

Comparison of the effectiveness of the irrigation material ethyl diamine tetra-acetic acid (edta) 17% and benzalkonium chloride 1% on the smear layer in the 1/3 apical

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ABSTRACT

A smear layer can form on the root canal walls during mechanical root canal preparation. Bacteria that cause root canal infections can grow on the smear layer, which can also lead to micro-leakage in the top one-third of the root canal. Benzalkonium chloride (BAC) is a necessary and effective substance for root canal irrigation. In addition to being a cationic nitrogen agent having a quaternary ammonium group, BAC is a combination of alkyl dimethyl benzyl ammonium chloride, which has broad spectrum antibacterial activity. The goal of this study was to assess the efficacy of 1% BAK and 17% EDTA on the smear layer of the apical 1/3 and to measure the degree of cleanliness that BAK produced compared to 17% EDTA. The research was conducted utilising a scanning electron microscope (SEM) approach in a laboratory setting. Findings: SEM analysis of the sample smear layer yielded BAK 1% 0.667 and EDTA 17% 1.667 according to the Garberoglio and Becece scoring system. Kruskal-Wallis analysis of the data showed that the 17% EDTA group had an average range of 11.50, the 1% BAK group of 5.58, the 2.5% NaOCl group of 11.92, and the negative control group of 21.00. In conclusion, when comparing the groups with and without the negative control, the 17% EDTA group had a p value of 0.0069, the 1% BAK group a p value of 0.0019, and the 2.5% NaOCl group a p value of 0.0013, all determined by Mann-Whitney data analysis. Thus, it is evident that BAK 1% is more efficient than EDTA 17% in affecting the smear layer at the apical 1/3.

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INTRODUCTION

The success of endodontic treatment depends on many factors, including host factors, preparation, *microorganisms* and others. The main cause of root canal treatment failure is the persistence of

infection in the root canal which inhibits healing of the apical area (Ariani & Hadriyanto, 2013). RCT is a treatment with the principle of *triad endodontic*, that is *cleaning and shaping*, medication and disinfection, and root canal obturation (John, 2019).. Mechanical root canal preparation can result *smear layer* on the wall of the tooth root canal. *Smear layer* can be a substrate for bacteria that are the cause of root canal infections, and can cause *microleakage* in the 1/3 apical of the root canal (Nevi Yanti & Prasetya, 2017).

Irrigation is one way that can be done to reach hard-to-reach areas with *instrument*, especially in the area of the 1/3 apical of the root canal due to the large amount of accessory tissue (Haapasalo dkk., 2010). Instrumentation in root canals must be supported by the use of anti-microbial irrigation fluids. Irrigation is necessary to rinse away debris during instrumentation, also acts as a lubricant for the instrument, and removes *smear layer* on the instrumented dentin surface (Nurliza & Abidin, 2014).

Various dental root canal irrigation materials that are used commonly includes : *sodium hypochlorite* (NaOCl), *chlorhexidine gluconate* (CHX), *ethyl diamine tetra-acetic acid* (EDTA), *mixture tetracyclin isomer-acid-detergent* (MTAD), and *iodine potassium iodide* (Bariah & Suwelo, 2003). The irrigant that currently commonly used is NaOCl. NaOCl has the advantage of being able to dissolve tissue, being bactericidal, and also being relatively cheap (Murray, 2015). NaOCl has the weakness of not being able to dissolve inorganic tissue (Plotino dkk., 2016).

Another material that is generally used is EDTA. This material aims to eliminate *smear layer*. EDTA with a concentration of 17%, has the ability to act as *chelating agent* and can also clean inorganic substances (Deviyanti, 2018) (Prada dkk., 2019). EDTA 17% cannot remove organic substances and has no antibacterial activity (Mohammadi dkk., 2013). There are materials that can also be used as root canal irrigation materials, one of which is *benzalkonium chloride* (BAC) (Blaurock-Busch, 2016). BAC is a mixture of *alkyl dimethyl benzyl ammonium chloride* is also a cationic nitrogen containing group agent *quaternary ammonium* with broad antimicrobial activity (*broad spectrum*) (Merchel Piovesan Pereira & Tagkopoulos, 2019).

In the research of M.T Arias-Moliz et al in 2015 stated that adding a BAK concentration of 2% or higher to *AH Plus* exhibits antimicrobial and antibiofilm activity without affecting the properties specified in ANSI/ADA standards (Arias-Moliz dkk., 2015). The results also showed that BAC at a concentration of 0.1 - 0.2% had a fast reaction and high stability. Currently, BAC is widely used as a cosmetic product such as conditioner, and *lotion* body, eye drops and medications. BAK is also an active ingredient commonly used in disinfectants such as in residential, industrial, agricultural and clinical environments (Merchel Piovesan Pereira & Tagkopoulos, 2019).

BAC is a mixture of *alkyl benzyl dimethylammonium chloride* and is a cationic nitrogen containing group agent *quaternary ammonium* with broad antimicrobial activity (*broad spectrum*). BAC-based cavity cleaners are easy to obtain and are classified as safe for the human body.¹⁴ BAC can clean microorganisms by damaging cell membranes, the standard production for BAC production is 50% and 80% in liquid preparations, BAC is very effective if used in small doses, BAC has a fast reaction, and also high stability at a concentration of 0.1% - 0.2%. The application for adding domestic BAC has been widely used as a cosmetic product such as conditioner, *lotion* body as well as eye drops and medications. BAC is also included as an active ingredient commonly used in disinfectants in several environments such as residential, industrial, agricultural and clinical environments (Merchel Piovesan Pereira & Tagkopoulos, 2019).

BAC consists of a mixture of *quaternary ammonium compounds*, *benzyl-C12-16-alkyldimethyl chlorides* which has the formula $[C_6H_5CH_2N(CH_3)_2R]Cl$, R represents alkyl mixture. The BAC content must be no less than 95% nor more than 104% *alkyldimethylammonium chloride* ($C_{22}H_{40}NCl$). Four main components of BAK with different alkyl chain lengths (R), namely BAC-C10, -C12, -C14, and -C16, were determined, and their respective retention times were 2.98, 3.25, 3.50, and 3.73 minutes. This amount is considered the total BAK. BAK with different alkyl chain lengths has

different levels of toxicity, protein binding, inhibition of histamine release, and antimicrobial activity (Kwon dkk., 2019) (Johnson, 2018).

SEM is a type of electron microscope that is capable of producing high resolution images of the surface of a sample. The working principle of the SEM tool is to utilize electron backscatter (*electron beam*) on the surface of the object and take images by detecting electrons appearing on the surface of the object (Setyaningsih & Septiano, 2019). Advances in the use of SEM make it possible to scan large areas and collect large amounts of data to obtain sample characteristics, including counting objects and collecting statistics on the objects, one of which is obtaining morphological images of size to determine size distribution (Mohammed & Abdullah, 2018). SEM testing makes it possible to obtain morphological and concentration images of the mixture of materials (Setyaningsih & Septiano, 2019) (Septiano dkk., 2021).

Extensive research has been conducted on various irrigation materials and their effectiveness in removing the smear layer. However, there may be limited direct comparisons between EDTA and benzalkonium chloride, especially concerning their efficacy in the apical third. Previous studies might have focused on different concentrations, application techniques, or other variables, making direct comparisons necessary to draw definitive conclusions.

RESEARCH METHOD

By utilising scanning electron microscopy (SEM) to detect a smear layer in the upper one-third of the root canal, this study builds on previous work that used conventional irrigation methods to examine the same phenomenon. For the purpose of observation and data assessment, all of the research data is entered into a table. After then, non-parametric techniques are used to handle and analyse the data. Schwarz-Wallis pair When there are more than two groups to compare, we use MegaStat version 10.4 to find statistical differences in ranks on categorical scale independent variables. Then, we use Mann-Whitney to see if the differences were significant.

Researchers would collect quantitative data on the thickness of the smear layer in micrometers (µm) or other suitable units from multiple locations within the apical 1/3 of each root canal. Data collection should be blinded, meaning the researchers assessing the smear layer should be unaware of the treatment group assigned to each sample to minimize bias. Statistical analysis would be performed on the collected data using appropriate methods. This analysis would determine if there are significant differences in the effectiveness of EDTA and benzalkonium chloride in removing the smear layer. Any potential confounding factors, such as tooth morphology or operator variability, would also be accounted for in the analysis.

RESULTS AND DISCUSSIONS

Based on smear *layer* observations sample using *scanning electron microscope* (SEM), the results obtained are as in Table 1, the Garberoglio and Becce scoring systems are divided *smear layer* as follows.

1. 0: No smear layer is formed in the apical 1/3 of the tooth root canal.
2. 1: There is debris on the apical 1/3 of the tooth root canal.
3. 2: There is *smear layer* thin and debris in the apical 1/3 of the tooth root canal.
4. There is *smear layer* thick in the apical 1/3 of the tooth root canal.

Table 1. Irrigation results based on garberoglio and becce (1994) scoring

Repetition	EDTA 17%	BAC 1%	NaOCl 2,5%	Negative Control
1	0	0	1	3
2	1	0	2	3
3	2	0	2	3
4	2	1	2	3
5	2	1	2	3
6	3	2	2	3

Repetition	EDTA 17%	BAC 1%	NaOCl 2,5%	Negative Control
Rates	1,667	0,667	1,834	3,000

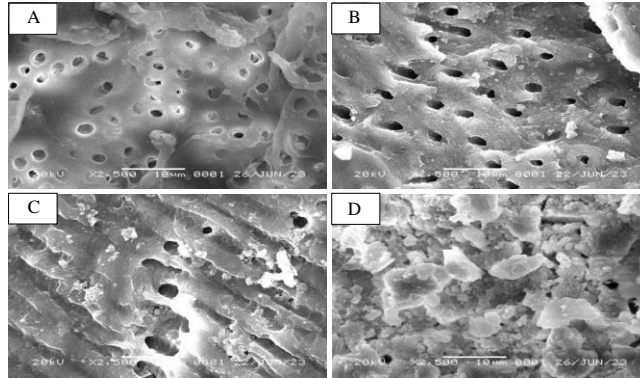


Figure 1. Irrigation Results in the first repetition. (A) EDTA 17%, (B) BAK 1%, (C) NaOCl 2.5%, and (D) negative control

In the first iteration, there was debris in the apical 1/3 of the root canal of the premolar tooth that was irrigated using 17% EDTA and 2.5% NaOCl. There is no *smear layer* formation at the 1/3 apical root canal of premolar teeth irrigated using BAC 1%. In the apical 1/3 of the root canal of untreated premolar teeth, there is *smear layer* thick enough to cover the dentinal tubules. The irrigation results in the first repetition can be seen in Figure 1.

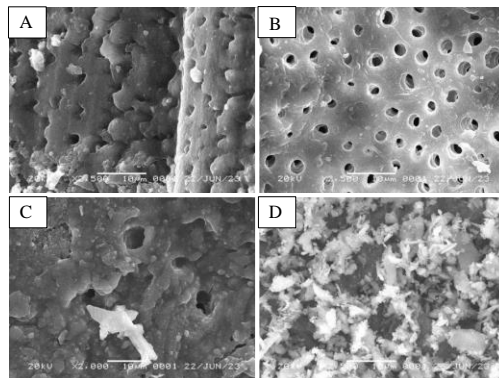


Figure 2. Irrigation Results in the second repetition. (A) EDTA 17%, (B) BAC 1%, (C) NaOCl 2.5%, and (D) negative control

In the second repetition, there was still debris in the apical 1/3 of the root canal of the premolar tooth which was irrigated using 17% EDTA and 2.5% NaOCl and the number of visible dentinal tubules was also decreasing. There is no *smear layer* formation on the apical 1/3 of the root canal of premolar teeth irrigated using 1% BAK. In the apical 1/3 of the untreated premolar root canal, there is a lot of debris and thick *smear layer* covering of the dentinal tubules. The irrigation results in the first repetition can be seen in Figure 2

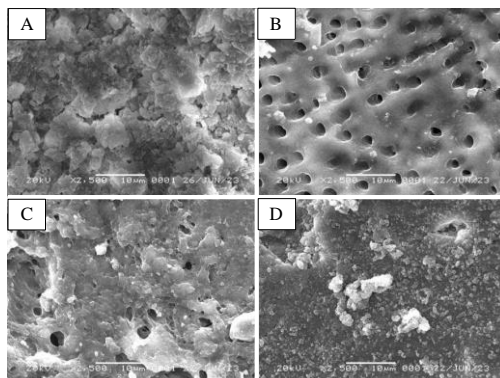


Figure 3. Irrigation Results in the third repetition. (A) EDTA 17%, (B) BAC 1%, (C) NaOCl 2.5%, and (D) negative control

In the third repetition, there was still debris in the apical 1/3 of the root canal of the premolar tooth which was irrigated using 17% EDTA and 2.5% NaOCl and fewer dentinal tubules were visible. There is no *smear layer* formation on the apical 1/3 of the root canal of premolar teeth irrigated using 1% BAK. In the apical 1/3 of the root canal of untreated premolar teeth, there is *smear layer* thick enough to cover the dentinal tubules. The irrigation results in the first repetition can be seen in Figure 3.

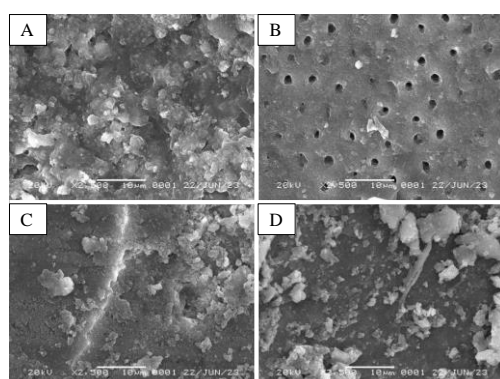


Figure 4. Irrigation Results in the fourth repetition. (A) EDTA 17%, (B) BAC 1%, (C) NaOCl 2.5%, and (D) negative control

In the fourth repetition, irrigation of the apical 1/3 of the premolar root canal using 17% EDTA and 2.5% NaOCl still gave the same results as the previous repetition, where there was still debris covering the dentinal tubules. There is debris in the 1/3 apical of the root canal of the premolar tooth which was irrigated using 1% BAK but the dentinal tubules can still be seen. In the 1/3 apical of the root canals of untreated premolars, there are *smear layer* thick enough to cover the dentinal tubules. The irrigation results in the first repetition can be seen in Figure 4.

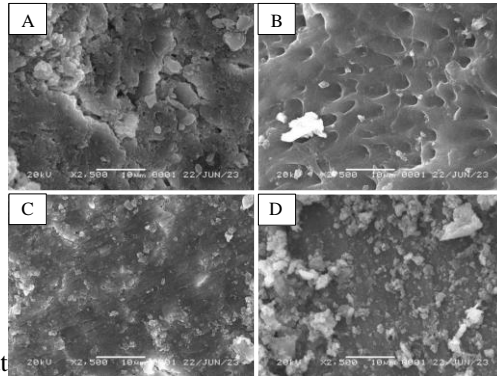


Figure 5. Irrigation results in the fifth repetition. (A) EDTA 17%, (B) BAC 1%, (C) NaOCl 2.5%, and (D) negative control

In the fifth repetition, irrigation of the 1/3 apical of the premolar root canal using 17% EDTA and 2.5% NaOCl still gave the same results as the previous repetition, where there was still debris covering the dentinal tubules. There is debris in the 1/3 apical of the root canal of the premolar tooth which was irrigated using 1% BAK but the dentinal tubules can still be seen. In the 1/3 apical of the root canals of untreated premolars, there are *smear layer* thick enough to cover the dentinal tubules. The irrigation results in the first repetition can be seen in Figure 5.

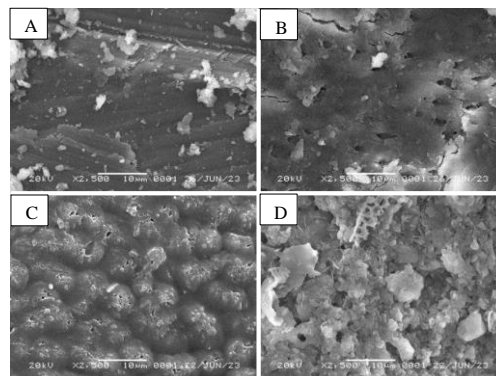


Figure 6. Irrigation results in the sixth repetition. (A) EDTA 17%, (B) BAC 1%, (C) NaOCl 2.5%, and (D) negative control

At the sixth repetition, the dentinal tubules could no longer be seen in the apical 1/3 of the root canals of premolars irrigated using 17% EDTA and which had not been treated due to the presence of a lot of debris and thick *smear layer* covering of the dentinal tubules. There is debris in the 1/3 apical of the root canal of the premolar tooth which was irrigated using 1% BAK but the dentinal tubules can still be seen the same as several previous repetitions. The irrigation results in the first repetition can be seen in Figure 6. Based on data analysis using *Kruskal-Wallis*, obtained a p value of 0.0012 or $p < 0.05$. The results of data analysis using *Kruskal-Wallis* can be seen in Table 2.

Table 2. Hypothesis test data analysis results (*Kruskal-Wallis*)

Group	Middle Value	N	Average Rank
EDTA 17%	2,00	6	11,50
BAK 1%	0,50	6	5,58
NaOCl 2,5%	2,00	6	11,92
K-	3,00	6	21,00

Total	2,00	24	-
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Based on data analysis using *Mann-Whitney*, obtained a p value of 0.0069 in the 17% EDTA group, 0.0019 in the 1% BAC group, and 0.0013 in the 2.5% NaOCl group compared with the negative control, and a p value of 0.0190 in the BAK group 1% compared to the 2.5% NaOCl group. The results of data analysis using *Mann-Whitney* can be seen in Table 3.

Table 3. Comparative test data analysis results (*Mann-Whitney*)

	EDTA 17%	BAK 1%	NaOCl 2,5%	K-
EDTA 17%				
BAK 1%	0,0943			
NaOCl 2,5%	0,8485	0,0190		
K-	0,0069	0,0019	0,0013	

Discussion

Analysing data to test a hypothesis The mean value using Kruskal-Wallis with a sample size of 24 was 2.00 in the group treated with 17% EDTA and 2.5% NaOCl, 0.50 in the group treated with 1% BAC, and 3.00 in the control group that did not receive any treatment. Data analysis yielded an average rangeThe 17% EDTA group had a Kruskal-Wallis of 11.50, the 1% BAC group of 5.58, the 2.5% NaOCl group of 11.92, and the negative control group of 21.00. According to these findings, we can accept H1 that BAK 1% affects the smear layer 1/3 apical to the root canal of premolars and is more effective than EDTA 17%; we can reject H0 that BAK 1% does not affect the smear layer 1/3 apical to the root canal of premolars and is less effective than EDTA 17% (Aguayo Bonniard dkk., 2016).

According to data-driven comparison testingUsing the Mann-Whitney U test, we found that EDTA 17%, BAC 1%, and NaOCl 2.5% were more effective than the negative control group in removing the smear layer from one-third of the premolars' root canals ($p = 0.0069, 0.0019, \text{ and } 0.0013$, respectively) (Deutschle dkk., 2006). A p value of 0.0190 indicates that 1% BAC is significantly more effective than 2.5% NaOCl in removing the smear layer from one-third of the root canals of premolars (Rasmussen dkk., 2014).

Additionally, the smear layer scoring findings observed using SEM corroborate this, with BAK exhibiting an average range of 1% and a lowest value of 5.58. A score of 0 was assigned to 1% BAK because, as observed using scanning electron microscopy (SEM), it caused structural disorganisation of the smear layer in three premolar root canals that had previously formed no smear layer. In the other two premolar root canals, 1% BAK was successful in dissolving the smear layer, leaving only the apical 1/3 of the canal with debris; thus, it received a score of 1. It takes a score of 2 to dissolve the smear layer in the last 1 premolar root canal using 1% BAK.A component of BAK, according to 2017 study by Yang Jiao: quaternary ammonium, which possesses antibacterial, antifungal, antiviral, and anti-matrix metalloprotein properties, can dissolve chemical molecules.¹⁹ The capacity of BAK to inhibit inorganic chemicals found in fresh water was demonstrated by study conducted by Hiroshi Takahashi in 2022.²⁰ In this study, the apical 1/3 of the root canals of the premolars were shown to have smear layer structural disorganisation, which was caused by BAK 1% dissolving both organic and inorganic molecules that comprise the smear layer (Jiao dkk., 2017) (Takahashi & Minami, 2022).

The average rank smear layer in premolars treated with 17% EDTA Since 17% EDTA could only create structural disorganisation of the smear layer on a single tooth, the resultant result was 11.50. Debris is still visible in the smear layer at the very top of one premolar tooth's root canal and in the very bottom of another premolar's root canal, on other teeth. Janhavi Jagzap found in 2017 that EDTA, being a 17% chelating agent, can only dissolve inorganic compounds. However, the smear layer contains both organic and inorganic compounds. As a result, the apical 1/3 of the root canals of the premolars in this study were less effectively structurally disorganised by EDTA 17%.

(Jagzap dkk., 2017).

The average rank smear layer in premolars that were treated with 2.5% NaOCl or served as a positive control Due to the persistence of debris and a thin layer of smear in the apical 1/3 of the root canal of the premolar tooth following the administration of 2.5% NaOCl, the result obtained was 11.92. The results of this study show that EDTA 17% is less effective in inducing structural disorganisation of the smear layer in the apical 1/3 of the root canals of the premolars compared to using 2.5% NaOCl as an irrigation solution. This is due to the fact that NaOCl can only dissolve organic compounds, alter the shape of dentin, and be erosive.

(Jagzap dkk., 2017).

The average rank smear layer thickness of the apical 1/3 of the root canal was 21.00 in premolars that were not treated or served as a negative control; this was due to the fact that these teeth did not receive any irrigation solution.

CONCLUSION

The smear layer in the apical 1/3 is affected by 1% benzalkonium chloride (BAK), which is more effective than 17% ethylene diamine tetraacetate acid (EDTA). The study may focus solely on comparing the effectiveness of EDTA and benzalkonium chloride, neglecting potential interactions with other irrigants, medicaments, or instrumentation techniques used in clinical practice.

Exploring the potential synergistic effects of combining EDTA and benzalkonium chloride with other irrigants, chelators, surfactants, or antimicrobial agents could enhance smear layer removal and antimicrobial activity while minimizing adverse effects.

References

- Aguayo Bonniard, A., Yeung, J. Y., Chan, C. C., & Birt, C. M. (2016). Ocular surface toxicity from glaucoma topical medications and associated preservatives such as benzalkonium chloride (BAK). *Expert Opinion on Drug Metabolism & Toxicology*, 12(11), 1279-1289. <https://doi.org/10.1080/17425255.2016.1209481>
- Ariani, N. G. A., & Hadriyanto, W. (2013). Perawatan Ulang Saluran Akar Insisivus Lateralis Kiri Maksila dengan Medikamen Kalsium Hidroksida-Chlorhexidine. *Majalah Kedokteran Gigi Indonesia*, 20(1), Article 1. <https://doi.org/10.22146/majkedgiind.8342>
- Arias-Moliz, M. T., Ruiz-Linares, M., Cassar, G., Ferrer-Luque, C. M., Baca, P., Ordinola-Zapata, R., & Camilleri, J. (2015). The effect of benzalkonium chloride additions to AH Plus sealer. Antimicrobial, physical and chemical properties. *Journal of Dentistry*, 43(7), 846-854. <https://doi.org/10.1016/j.jdent.2015.05.003>
- Bariah, B., & Suwelo, I. S. (2003). PERAWATAN PULPOTOMI MORTAL SATU KALI KUNJUNGAN PADA MOLAR SULUNG NON VITAL (Laporan Kasus). *Journal of Dentistry Indonesia*, 10(3), 699-704. <https://core.ac.uk/download/pdf/297925902.pdf>
- Blaurock-Busch, E. K. (2016). EDTA: Ethylene diamine tetra acetic acid-A review. *Occup. Med. Health Aff*, 4(2). https://microtraceminerals.com/fileadmin/uploads/pdf/en/EDTA_OccupHealth16.pdf
- Deuschle, T., Porkert, U., Reiter, R., Keck, T., & Riechelmann, H. (2006). In vitro genotoxicity and cytotoxicity of benzalkonium chloride. *Toxicology in Vitro*, 20(8), 1472-1477. <https://doi.org/10.1016/j.tiv.2006.07.006>
- Deviantyanti, S. (2018). POTENSI LARUTAN CHITOSAN 0,2% SEBAGAI ALTERNATIF BAHAN IRIGASI DALAM PERAWATAN SALURAN AKAR GIGI. *Jurnal Ilmiah Dan Teknologi Kedokteran Gigi*, 14(1), Article 1. <https://doi.org/10.32509/jitekgi.v14i1.642>
- Haapasalo, M., Shen, Y., Qian, W., & Gao, Y. (2010). Irrigation in Endodontics. *Dental Clinics*, 54(2), 291-312. <https://doi.org/10.1016/j.cden.2009.12.001>
- Jagzap, J. B., Patil, S. S., Gade, V. J., Chandhok, D. J., Upagade, M. A., & Thakur, D. A. (2017). Effectiveness of Three Different Irrigants - 17% Ethylenediaminetetraacetic Acid, Q-MIX, and Phytic Acid in Smear Layer Removal: A Comparative Scanning Electron Microscope Study. *Contemporary Clinical Dentistry*, 8(3), 459-463. https://doi.org/10.4103/ccd.ccd_524_17

- Jiao, Y., Niu, L., Ma, S., Li, J., Tay, F. R., & Chen, J. (2017). Quaternary ammonium-based biomedical materials: State-of-the-art, toxicological aspects and antimicrobial resistance. *Progress in Polymer Science*, 71, 53–90. <https://doi.org/10.1016/j.progpolymsci.2017.03.001>
- John, L. C. (2019). CRUCIAL RADIOGRAPHIC APPRAISAL OF ROOTS-A KEY TO ENDODONTIC SUCCESS. *JIDAK*, 1(2), 44–50. http://www.jidakochi.org/co_editor/upload/1602778437JIDA%20Kochi%202019%20Vol%201%20Iss%202%20Pages%2044-50.pdf
- Johnson, N. F. (2018). Pulmonary Toxicity of Benzalkonium Chloride. *Journal of Aerosol Medicine and Pulmonary Drug Delivery*, 31(1), 1–17. <https://doi.org/10.1089/jamp.2017.1390>
- Kwon, D., Lim, Y.-M., Kwon, J.-T., Shim, I., Kim, E., Lee, D.-H., Yoon, B.-I., Kim, P., & Kim, H.-M. (2019). Evaluation of pulmonary toxicity of benzalkonium chloride and triethylene glycol mixtures using in vitro and in vivo systems. *Environmental Toxicology*, 34(5), 561–572. <https://doi.org/10.1002/tox.22722>
- Merchel Piovesan Pereira, B., & Tagkopoulos, I. (2019). Benzalkonium Chlorides: Uses, Regulatory Status, and Microbial Resistance. *Applied and Environmental Microbiology*, 85(13), e00377-19. <https://doi.org/10.1128/AEM.00377-19>
- Mohammadi, Z., Shalavi, S., & Jafarzadeh, H. (2013). Ethylenediaminetetraacetic acid in endodontics. *European Journal of Dentistry*, 07(S 1), S135–S142. <https://doi.org/10.4103/1305-7456.119091>
- Mohammed, A., & Abdullah, A. (2018). Scanning electron microscopy (SEM): A review. *Proceedings of the 2018 International Conference on Hydraulics and Pneumatics – HERVEX, Băile Govora, Romania, 2018*, 7–9. <https://fluidas.ro/hervex/proceedings2018/77-85.pdf>
- Murray, P. (2015). *A Concise Guide to Endodontic Procedures*. Springer. <https://doi.org/10.1007/978-3-662-43730-8>
- Nevi Yanti, D., & Prasetya, W. (2017). The ability of root canal irrigant with ethanol extract of Lerak fruit (*Sapindus Rarak Dc*) in removing root canal smear layer (a sem study). *IOSR Journal of Dental and Medical Sciences*, 16, 24–30. <https://citeseerx.ist.psu.edu/document?repid=rep1&type=pdf&doi=a0f08c2bbb431c2314f83fcd8c8e6e60d85d5fd>
- Nurliza, C., & Abidin, T. (2014). Prinsip-Prinsip Dasar Preparasi Saluran Akar Secara Khemomekanis. *Dentika Dental Journal*, 18(2), 177–184. <https://talenta.usu.ac.id/dentika/article/download/2027/1459>
- Plotino, G., Cortese, T., Grande, N. M., Leonardi, D. P., Di Giorgio, G., Testarelli, L., & Gambarini, G. (2016). New Technologies to Improve Root Canal Disinfection. *Brazilian Dental Journal*, 27, 3–8. <https://doi.org/10.1590/0103-6440201600726>
- Prada, I., Micó-Muñoz, P., Giner-Lluesma, T., Micó-Martínez, P., Muwaquet-Rodríguez, S., & Albero-Montegudo, A. (2019). Update of the therapeutic planning of irrigation and intracanal medication in root canal treatment. A literature review. *Journal of clinical and experimental dentistry*, 11(2), e185. <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6383907/>
- Rasmussen, C. A., Kaufman, P. L., & Kiland, J. A. (2014). Benzalkonium Chloride and Glaucoma. *Journal of Ocular Pharmacology and Therapeutics*, 30(2–3), 163–169. <https://doi.org/10.1089/jop.2013.0174>
- Septiano, A. F., Sutanto, H., & Susilo. (2021). Synthesis and characterization of resin lead acetate composites and ability test of X-ray protection. *Journal of Physics: Conference Series*, 1918(2), 022003. <https://doi.org/10.1088/1742-6596/1918/2/022003>
- Setyaningsih, N. E., & Septiano, A. F. (2019). Optimasi kualitas citra scanning electron microscopy (sem) dengan metode contrast to noise ratio (CNR). *Prosiding Seminar Nasional IV Hasil Penelitian Pranata Laboratorium Pendidikan Indonesia, IV-ISSN*, 2548–1924.
- Takahashi, H. A., & Minami, M. (2022). Assessment of the influence of benzalkonium chloride addition on radiocarbon analysis of dissolved inorganic carbon. *Limnology and Oceanography: Methods*, 20(10), 605–617. <https://doi.org/10.1002/lom3.10508>