# Assessment of microbial reduction following rapid chair-side sterilization of contaminated endodontic files

Ban M. Jassim<sup>1</sup>, Hind M. Ahmed<sup>2</sup>, Majida K. Al-Hashimi<sup>3</sup>, Yasir B. Fadhil<sup>4</sup>

<sup>1</sup>College of Dentistry, AL-Esrass University, Baghdad, Iraq <sup>2</sup>Department of Applied Sciences, Technology University, Baghdad, Iraq <sup>3</sup>Department of Dentistry, Ashur University College, Baghdad, Iraq <sup>4</sup>Department of Biology, College of Education, AL-Iraqia University, Baghdad, Iraq

# ABSTRACT

**Background.** Endodontic treatment is carry out in order to preserve a tooth whose pulp has been damaged where the injury extends to the peri-radicular tissue. The endodontic treatment is cleaning, disinfection of the root canal space and shaping it, followed by the obturation of the root canal system. The presence and re-entry of microbes inside the canal is the main reason for post-treatment infections, which is considered as an endodontic failure. Therefore, disinfection throughout the treatment is very important. The aims of the study were to find microbial growth on brand sealed endodontic files, taken directly from the manufacture's packages, compare between different chemical compounds in rapid sterilization of contaminated endodontic files and determine the most effective chemical method in the sterilization.

**Methods.** The present study was performed to compare between different chemical compounds: 3% sodium hypochlorite, dendroid, chloroX and saline as the positive control in rapid sterilization of endodontic files in root canal treatment in dental practice using sealed endodontic files, taken directly from the manufacture's packages and sterile K-files sterilize by autoclaving.

**Results.** The results of this study, concerning *E. coli* bacteria, indicated that Dentroid scored the best results (90% free of any *E. coli* bacteria), followed by CHX (20% free of *E. coli* bacteria), then 3% NaOCL (10% free of *E. coli* bacteria). The results, concerning *S. aureus*, showed that normal saline and 3% NaOCL scored the highest contamination percentages (100%). While least and equal distribution of CHX and dentroid (90% free of *S. Aureus*).

**Conclusion.** The disinfecting solutions as CHX and Dentroid could be used as a method of rapid chair-side sterilization in clinical practice.

**Keywords:** Escherichia coli, Staphylococcus aureus endodontic files, sodium hypochlorit, dendroid, chloroX

# INTRODUCTION

Different microorganisms may induce different infectious diseases. Direct or indirect contamination may lead to transmission of infectious organisms [1]. In dentistry cross-contamination of infectious diseases among patients has been a big problem.

The main purpose of endodontic treatment is cleaning, disinfection of the root canal space and shaping it, followed by the obturation of the root canal system. The presence and re-entry of microbes inside the canal is the main reason for post-treatment infections, which is considered as an endodontic failure [2]. Therefore, disinfection throughout the treatment is very important.

Proper and good sterilization will prevent the spread of infectious diseases. In endodontics, different reusable instruments as reamers, files, gates glidden drill and peeso reamers are utilized for debridement and shaping of the root canal and in order to eliminate the bacterial colonies from the pulp canal space. Therefore, complete and aseptic techniques are mandatory. There are many methods to sterilize these instruments, such as dry heat sterilizer, autoclave, ethylene oxide gas or glass-bead sterilizer [3]. These techniques are not limited to, rubber dam isolation of the treated tooth, the use of sterilized instrument, and the introduction of antimicrobial irrigants during the treatment procedures [4].

Murgel, et al. 2017 evaluated new files directly from manufacturers packaging for the presence of surface debris and microorganisms. They showed that none of the instruments could be considered clean after removal from the package. Grease, epithelial cells, plastic and metallic spurs were found on the tested instruments. The purpose of the present study was to evaluate and compare four sterilization solutions, 3% sodium hypochlorite, dendroid, chloroX and saline as the positive control, in rapid sterilizing of endodontic files in dental practice [5].

The aims of the study were to find microbial growth on brand sealed endodontic files, taken directly from the manufacture's packages, compare between different chemical compounds in rapid sterilization of contaminated endodontic files and determine the most effective chemical method in the sterilization.

## MATERIAL AND METHOD

Materials used in this study were:

- 1. One hundred and sixty (160) test tubes.
- 2. Brain heart infusion broth.
- 3. K-files (CH-1338 Ballaigues-Switzerland), 21 mm long, size 25.
- 4. Sterilization solutions tested were 3% sodium hypochlorite, dentroid, chloroX and saline as the positive control.

#### Initial sample and culturing

One hundred and sixty (160) test tubes, containing 10 ml of Brain heart infusion broth which generally supports the growth of bacteria, were prepared. All the tubes were autoclaved at 121°C for 15 minutes; then they were divided into three experimental groups and one group as the negative control (forty test tubes each).

- Group A: inoculated with E. coli bacteria; after inoculation period of 24 hours at 37° C. Then sterile K-files (CH-1338 Ballaigues-Switzerland) were put in the test tubes, each test tube got 2 K-files.
- Group B: inoculated with Staphylococcus aureus bacteria; inoculation period of 24 hours at 37° C is followed. Sterile K-files were put in the test tubes; each test tube got 2 K-files.
- Group C: test tubes, left without any bacterial inoculation, got K-files, directly from manufacturer's package; 2 K-files in each test tube.
- Group D: without any bacterial inoculation (negative controls). Two sterile K-files were put in each test tube.

Any bacterial growth or turbidity in these test tubes considered as bacterial contamination.

The experimental test tubes with the files were incubated for two hours at 37° C, and then K-files were put in Petri-dishs and treated with different tested sterilization solutions, as follows:

All test group (A,B,C,and D), were divided into four subgroups ten tubes each. The files of the,

- first subgroup were treated with 3% sodium hypochlorite.
- second subgroup were treated with dendroid.
- third subgroup were treated with chloroX.
- last subgroup were treated with normal saline as the positive control.

All the files were treated with different tested sterilization solutions for only one minute. Then the files were washed with sterile distilled water for two minutes.

After washing the files were transferred into new sterile test tubes, each tube contains 10 ml of Brain heart infusion broth. The tubes were incubated for 24 hours at 37° C and checked for any turbidity that indicates the effectiveness of the tested sterilization solutions; turbidity for the positive result of bacterial growth and clear broth as the negative result, (Figure 1).



+ve control: Sealed K-file (without Autoclaving) S. aureus: Sterilized K-file incubated with S. aureus for 4 hrs. E. coli: Sterilized K-file incubated with E. coli for 4 hrs. Growth exam: Turbidity checked, and double checked by subculture on selective media

FIGURE 1. Flow chart of the study

For conformation of the results extra test was performed by taking 0.5 ml from all plane tubes and spreading it by a spreader in MacConkey ager which is a selective and differential media for G-ve *E. coli* and mannitol salt ager for *Staph. aureus*. After incubation (37°C for 24 hours) the colony seen on agar plate for definitive results which indicated the growth of bacteria.

## RESULTS

The results of this study, concerning *E. coli* bacteria, indicated that Dentroid scored the best results (90% free of any *E. coli* bacteria), followed by CHX (20% free of *E. coli* bacteria), then 3% NaOCL (10% free of *E. coli* bacteria). However, normal saline scored 100% contamination with *E. coli* bacteria. Significant differences between these solutions were found. Furthermore, multiple pair comparisons between them showed significant differences when compared each one of the tested solutions with Dentriod while others had no statistically significant differences (Figure 2).



FIGURE 2. Distribution and association of *E. coli* with the solution tested

The results, concerning *S. aureus*, showed that normal saline and 3% NaOCL scored the highest contamination percentages (100%). While least and equal distribution of CHX and dentroid (90% free of *S. aureus*) were found with significant association between them. Furthermore, multiple pair comparisons between them were performed. When each solution compared with Dentroid, the results were significant except when compared with CHX, which was not significant. Comparing the CHX with both normal saline and 3% NaOCL, the results were significant (Figure 3).



**FIGURE 3.** Distribution and association of *S. aureus* with tested solutions

#### DISCUSSION

The present study was performed because in certain cases the endodontist may need urgent or rapid sterilization of the endodontics files or reames. Therefore, this study tested the chair-side sterilization of K-files intentionally contaminated with two types of bacteria cultures, (*E. coli* and *S. aureus*).

NaOCl is used in this study since it has been widely used as an endodontic irrigant and has a good sterilizing action on contaminated cones [6]. CHX is a broad spectrum antimicrobial solution, which has low toxicity [7]. It has been used in endodontics either as an irrigant solution or as an intracanal medication solution, giving good and acceptable performance [8, 9].

The results of this study, Dentroid which is used for sterilization the endodontic files, contaminated with *E. coli* bacteria, showed complete sterilization of the endodontic instrument (90%) while CHX (20%) was less effective in sterilizing the files from *E. coli* bacteria. It was clear that both Dentroid and CHX were capable of sterilizing endodontic instruments, better than the efficiency of 3% NaOCL [10,11].

## CONCLUSION

Within the limitations of this study, it can be concluded that the disinfecting solutions as CHX and Dentroid could be used as a method of rapid chairside sterilization in clinical practice.

*Conflict of interest:* none declared *Financial support:* none declared

#### REFERENCES

- Miller CH. Infection control and office safety sterilization: Disciplined microbial control. Den Clin North Am. 1991;35:339-55. PMID: 2032579.
- Ravi Chandra PV, Kumar VH, Reddy SJ, Kiran DR, Krishna MN, Kumar GV. Biofilm forming capacity of Enterococcus faecalis on Gutta-percha points treated with four disinfectants using confocal scanning laser microscope: An in vitro study. *Dent Res J (Isfahan)*. 2015 Jul-Aug;12(4):331-6. doi: 10.4103/1735-3327.161446. PMID: 26288622; PMCID: PMC4533190.
- Raju TB, Garapati S, Agrawal R, Reddy S, Razdan A, Kumar SK. Sterilizing Endodontic Files by four different sterilization methods to prevent cross-infection - An In-vitro Study. *J Int Oral Health.* 2013 Dec;5(6):108-12. Epub 2013 Dec 26. PMID: 24453454; PMCID: PMC3895727.
- Rajkumar K, Lakshminarayanan L. The Effectiveness of two commonly used methods of Sterilizing Endodontics. *J Indian Dent Assoc.* 2001;72:245-8. https://scholar.google.co.in/citations?view\_op=view\_ citation&hl=en&user=YNoE23gAAAAJ&citation\_for\_ view=YNoE23gAAAAJ:RGFaLdJalmkC
- Ferreira Murgel CA, Walton RE, Rittman B, Pecora JD. A comparison of techniques for cleaning endodontic files after usage: a quantitative scanning electron microscopic study. *J Endod*. 1990 May;16(5):214-7. doi: 10.1016/S0099-2399(06)81672-9. PMID: 2074413.

- Isci S, Yoldas O, Dumani A. Effects of sodium hypochlorite and chlorhexidine solutions on Resilon (synthetic polymer based root canal filling material) cones: an atomic force microscopy study. *J Endod*. 2006 Oct;32(10):967-9. doi: 10.1016/j.joen.2006.04.001. Epub 2006 Jul 3. PMID: 16982275.
- Valois CR, Silva LP, Azevedo RB. Effects of 2% chlorhexidine and 5.25% sodium hypochlorite on gutta-percha cones studied by atomic force microscopy. *Int Endod J.* 2005 Jul;38(7):425-9. doi: 10.1111/j.1365-2591.2005.00940.x. PMID: 15946261.
- Gomes BP, Vianna ME, Zaia AA, Almeida JF, Souza-Filho FJ, Ferraz CC. Chlorhexidine in endodontics. *Braz Dent J.* 2013;24(2):89-102. doi: 10.1590/0103-6440201302188
- Merdad KA, Alghamdi FT. Assessment of the Sterility of New Endodontic Files Received From the Manufacturer Using Microbial Culture and Scanning Electron Microscopic Analysis: An In Vitro Study. *Cureus.* 2022 Aug 17;14(8):e28092. doi: 10.7759/cureus.28092
- Gnau HL, Goodell GG, Imamura GM. Rapid chairside sterilization of endodontic files using 6% sodium hypochlorite. *J Endod.* 2009 Sep;35(9):12534. doi: 10.1016/j.joen.2009.05.032
- Roth TP, Whitney SI, Walker SG, Friedman S. Microbial contamination of endodontic files received from the manufacturer. *J Endod.* 2006 Jul;32(7):649-51. doi: 10.1016/j.joen.2005.09.006