# Comparative Evaluation of Anti-Bacterial Efficacy of Chlorine Dioxide ($ClO_2$) & Sodium Hypochlorite (NaOCl) on E. Faecalis : An in Vitro Study

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## Abstract

### Aim

The aim of this study was to evaluate the comparative antibacterial efficacy of 10% chlorine dioxide and 3% Sodium hypochlorite as an endodontic irrigant.

### Material & Methods

Thirty intact extracted teeth were taken. Apical 5mm was removed with diamond disc& roots were further sectioned into cylindrical discs of 5mm height lumens were standardized with (no: 25) round bur. Smear layer was removed with EDTA & Sodium hypochloride (NaOCl). Disks were autoclaved and then infected with standardized concentrations of Enterococcus (E) faecalis (ATCC) in brain heart infusion broth for 72hr .The discs were divided into two groups of fifteen each & were exposed to 10% Chlorine dioxide($ClO_2$) & 3% NaOCl for 5, 15 , 30 min respectively. At intervals of 5, 15, 30 Dentinal shavings were collected from the lumen (Fig. 4) and weighed & transferred to tube containing known quantity of normal saline and were then frozen, pulverized, serially diluted in phosphate buffered saline, and plated cultured in Bile-Esculin Agar (BEA), which is a specific culture medium for E. faecalis[12]. The plates were then incubated at and later evaluated for the presence of bacteria and the constituents of the colonies.

### Results & Conclusion

After 5 minutes 10% $ClO_2$ was significantly more effective than NaOCl in removing E. fecalis. Efficacy at 15& 30 min were comparable.

**Keywords:** Antimicrobial, Endodontics, Enterococcus faecalis, irrigation, Sodium hypochlorite, chlorine dioxide.

### Introduction

The success of endodontic treatment depends, to a great degree, on the elimination of microorganisms from the root canal system. [1]

Irrigating the canals with antimicrobial solution is an important step to the decrease the number of microorganisms from root canal system .[2]

E. Faecalis is a recognized pathogen in post treatment endodontic failure. E. faecalis is found to be resistant against Ca $(OH)_2$ & is can survive in the root canals without synergistic effect of other bacteria.[3,4]

Different kinds of antimicrobial rinses have been introduced for disinfecting root canal system. However, no irrigant has been formulated that can accomplish all the desirable qualities while avoiding the undesirable one's & still show high rate of bactericidal efficacy[5].

NaOCl is the most commonly used irrigant for the Root canal system. However it is extremely toxic to the periapical tissues if it is passed beyond the apex.[6] Furthermore, NaOCl can't eliminate E. Faecalis from the canal system. [7]

$ClO_2$ has long been used as a disinfectant for drinking water. Due to its disinfectant property it has found its applications in various industries like [8]:

- Dairy industry.
- Beverage industry.
- The pulp and paper industries.
- Fruit and vegetable processing.
- Increased use in municipal potable water treatment facilities.

Chlorine dioxide has the following advantages:

- More effective than chlorine and chloramine.
- Taste and odour are acceptable
- Environment friendly
- Safety-no byproducts released
- Does not create resistant strains
- Easy to use
- Economic

This strong antibacterial activity makes it a potentially useful endodontic irrigant.

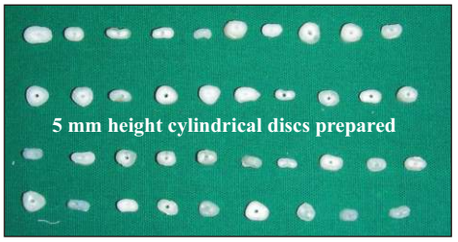
### Aim & Objectives

The purpose of this in vitro and in vivo study was to compare the antimicrobial effects of 3% NaOCl & 10% $ClO_2$ as root canal irrigant on E. faecalis-contaminated root canals of human teeth.

### Material & Method

Thirty sound single-rooted human teeth which had been extracted because of periodontal diseases or orthodontic treatment were selected. The teeth included maxillary and mandibular incisors and premolars and had one canal each only. ( Fig. 1)

The external surfaces of the roots were immediately debrided after extraction and all the adherent tissues were removed using a curette. Then all teeth were placed in 0.5% NaOCl for 24 hours for surface disinfection and were stored in 0.9% sterile physiologic serum at room temperature until used.All teeth were radio graphically investigated to ensure that they have single canals and that there are no calcifications. 5 mm ht cylindrical discs were then prepared using diamond disks[9]. (Fig. 2)



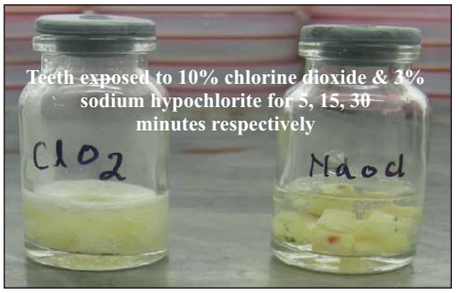

Pure culture of E.faecalis grown in Brain Heart Infusion broth (BHI) was used to contaminate the root canals. Standardized lumens were filled with either sterile Brain Heart Infusion Broth (contamination controls, n = 10) or BHI containing placed in a broth containing infections pure E. faecalis suspensions (ATCC 29212).

Next, all specimens were kept in special flasks in an incubator at 37ºC for 24 hours. All the specimens were monitored during this period [11]. Subsequent to incubation, all the specimens were separately retrieved from the flasks under aseptic conditions. Then each specimen was immersed in 3 ml of sterile physiologic serum in a test tube and was shaken 3 times for 30 seconds each time on a rotator to remove the excess of the culture media from the specimens. To simulate endodontic instrumentation the lumens were again enlarged.

The study disks were divided into 2 groups according to the solution used for the irrigant:

A. 10% chlorine dioxide
B. 3% sodium hypochlorite

The teeth were then exposed to 10% chlorine dioxide & 3% sodium hypochlorite for 5, 15, 30 min respectively. (Fig. 3)




At intervals of 5, 15, 30 Dentinal shavings were collected from the lumen (Fig. 4) and individually transported and

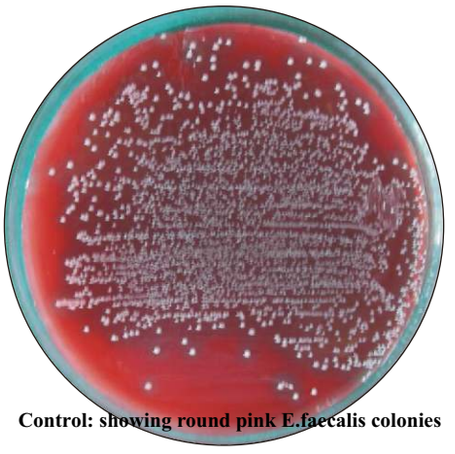

cultured in Bile-Esculin Agar (BEA), which is a specific culture medium for E. faecalis[12]. The plates were then incubated at and later evaluated for the presence of bacteria and the constituents of the colonies (Fig. 5)


Significance in the distribution of negative cultures between two groups was tested by Fisher's Exact test.

## Results

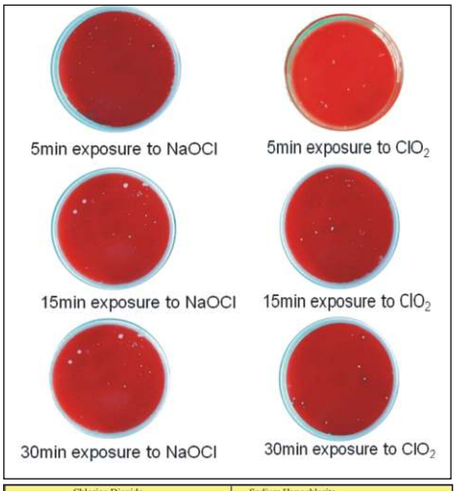
E. Faecalis was taken as a control as round pink colonies on BHI plate with ATCC(29212). (Fig. 6)

For statistical analysis Fisher Exact test was done. 10% $ClO_2$ was significantly more effective in removing E. faecalis ($p<0.05$) at



**Control: showing round pink E.faecalis colonies**

5 min. However, comparable results were found at 15 and 30 minutes (Fig 7 & Table 1).




| | Chlorine Dioxide | | | | Sodium Hypochlorite | | | |
|---|---|---|---|---|---|---|---|---|
| | 5 min | 15 min | 30 min | | 5 min | 15 min | 30 min |
| A1 | NG | NG | NG | B1 | NG | NG | NG |
| A2 | NG | NG | NG | B2 | G | NG | NG |
| A3 | NG | NG | NG | B3 | NG | NG | NG |
| A4 | G | G | NG | B4 | NG | G | G |
| A5 | NG | NG | NG | B5 | G | NG | NG |
| A6 | NG | NG | NG | B6 | NG | NG | NG |
| A7 | NG | NG | NG | B7 | G | NG | NG |
| A8 | G | G | NG | B8 | NG | NG | NG |
| A9 | NG | NG | NG | B9 | G | NG | NG |
| A10 | NG | NG | NG | B10 | NG | NG | NG |
| A11 | NG | NG | NG | B11 | NG | G | NG |
| A12 | NG | NG | NG | B12 | NG | NG | NG |
| A13 | G | G | NG | B13 | G | NG | NG |
| A14 | NG | NG | G | B14 | G | NG | NG |
| A15 | NG | NG | NG | B15 | G | NG | NG |
| | 3/12 | 3/12 | 1/14 | | 7/8 | 2/13 | 1/14 |

**NG: No growth ; G: Growth of E. faecalis**

## Discussion

The objective of this study was to compare the bactericidal activity of chlorine dioxide with sodium hypochlorite as root

canal irrigant in endodontic treatment protocols.

E faecalis is taken for the study as it is the most common detected in asymptomatic, persistent endodontic infections. Its prevalence in such infections ranges from 24% to 77%[13].

The percentage of irrigants and duration of exposure to irrigating solutions were selected on the basis of previous studies[9, 14, 15,16].

NaOCl is a commonly used intra-canal irrigating solution and its antibacterial properties are attributed to hypochlorous acid[17]. Sjögren *et al.* demonstrated that approximately 40% of the canals remain contaminated subsequent to debridement with 2.5% NaOCl[18]. Siqueira *et al.* demonstrated that the rate of canal contamination subsequent to the use of NaOCl is 30-40%[19].

The main drawback of NaOCl is the toxicity to the periapical tissues[20,21], bad odour, discolouration of dental equipment, and destruction of permanent tooth follicles and oral mucosa. It can also cause pharyngeal oedema and oesophageal burns when unintentionally swallowed[22]

Chlorine dioxide was selected for this study because of its strong antimicrobial properties, ease of use and widespread availability.

This study demonstrated that 10% $ClO_2$ is more effective than 3% sodium hypochlorite at 5min in effectively removing E. faecalis.

At 30 min both were equally effective; this is comparable to a previous study[9].

## Conclusion

This study demonstrated that 10% $ClO_2$ is more effective than 3% sodium hypochlorite at 5 min in effectively removing E. faecalis.

Further studies are needed to determine the effect of these findings in clinical settings and also to establish chlorine dioxide toxicity, effects on dentin, and minimum inhibitory concentration.

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