MedCrave
Step into the World of Research
Journal of Dental Health, Oral Disorders & Therapy

Short communication
Open Access
CrossMark

Cold plasma: a good *Enterococcus faecalis* inhibitor – an in vitro tooth root canal pilot study

Abstract

Tooth root canals with endodontic treatment can be reinfected because the continued presence of *Enterococcus faecalis* biofilm. Many techniques of has been proposed to eliminated this bacteria, however, these techniques no offer the fully elimination of the E. faecalis. In this pilot study cold plasma was effective in the fully inhibition of E. faecalis biofilm in infected tooth root canals.

Keywords: E. faecalis, disinfection, cold plasma

Volume 13 Issue 1 - 2022

Rufo Figueroa-Banda,[1] Luis Ponce-Soto,[2] Gustavo Obando-Pereda[2]
[1]Dentistry School of Universidad Catolica de Santa Maria, Peru
[2]Proteomics, pain and inflammation laboratory of Universidad Catolica de Santa Maria, Peru

Correspondence: Gustavo Obando Pereda, Proteomics, pain and inflammation laboratory of Universidad Catolica de Santa Maria, Arequipa, Peru, Email gobando@uscm.edu.pe

Received: December 12, 2021 | **Published:** January 21, 2022

Introduction

The main of endodontic treatment is to eliminate total microbial biofilm inside of the root canal system to prevent a reinfection.[1] Traditional and modern treatments as mechanical debridement, chemicals and physics irrigation employing sodium hypochlorite, chlorhexidine, laser irradiation and ultrasound, perform a good microbial removing but not a complete elimination thus being able to reinfect the tooth canal system.[2] Persistent endodontic infection are frequently cause by *Enterococcus faecalis* who has the capacity to live in dentinal tubules forming a mature biofilms capable to resist any antibiotic and resist the host immune defenses.[3] The elimination of *E. faecalis* from root canal systems can be make using many strategies as the use mainly calcium hydroxide (Ca[OH]2) as intracanal medication.[3,4] However, some studies showed than *E. faecalis* still in the root canals more than 10 days of treatment provoking a future reinfection of the root canal systems.[5] Atmospheric pressure plasma technologies have been used in various applications such disinfection, sterilization mainly.[6] In dental application is use in dental whitening a disinfection of dental materials.[7] In this manner, the main of the study is investigate the capacity of cold plasma in the inhibition of *E. faecalis* biofilm.

Materials and methods

Root canal samples

Thirty single rooted extracted human teeth were cleaned and stored in 0.1% thymol solution at 4°C before the procedures. The tooth were decoronated below the cementoenamel junction to obtain a standard length of 10 mm.[8] All tooth specimens were prepared with nickel-titanium hand files up to size #40 in a step-back manner. The canals were irrigated with 5.25% sodium hypochlorite (NaOCl), the smear layer was cleaned using 20% EDTA for 5 minutes and an ultrasonic bath of 5.25% NaOCl for 5 minutes.[8] After that, root apical foramens were sealed with composite resin. Finally, the root samples were sterilized in an autoclave for 15 minutes at 121°C at 1 atmosphere pressure.[9]

Bacterial culture and root canal infection

ATCC of *E. faecalis* (29212) was cultured in brain-heart infusion (BHI) broth under at 37°C in aerobic condition. The experimental bacterial concentration was adjusted to 1.5 x 108 colony-forming units (CFU)/mL as measured by spectrophotometer. 100 uL of *E. faecalis* inoculum were placed into the root canal and using a sterile dental wax the coronal portion was sealed. The samples were place in sterile 10 mL centrifuge tubes with 3 mL of BHI and were incubated for 21 days at aerobic conditions. After that, the samples were washed three times with sterile water until the cold plasma treatment.[9]

Cold plasma device

The cold plasma device was built as described by Pan.[9] Briefly, one Teflon tube, a 1-mΩ resistor, and an outer cooper foil surrounding the Teflon tube connected to a high-voltage (18 kv). Premixed argon (98%) and oxygen (2%) passed into the teflon tube at a flow rate of 5L/min. The plasma is generated inside the tube by the high-voltage electrodes.

Disinfection of the root canal

Twenty-five specimens were used to verify the effectiveness of cold plasma against the E. faecalis biofilm in root canal system. The tooth were divide in 5 groups of 5 teeth. Group 1 was used as negative control, groups 2 – 4 were treated by cold plasma for 2, 5 and 10 minutes, respectively (Figure N°1). Group 5 (positive group) were treated using NaOCl irrigation. After that, root canal was washed with sterile water and sterile paper points were inserted into the canal to retire the bacterial. The paper points were places into sterile tubes with BHI for 7 days in aerobic conditions at 37°C to observe microbial growth.[9]

Statistical analysis

Analysis was performed using Anova One-way post hoc Tukey, and a P value < 0.05 was considered significant.

Results

The results of this experiment (Figure N°2) showed that NaOCl canal treatment (positive group) do not have a statistical difference in compare with 2 minutes cold plasma treatment ($p = 0.622$). Cold plasma treatment at 5 and 10 minutes, showed the total inhibition of *E. faecalis* growth in the root system canals. Negative control showed a high *E. faecalis* presence in compare with the other experimental groups ($p < 0.0001$).

Figure 1 Photograph of cold plasma jet during in vitro tooth root canal.

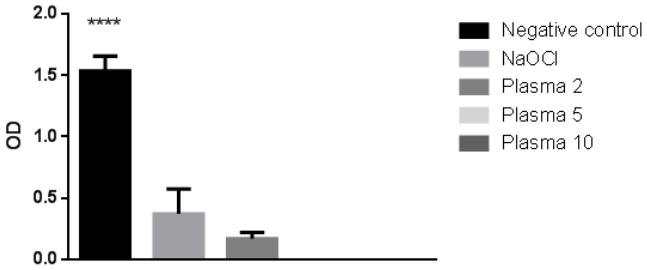

Figure 2 Cold plasma jet in 5 and 10 minutes showed a total inhibition of *E. faecalis* growth in compare with other treatments.

Discussion

Reinfection of the root canal system after the endodontics procedures can be explain by the presence of *E. faecalis* (mainly) in the dentinal tubules that in time can be activate and avoid the immune system response provoking a fully reinfection needs a retreatment or total tooth retire.[3-5] Many techniques for tooth disinfection have been proposed such ultrasonic, laser, and chemicals, however, the literature report that these techniques some times are functional and other do not.[1,2]

Cold plasma technology has been used for many proposed since disinfection of alimentary to medical device.[9] In this way many reports in dental sciences have been used this technology to disinfection of wounds and other application.[7] The study showed great effects in disinfection of E. faecalis in root canal systems. Application of cold plasma in the root canal over 5 minutes obtain a total disinfection of *E. faecalis*. These results are in concordance with previously studies that encounter total disinfection of other bacteria include *E. faecalis* from root canal systems.[9-11]

Microbial inactivation by cold plasma is produce by the vibration of O_2 and N_2 molecules producing the reactive oxygen and nitrogen species[12] that provoke a damage by oxidation of microbial cell membrane, proteins and DNA strands.[13] In this manner the cold plasma technology can be used has a disinfection therapy in dental endodontic procedures.

Conclusion

With the limits of this study can be suggest that cold plasm inhibit totally the *E. faecalis* biofilm growth in root canal system.

References

1. Kandaswamy D, Venkateshbabu N. Root canal irrigants. Journal of conservative dentistry: *JCD.* 2010;13(4):256–264.
2. Susila A, Minu J. Activated Irrigation vs. Conventional non–activated Irrigation in Endodontics – A Systematic Review. *European endodontic journal.* 2019;4(3):96–110.
3. Alghamdi F, Shakir M. The Influence of Enterococcus faecalis as a Dental Root Canal Pathogen on Endodontic Treatment: A Systematic Review. *Cureus.* 2020;12(3):e7257.
4. Sun X, Li S, Wang S, et al. The evaluation of E. faecalis colonies dissolution ability of sodium hypochlorite in microenvironment by a novel device. *Biomedical microdevices.* 2018;20(2):36.
5. Prada I, Mico–Munoz P, Giner–Lluesma T, et al. Influence of microbiology on endodontic failure. Literature review. Medicina oral, patologia oral y cirugia bucal. 2019;24(3):e364–e72.
6. Liao X, Muhammad AI, Chen S, et al. Bacterial spore inactivation induced by cold plasma. *Critical reviews in food science and nutrition.* 2019;59(16):2562–2572.
7. Wagner G, Eggers B, Duddeck D, et al. Influence of cold atmospheric plasma on dental implant materials – an in vitro analysis. Clinical oral investigations. 2021.
8. Yoshii S, Fujimoto M, Okuda M. In Vitro Evaluation of a Novel Root Canal Endoscope for Visualizing the Apex of Curved Root Canal Models and an Extracted Tooth. *Journal of endodontics.* 2018;44(12):1856–1861.
9. Pan J, Sun K, Liang Y, et al. Cold plasma therapy of a tooth root canal infected with enterococcus faecalis biofilms in vitro. *Journal of endodontics.* 2013;39(1):105–110.
10. Theinkom F, Singer L, Cieplik F, et al. Antibacterial efficacy of cold atmospheric plasma against Enterococcus faecalis planktonic cultures and biofilms in vitro. *PloS one.* 2019;14(11):e0223925.
11. Li Y, Sun K, Ye G, et al. Evaluation of Cold Plasma Treatment and Safety in Disinfecting 3–week Root Canal Enterococcus faecalis Biofilm In Vitro. *Journal of endodontics*. 2015;41(8):1325–1330.
12. Ji WO, Lee MH, Kim GH, et al. Quantitation of the ROS production in plasma and radiation treatments of biotargets. *Scientific reports.* 2019;9(1):19837.
13. Guo L, Zhao Y, Liu D, et al. Cold atmospheric–pressure plasma induces DNA–protein crosslinks through protein oxidation. *Free radical research.* 2018;52(7):783–798.

Citation: Figueroa-Banda R, Ponce-Soto L, Obando-Pereda G. Cold plasma: a good *Enterococcus faecalis* inhibitor – an in vitro tooth root canal pilot study. *J Dent Health Oral Disord Ther.* 2022;13(1):1–2. DOI: 10.15406/jdhodt.2022.13.00561