

# Effectiveness of Root Canal Irrigants in Different Concentrations on Facultative Anaerobic & Obligate Aerobic Microorganisms : An Exvivo Study

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## Abstract

The aim of this study was to test the effect of different concentrations of sodium hypochlorite & Chlorhexidine on the following bacterial strains-

- Escherichia Coli ATCC 25922 (Facultative anaerobic)
- Enterococcus Faecalis ATCC 29212 (Facultative anaerobic)
- Pseudomonas A eruginosa ATCC 27853 (obligate aerobic)

This experiment was carried out using a microorganism concentration equal to approximately  $1.5 \times 10^8$  bacteria.

Root canal irrigants were used in the following concentration-

Chlorhexidine-0.2%, 0.4%, 0.5%, 1% & Sodium hypochlorite in the concentration of 0.5%, 1%, 2.5%, 3%. Each substance was kept in contact with the bacterial species for 10, 20 or 30 min. For every bacterial strain starting from a concentration equal to  $1.5 \times 10^7$  CFU/ml

Serial tenfold dilutions were made, showed results that all irrigants had a bactericidal effect on both facultative anaerobic and obligate aerobic strains in all concentration and even after short period of contact. Thus it can be implied- that clinically Chlorhexidine is a useful substitute in patients who are allergic to sodium hypochlorite.

**Key Words:** Chlorhexidine (CHX), Sodium hypochlorite (NaOCl), Facultative anaerobic, Facultative anaerobic, Obligate aerobic.

## Introduction

The reduction and elimination of bacteria and their byproducts should be given the utmost importance towards achieving a successful endodontic therapy.<sup>1</sup>

Root canal irrigation plays a key role in the success of endodontic treatment, because it helps in the progressive removal of the smear layer and neutralizes the root canal microbic flora.<sup>1</sup>

## Objective

The purpose of this invitro experiment was to test the antimicrobial efficiency of Sodium hypochlorite, Chlorhexidine in different concentrations on the microorganisms that are most frequently found inside the root canal system.

## Materials & Method

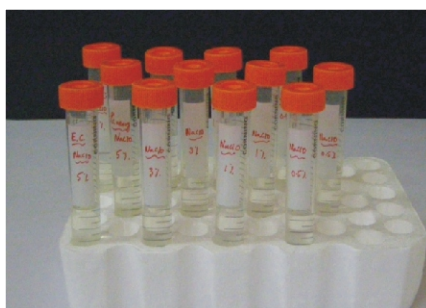


Sodium Hypochlorite

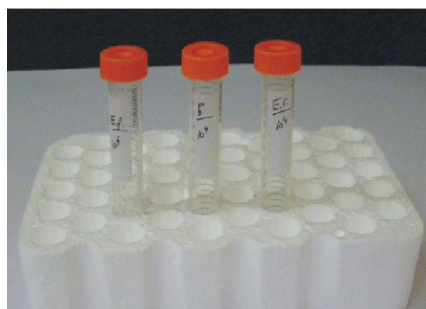
Sterile Water

Chlorhexidine Gluconate

- Sodium hypochlorite
- Sterile water
- Chlorhexidine gluconate
- Conical tubes
- Mcfarland
- Serial 10 Fold Dilutions are Prepared for Enterococcus Faecalis-ATCC29212, Escherichia Coli-ATCC25922, Pseudomonas Aeruginosa-ATCC27853
- Control plates
- Incubator



Concentration of sodium hypochlorite



Serial 10 Fold Dilutions are Prepared for Enterococcus Faecalis-ATCC29212, Escherichia Coli-ATCC25922, Pseudomonas Aeruginosa-ATCC 27853



Control Plates

## Methodology

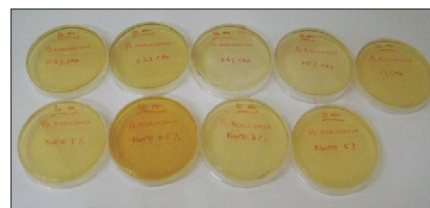
- Root canal irrigants were used in the following concentrations-
  - Sodium hypochlorite - 0.5%, 1%, 3%, 5%
  - Chlorhexidine - 0.2%, 0.3%, 0.4%, 0.5%, 1%
- These concentrations were prepared using sterile distilled water.
- Irrigants were kept in contact with the bacterial species for 10, 20 and 30 min.
- For every bacterial strain starting from a conc. equal to 0.5Mcfarland serial ten fold dilutions were made thus obtaining a

final dilution corresponding to  $10^4$  CFU/ml.

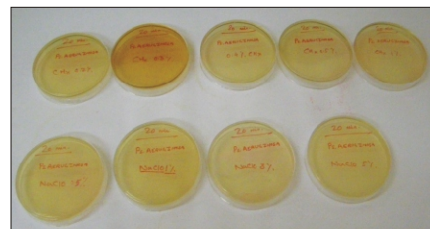
- At 10 minutes interval 1ml of the contents of the tube were placed in brain heart infusion agar and incubated at 37° centigrade for 24 hours.
- Same procedure was followed at 20 minutes and at 30 minutes and finally all the samples were incubated at 37° centigrade for 24 hours.

## Results

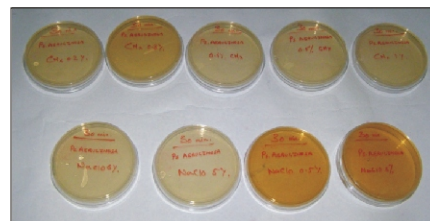
- Pseudomonas Aeruginosa



After 10 Minutes of Incubation

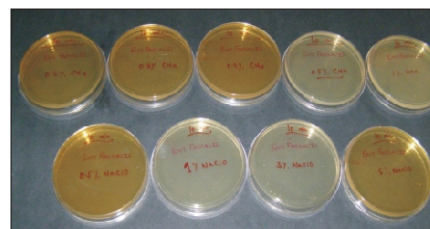


After 20 Minutes of Incubation

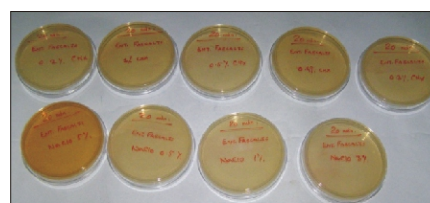


After 30 Minutes of Incubation

- Enterococcus Faecalis



After 10 Minutes of Incubation



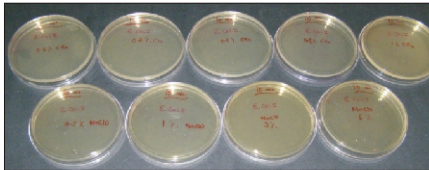
After 20 Minutes of Incubation



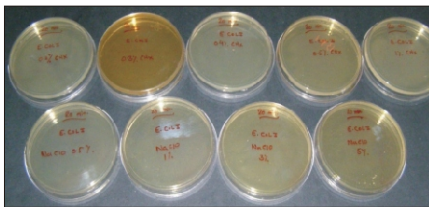


After 30 Minutes of Incubation

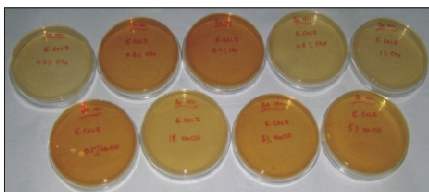
• **Escherichia Coli**



After 10 Minutes of Incubation

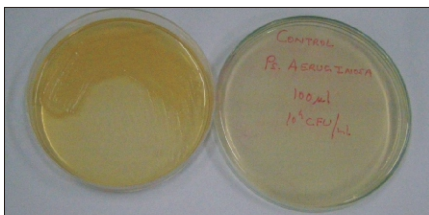


After 20 Minutes of Incubation



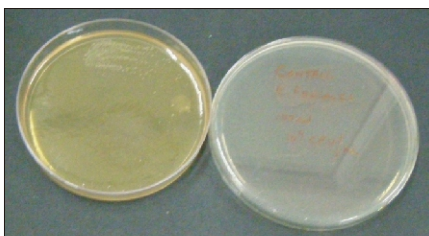
After 30 Minutes of Incubation

**Pseudomonas Aeruginosa**



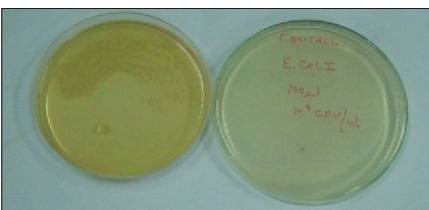
Control Plates

**Enterococcus Faecalis**



Control Plates

**Escherichia Coli**



Control Plates

- There was no difference among the irrigants tested-sodium hypochlorite and Chlorhexidine in varying concentrations.
- The vitality of all microorganisms tested was always 0 after a contact period of 10min with each root canal irrigant.
- The same results were obtained after a period of 20 and 30 min of contact.
- Thus, all irrigants tested had a strong bactericidal effect on all bacterial strains.

**Discussion**

The elimination of microorganisms from infected root canal systems is a complicated task involving the use of various instrumentation techniques, irrigation regimens and intracanal medicaments. Mechanical instrumentation alone does not result in a bacteria free root canal system and when complex anatomy of root canal system is considered (Hess 1925). On the other hand, ex vivo and clinical evidence has shown that mechanical instrumentation leaves significant portions of root canal walls untouched (Peterd et al 2001) and complete elimination of bacteria by instrumentation alone is unlikely to occur.<sup>2</sup> Necrotic tissue remnants in the root canals, serve as a nutrient source for any remaining microorganisms<sup>3</sup>. Furthermore, tissue remnants also impede the antimicrobial effects of root canal irrigants and medicaments. Therefore, some form of disinfection and irrigation is necessary to remove residual tissue and microorganisms<sup>2</sup>. Unfortunately, irrigation is one of the most neglected phases of endodontic treatment.<sup>4</sup>

Irrigants serve following purposes: Lubricate the canal walls during instrumentation, removes debris by flushing the canals, dissolve organic matter, dissolve inorganic matter, destroy microbial organisms and to aid cleaning in areas that are inaccessible to mechanical cleansing methods.<sup>5</sup> Through the years, numerous agents have been tested for their suitability as a root canal irrigant.<sup>6</sup> Prior to 1940, water was the most commonly used endodontic irrigant. It was readily available, inexpensive and like all non-viscous fluids provided a lubricating effect making instrumentation of the canal walls easier.

Acids such as 30% hydrochloric acid and 50% Sulphuric acid were used as late as the 1940's with little or no understanding of the hazards these agents posed for the periradicular tissues. In present study some easily available irrigants were used. NaOCl is the most commonly used irrigant. It is an excellent organic solvent and effective antimicrobial agent.<sup>7</sup> The antimicrobial efficacy of the solution is due to its ability to oxidize and hydrolyze cell proteins. NaOCl has an approximate pH 11-12 and when comes in contact with tissue proteins, nitrogen, formaldehyde and acetaldehyde are formed within a short time and peptide links are broken resulting in dissolution of proteins. During this process, hydrogen in amino groups is replaced by chlorine thereby forming chloramines which is responsible for antimicrobial effectiveness. Necrotic tissue and pus are thus dissolved and antimicrobial agent can reach and clean the infected area

better.<sup>8</sup> However, NaOCl is malodorous and very caustic. It can cause violent tissue reactions characterised by pain, swelling, haemorrhage and in some cases the development of secondary infection and parasthesia when solution is injected beyond the apex.

Hypersensitivity reactions have also been reported. Chlorhexidine is being suggested as an useful substitute in patients who are allergic to sodium hypochlorite.<sup>1</sup> Chlorhexidine was developed in the late 1940s in the research laboratories of Imperial Chemical Industries Ltd.

CHX is a potent antiseptic which is widely used for chemical plaque control in the oral cavity. CHX is active against a wide range of gram positive and gram negative organisms, yeast, fungi, facultative anaerobes and aerobes.<sup>9</sup> CHX is particularly effective against *Enterococcus faecalis*, a microorganism that has been implicated in treatment failures. CHX has also been shown to have a long term antimicrobial properties because of its unique ability to bind to hydroxyapatite. A gradual release of this bound CHX could maintain an even level of molecule sufficient to create a bacteriostatic milieu in the root canal over a period of time.<sup>10</sup>

Chlorhexidine has been studied for its various properties with an objective of being an alternative to sodium hypochlorite.

- Chlorhexidine was found to be equally effective and exhibited bactericidal effect towards the bacteria most frequently present in the root canal system.
- From a microbiological point of view no difference was found between 1% and 0.2%
- In this study only the microbiological action of sodium hypochlorite and chlorhexidine at different concentrations is assessed. However, Antimicrobial activity is not the only requirement of an endodontic irrigant.
- Root canal irrigants should have other characteristics and dissolution of pulp tissue is one of the property which a root canal irrigant should possess so until chlorhexidine is effective as pulp tissue debridement.
- Sodium hypochlorite, must be considered the irrigant of choice because it has tissue dissolving properties.

**Conclusion**

- Sodium hypochlorite is one of the commonly used root canal irrigant owing to its bactericidal properties.
- However, Chlorhexidine may still be useful as an alternative endodontic irrigant.
- Its excellent antimicrobial properties indicate it could be a useful substitute in patients who are allergic to sodium hypochlorite

**References**

References are available on request at [editor@healtalkt.com](mailto:editor@healtalkt.com)