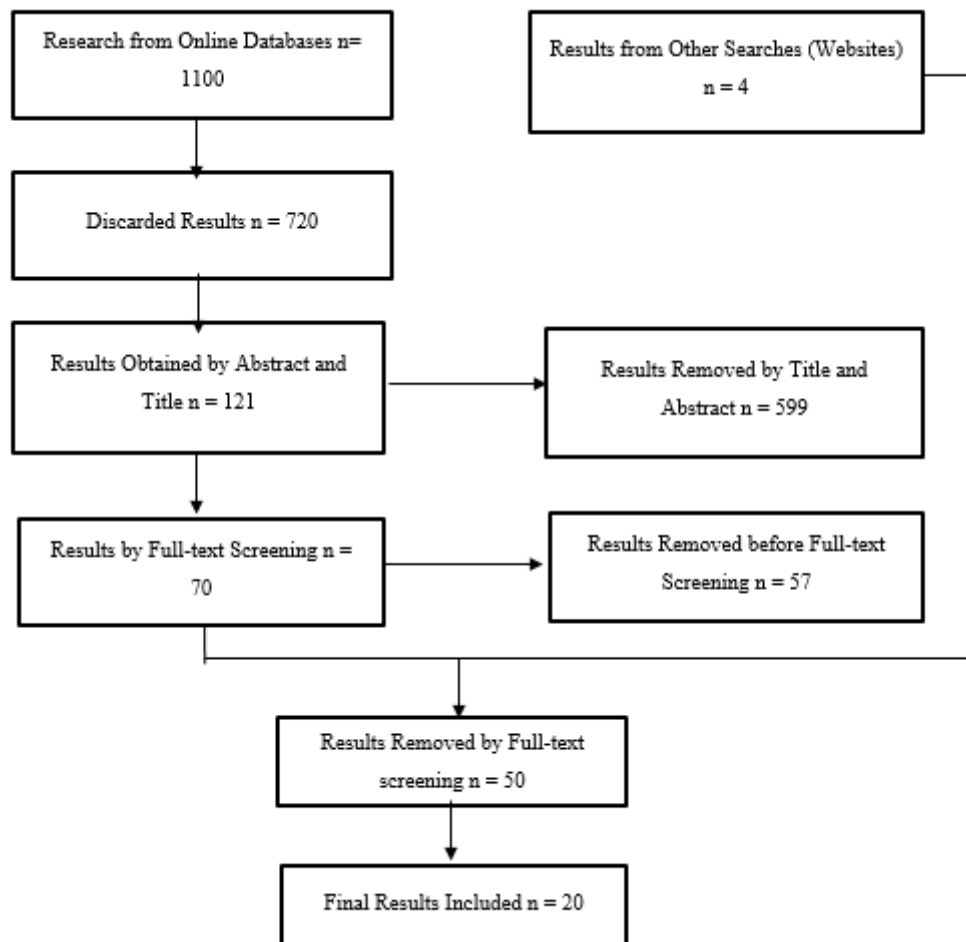
Intracanal medication using Chlorhexidine resulted in promising results where all sample teeth showed progress in eliminating bacteria and the infection. The treatment using Chlorhexidine was compared to other medications such as calcium hydroxide, plant extracts, and antibiotics.

### Data Synthesis

We applied a narrative synthesis of the results to assess the studies we included in our research. We analyzed Chlorhexidine's effect on different roots and presented the results separately for all the roots. Moreover, we used studies with study designs, outcome measures, and consistent interventions.

### Results

During our electronic search on all the online databases, we discovered 1100 references, which we later reduced to 720 after the exclusion of duplicates. We later reduced the number to 121 records after conducting title and abstract screening. We identified an additional four more studies using other search methods. Out of the 127 sources we came up with, we disregarded 57 before the full-text screening. This was done because 49 were not in English, 5 were ongoing studies, and 3 were literature reviews. From among the remaining sources, 50 were excluded following full-text screening since they focused on intracanal medications using other treatments aside from Chlorhexidine. Finally, 20 studies qualified for inclusion in the systematic review. The remaining 20 studies were included in the final review since they focused on the effect of intracanal medication using Chlorhexidine (**Figure 1**).



**Figure 1:** PRISMA Diagram

One of the articles compared the effect of both sodium hydroxide and Chlorhexidine on *Enterococcus faecalis*. [15] The results from each of the two were compared, and Chlorhexidine emerged to be more effective at killing *Enterococcus*

faecalis in the root canals. The experiment involved bovine incisors extracted and placed in NaOCI after removing all soft tissues. Similar to previous research on two groups of patients showed better results among the patients treated with Chlorhexidine than calcium hydroxide, which also contains antimicrobial and physicochemical properties useful for killing bacteria. [16, 17] In an experimental study, the root's apical part was removed, and the internal diameter was prepared into a round bur. The roots were, then, cut into small segments and the crowns removed. To extract the smear layer, the dentin segments were put in an ultrasonic bath and sterilized before being placed in a soy broth containing *Enterococcus faecalis* and being incubated to facilitate the infection of the tubules. [15]

While Vasudeva et al. (2017) pointed out that the *Enterococcus Faecalis* can penetrate and infect the dental tubules, the segments were removed five days later and rinsed in sterile water, dried, and divided into groups. Each segment's canal space in group A was filled with calcium hydroxide paste, and group B was filled with chlorhexidine paste. The two groups were incubated for an additional 1 week at 100% humidity, after which the segments were retrieved and the pastes removed using sterile water. In this stage, bacterial survival was tested by collecting dental shavings inside the canal. The collected dental shavings were weighed and suspended in a reduced transport fluid solution. A dilution of the resultant solution was created and incubated for a day, after which colony-forming units were counted. [18] When compared the number of colony-forming units between the two groups, the number of colonies in the inner and outer layers of the dental shavings was higher in group A as compared to group B. Group B dental shavings should have significantly progressed in the elimination of the *Enterococcus faecalis* as the bacterial proliferation was minimal. This shows that Chlorhexidine is efficient in eliminating *Enterococcus faecalis* and other bacteria residing in the root canal. [19] The summary of the studies included in the review are presented in **Table 2**.

**Table 2:** Summary of the studies included in the review.

Author	Date Published	Selection Criteria	Results
Borzini, L., Condò, R., De Dominicis, P., Casaglia, A., & Cerroni, L.	2016 [20]	Published within the Last 5 Years	Endodontic treatment is efficient in the elimination of microorganisms.
Singh, G., Gupta, I., Elshamy, F. M., Boreak, N., & Homeida, H. E.	2016 [10]	Published within the Last 5 Years	Eradication of microbial load leads to successful intracanal medication.
Zancan, R. F., Vivan, R. R., Milanda Lopes, M. R., Weckwerth, P. H., De Andrade, F. B., Ponce, J. B., & Duarte, M. A.	2016 [17]	Published within the Last 5 Years	Calcium hydroxide has antimicrobial properties similar to Chlorhexidine.
Chuagal N, Mallya S.M, Lin L.M	2017 [21]	Published within the Last 5 Years	Disease-free treatment outcome is the goal of the endodontic treatment.
Lakhani, A. A.	2017 [22]	Comparison between Treatments	Chlorhexidine is more effective than calcium hydroxide.
Vasudeva, A., Sinha, D. J., Tyagi, S. P., Singh, N. N., Garg, P., & Upadhyay, D.	2017 [18]	Comparison between Treatments	Chlorhexidine is more efficient than traditional treatments.
Ghabraei, G., Bolhari, B., & Afshar, M. S.	2018 [15]	Comparison between Treatments	Chlorhexidine is more effective than calcium hydroxide.
Lal, V., Parveen, N., & Sajjad, A.	2018 [19]	Focus on the Research Question	Chlorhexidine is an efficient intracanal medication.
Martinho, F., Gomes, C. C., Nascimento, G. G., Gomes, A. P., Leite, F. R.	2018 [11]	Experimental Study	Intracanal medication is effective in dental disinfection.
Miçoğulları Kurt, S., & Çalışkan, M. K.	2018 [16]	Comparison between Treatments	Chlorhexidine is more efficient than calcium hydroxide.
Savitha. A SriRekha. A Vijay. R, Ashwija, Champa. C, and Jaykumar. T	2019 [23]	Comparison between Treatments	2% CHX with chitosan group showed the highest microbial reduction against <i>E. faecalis</i> during retreatment of failed endodontic cases.
Kumar, A., Iftekhar, H., & Tamanna, S.	2019 [14]	Published within the Last 5 Years	Calcium hydroxide may be better than Chlorhexidine in root-canal medication.
Lamarque, G. C., Méndez, D. A., Gutierrez, E., Dionisio, E. J., Machado, M. A., Oliveira, T. M., Rios, D., & Cruvinel, T.	2019 [9]	Focus on the Research Question	Chlorhexidine is an efficient antimicrobial agent.
Ribeiro, M. B., Vasconcelos, R. A., Soares, A. D., Zaia, A. A., Ferraz, C. C., Almeida, J. F., & Gomes, B. P.	2019 [24]	Comparison between Treatments	Calcium hydroxide can kill some levels of bacteria.
Badr, M., & Elhafez, E.	2020 [25]	Published within the Last 5 Years	Traditional medications are preferred.
Alsabawi N. A	2020 [26]	Comparative Study	Chlorhexidine at 2% produces a better antimicrobial effect among other concentrations.

Nadhirah Faiz, Mebin George Mathew	2020 [27]	Comparative Study	The highest inhibitory zone against <i>E. faecalis</i> was seen with 2% CHX.
Ibrahim, A. M., Zakhary, S. Y., & Amin, S. A.	2020 [28]	Published within the Last 5 Years	Calcium hydroxide is as good as Chlorhexidine in root-canal medication.
Punathil S, Moyin S, Bhat SS, Hedge S, Pai A, James J.	2020 [29]	Comparative Study	Ca (OH) <sub>2</sub> + 2% CHX are effective against <i>E. faecalis</i> .
Phadnaik, g. M., Ghike, M., Tripathi, R., & Phadnaik, M. B.	2020 [30]	Focus on the Research Question	Chlorhexidine is an efficient alternative in dental therapy.

## Discussion

We reviewed most studies on Chlorhexidine's efficacy as a root canal medication in different types of root specimens. Our evaluation focused on Chlorhexidine and its effect by assessing the possibility of bacterial re-occurrence after the treatment. We identified 20 studies and articles that met our inclusion criteria. The evidence given by the studies suggests that the efficacy of Chlorhexidine is sufficient for application as an intracanal medication. However, our study results' interpretation needs caution due to the scarcity of quality evidence and the given interventions. None of the studies obtained presented sufficient evidence on the complete termination of bacteria in the root canal. In line with Borzini *et al.*'s [20] previous findings, shaping the root and cleansing it biomechanically with antimicrobial irrigants may eliminate the bacteria from the root canal dentin.

As a result, residue bacteria in the tubules can increase exponentially and negatively affect the root canal therapy's progress. *Enterococcus faecalis* has been identified in non-healing dental canals as one of the most commonly isolated root microorganisms. [22] The endodontic retreatment in combination with 2% CHX and chitosan group showed the highest microbial reduction against *E. faecalis*. [23] Interestingly, several studies have established that calcium hydroxide is, on many occasions, not effective in eliminating *enterococcus faecalis* existing in root canals. [28] Consequently, more and more studies are being performed to establish Chlorhexidine's efficiency as a cationic biguanide that can be used as an intracanal medication due to its vast antibacterial activity range. Chlorhexidine has shown its bactericidal characteristics at high concentrations and bacteriostatic characteristics at low concentrations.

Additionally, Chlorhexidine can get absorbed in the dentin tissues and be gradually released as time progresses. [30] Most of the studies we came across indicate that Chlorhexidine is better than most traditional medications when eradicating *Enterococcus faecalis* and other bacterial formations in root canals. Moreover, Chlorhexidine showed more efficiency than the traditional calcium hydroxide medication in eradicating *Candida Albicans* in the root canal.

## Strengths and Limitations

The present study is one of the few systematic reviews conducted on the efficacy of Chlorhexidine as an intracanal medication. We struggled to minimize biases by thoroughly searching through a wide range of online databases and conducting citation tracking. We then minimized the time lag by contacting different authors to carry out repository trial searches. However, this study's main limitation is language bias, which occurred due to neglected studies not published in English. Additionally, the limited number of studies included in this review raises concerns about the scarcity of high-quality evidence. The implication in this is that caution needs to be taken when interpreting and generalizing the results. Most of the excluded studies were neglected due to the lack of Chlorhexidine as an intracanal medication. As a result, the use of Chlorhexidine's restrictive disability as an intracanal medication would have substantially reduced the pool of potential studies applicable in the review. However, some studies, such as Ribeiro *et al.* [24] concluded that calcium hydroxide is a better intracanal medication. However, our study shows that the concentration of chlorhexidine was not sufficient to eradicate bacteria.

Additionally, the minimal time of exposure may have led to Chlorhexidine's ineffectiveness as an intracanal medication. Chlorhexidine's significant drawback was the inability to remove the smear layer and dissolve soft tissue. [25] However, most of the included studies demonstrated Chlorhexidine's efficiency as an intracanal medication due to its ability to terminate bacterial proliferation.

## Conclusion

The efficacy of Chlorhexidine as an intracanal medication in root-canal treatment may help to eliminate bacterial proliferation in root canals. *Enterococcus faecalis* is a major bacteria that can penetrate and grow throughout the dental tubules. Many studies have established that *Enterococcus faecalis* can resist calcium hydroxide medication and other forms of traditional treatment. However, most studies show that Chlorhexidine is an antimicrobial agent efficient in root-canal treatment due to its ability to eradicate *Enterococcus faecalis* and other bacteria in the dental tubules. This systematic review evaluates the existing literature on Chlorhexidine's efficacy as a root-canal medication and comes up with certain interventions that show Chlorhexidine's positive effects as an intracanal medication.

## References

1. Narayanan LL, Vaishnavi C. Endodontic microbiology. *J Conserv Dent.* 2010;13(4):233–9.
2. DMD AC By Allen Ali Nasseh, DDS, MMSc, James Bahcall, DMD, MS, Anne L Koch. What is endodontic success? How successful is endodontic therapy? | Compendium [Internet]. [cited 2020 Nov 5].
3. Root Canal Treatment [Internet]. American Association of Endodontists. [cited 2019 Jul 25]. Available from: <https://www.aae.org/patients/root-canal-treatment/>
4. Greenstein G, Berman C, Jaffin R. Chlorhexidine. An adjunct to periodontal therapy. *J Periodontol.* 1986;57(6):370–7.
5. Athanassiadis B, Abbott PV, Walsh LJ. The use of calcium hydroxide, antibiotics, and biocides as antimicrobial medicaments in endodontics. *Aust Dent J.* 2007;52(1 Suppl): S64–82.
6. Gomes BP, Souza SF, Ferraz CC, Teixeira FB, Zaia AA, Valdrighi L, Souza-Filho FJ. Effectiveness of 2% chlorhexidine gel and calcium hydroxide against *Enterococcus faecalis* in bovine root dentine in vitro. *Int Endod J.* 2003;36(4):267–75.
7. Mohammadi Z, Abbott PV. The properties and applications of chlorhexidine in endodontics. *Int Endod J.* 2009;42(4):288–302.
8. Mohammadi Z. Chlorhexidine gluconate, its properties, and applications in endodontics. *Iran Endod J.* 2008;2(4):113–25.
9. Lamarque GC, Méndez DA, Gutierrez E, Dionisio EJ, Machado MA, Oliveira TM, Rios D, Cruvinel T. Could chlorhexidine be an adequate positive control for antimicrobial photodynamic therapy in- in vitro studies? *Photodiagnosis Photodyn Ther.* 2019 Mar;25:58–62.
10. Singh G, Gupta I, Elshamy FMM, Boreak N, Homeida HE. In vitro comparison of antibacterial properties of bioceramic-based sealer, resin-based sealer and zinc oxide eugenol based sealer and two mineral trioxide aggregates. *Eur J Dent.* 2016 Sep;10(3):366–9.
11. Martinho FC, Gomes CC, Nascimento GG, Gomes APM, Leite FRM. Clinical comparison of the effectiveness of 7- and 14-day intracanal medications in root canal disinfection and inflammatory cytokines. *Clin Oral Investig.* 2018;22(1):523–30.
12. Hamidi MR, Mahmoudi E, Moghadamnia AA, Zahedpasha S. Effect of Calcium Hydroxide and Chlorhexidine Medicaments on the Apical Seal. *Iran Endod J.* 2012;7(1):15–9.
13. NCI Drug Dictionary [Internet]. National Cancer Institute. 2011 [cited 2020 Nov 5]. Available from: <https://www.cancer.gov/publications/dictionaries/cancer-drug>
14. Kumar A, Tamanna S, Iftekhar H. Intracanal medicaments—their use in modern endodontics: a narrative review. *Journal of Oral Research and Review.* 2019;11(2):94.
15. Ghabraei S, Bolhari B, Sabbagh MM, Afshar MS. Comparison of Antimicrobial Effects of Triple Antibiotic Paste and Calcium Hydroxide Mixed with 2% Chlorhexidine as Intracanal Medicaments Against *Enterococcus faecalis* Biofilm. *J Dent (Tehran).* 2018;15(3):151–60.
16. Miçoğulları Kurt S, Çalışkan MK. Efficacy of chlorhexidine as a final irrigant in one-visit root canal treatment: a prospective comparative study. *Int Endod J.* 2018;51(10):1069–76.
17. Zancan RF, Vivan RR, Lopes MR, Weckwerth PH, de Andrade FB, Ponce JB, Duarte MA. Antimicrobial Activity and Physicochemical Properties of Calcium Hydroxide Pastes Used as Intracanal Medication. *J Endod.* 2016;42(12):1822–8.
18. Vasudeva A, Sinha DJ, Tyagi SP, Singh NN, Garg P, Upadhyay D. Disinfection of dentinal tubules with 2% Chlorhexidine gel, Calcium hydroxide and herbal intracanal medicaments against *Enterococcus faecalis*: An in-vitro study. *Singapore Dent J.* 2017;38:39–44.
19. Lal V, Parveen N, Sajjad A. The Effectiveness of 2% Chlorhexidine Gel in Reducing Intracanal Bacterial Count. *Med Clin Res.* 2018;3(2):1–3.
20. Borzini L, Condò R, De Dominicis P, Casaglia A, Cerroni L. Root Canal Irrigation: Chemical Agents and Plant Extracts Against *Enterococcus faecalis*. *Open Dent J.* 2016;10:692–703.
21. Chugal N, Mallya SM, Kahler B, Lin LM. Endodontic Treatment Outcomes. *Dent Clin North Am.* 2017;61(1):59–80.
22. Lakhani AA, Sekhar KS, Gupta P, Tejolatha B, Gupta A, Kashyap S, Desai V, Farista S. Efficacy of Triple Antibiotic Paste, Moxifloxacin, Calcium Hydroxide And 2% Chlorhexidine Gel In Elimination of *E. Faecalis*: An In-vitro Study. *J Clin Diagn Res.* 2017 Jan;11(1): ZC06–9.
23. Savitha A, SriRekha A, Vijay R, Ashwija, Champa C, Jaykumar T. An in vivo comparative evaluation of antimicrobial efficacy of chitosan, chlorhexidine gluconate gel and their combination as an intracanal medicament against *Enterococcus faecalis* in failed endodontic cases using real-time polymerase chain reaction (qPCR). *Saudi Dent J.* 2019;31(3):360–6.
24. Barbosa-Ribeiro M, Arruda-Vasconcelos R, de-Jesus-Soares A, Zaia AA, Ferraz CC, de Almeida JF, Gomes BP. Effectiveness of calcium hydroxide-based intracanal medication on infectious/inflammatory contents in teeth with post-treatment apical periodontitis. *Clin Oral Investig.* 2019 Jun;23(6):2759–66.
25. Badr MM, Elhafez E. Comparison of Antibacterial Effect and Smear Layer Removal of Herbal versus Traditional Irrigants-An in vitro Study. *Egyptian Dental Journal.* 2020;66:1863–1871.

26. Al-Sabawi NA. Physical, Chemical, and Antimicrobial Properties of Chlorhexidine Combine with Calcium Hydroxide as Intracanal Medicament. *Al-Rafidain Dental Journal*. 2020;13(3):388–95.
27. Faiz N, Mathew MG. Comparative Evaluation of Antimicrobial Efficacy of Chlorhexidine and Aloe Vera against *Enterococcus Faecalis*: An in Vitro Study. *Indian Journal of Public Health Research & Development*. 2020;11(4):214–218.
28. Ibrahim AM, Zakhary SY, Amin SAW. Calcium hydroxide intracanal medication effects on pain and flare-up: a systematic review and meta-analysis. *Restor Dent Endod*. 2020 Aug;45(3):e26.
29. Punathil S, Moyin S, Bhat SS, Hedge S, Pai A, James J. Comparison of antibacterial effect of calcium hydroxide combined with chlorhexidine and povidone-iodine against *Enterococcus faecalis* in dentinal tubules of human incisors: An in vitro comparative study. *Journal of Pharmacy And Bioallied Sciences*. 2020;12(5):448.
30. Phadnaik GM, Ghike M, Tripathi R, Phadnaik MB. Chlorhexidine: An Adjunct Anti-microbial in Dental Therapy. *IOSR-JDM*. 19(07):37–42.