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Original article

Effect of chlorine dioxide and sodium hypochlorite on the dissolution of human pulp tissue – An in vitro study

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ABSTRACT

Background: Organic tissue dissolution is an important property of an irrigant which aids in the success of root canal treatment. Recent studies have advocated the use of Chlorine dioxide as an endodontic irrigant. The aim of this study is to compare the dissolution efficacy of chlorine dioxide and sodium hypochlorite on human pulp tissue.

Methods: In this study, 2% Sodium hypochlorite, 5% Chlorine dioxide and isotonic saline solution (control) were used. Thirty human pulp tissue specimens were exposed to three test solutions ($n = 10$) for 30 min following which the loss of weight was compared from the original weight by using a digital analytical balance.

Results: Sodium hypochlorite was more efficient in dissolving human pulp tissue when compared to Chlorine dioxide. Isotonic saline solution failed to dissolve any of the specimens.

Conclusion: 5% Chlorine dioxide is capable of dissolving human pulp tissue but sodium hypochlorite was more effective.

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Introduction

Chemomechanical preparation of the root canal treatment involves efficient mechanical instrumentation and irrigation procedures. The use of an antimicrobial substance for irrigation is indicated because it significantly enhances bacterial elimination.^{1–3}

Sodium hypochlorite (NaOCl) remains the most widely used irrigant solution because of its pronounced antimicrobial

activity and the ability to dissolve organic matter.^{4,5} However, it has been shown that sodium hypochlorite has toxic effects on vital tissues, resulting in haemolysis, skin ulceration and necrosis.⁶ It has a pH of approximately 12 and causes injury primarily by oxidation of proteins. Therefore, the emphasis on finding an alternative root canal irrigant should continue.

Chlorine dioxide has recently come under consideration as a possible root canal irrigant. It is reported to be tuberculocidal, bactericidal, virucidal, and fungicidal. Chlorine

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dioxide can be more effective as a disinfectant when compared to sodium hypochlorite because HOCl^- (Hypochlorous acid) or OCl^- (hypochlorite ions), two effective components of sodium hypochlorite, when come in contact with negatively charged bacterial cell wall might be repelled as both are negatively charged, thus causing less penetration and absorption of the disinfectant into the membranes, whereas chlorine dioxide irrigant, exists as gas in water, which enables it to permeate through bacterial cell membranes and bring about its destruction at a wide range of pH from 3 to 9.⁷

The recent detection of Cytomegalovirus and Epstein–Barr virus associated with periradicular lesions may promote the use of chlorine dioxide, which kills both enveloped and non enveloped viruses, at the same time by adsorbing onto and penetrating the protein coat of the viral capsid.⁸ Also, for its antifungal activity, Chlorine dioxide damages *Candida albicans* ATCC10231 plasma membranes mainly by permeabilization, under minimal fungicidal concentration of 20 mmol/l.^{1,9}

Brian D et al concluded that chlorine dioxide is less cytotoxic as compared to Sodium hypochlorite.¹⁰ Sodium hypochlorite reacts with natural organic matter to produce trihalomethanes and haloacetic acids both of which are animal carcinogens and suspected human carcinogens. Chlorine dioxide produces little or no trihalomethanes, and may be a better dental disinfectant than NaOCl .¹¹

To date, there are no studies reported on the human pulp tissue dissolving effect of chlorine dioxide. Hence, the purpose of this study is to compare the dissolution efficacy of chlorine dioxide and sodium hypochlorite on human pulp tissue.

Materials and methods

Human pulp tissue was collected from 22 M teeth which were extracted for periodontal reasons or which were impacted. After the extraction, periodontal tissues were removed with a brush and the teeth placed in saline and refrigerated for 24 h. A continuous groove was prepared on the proximal root and crown using a diamond bur (Fig. 1) and then carefully split into

Fig. 1 – A groove prepared on the root surface.

two using a surgical blade (Fig. 2), pulp tissue was then removed entirely using an excavator, washed in distilled water to remove the blood and refrigerated for 30 min so as to assist in pulp tissue sectioning. Total 30 pulp specimens were acquired by sectioning with a surgical blade so that each specimen had a standard weight of 0.015 g. Before weighing, each piece of the pulp tissue was blotted on a filter paper and dried and then weighed using a digital analytical balance (Shimadzu, EL Series) (Fig. 3). Specimens were handled only with cotton pliers to avoid contamination. Pulp tissues were then divided randomly into three experimental groups ($n = 10$).

1. Group A: pulp specimens were placed in 5 ml of 2% Sodium Hypochlorite solution.
2. Group B: Pulp specimens were placed in 5 ml of 5% Chlorine dioxide solution. The 5% chlorine dioxide solution (BLO-OUT, GOODLOOKS, INDIA) was prepared by mixing equal parts of solutions A and B according to the manufacturer's instructions just before use. The pH of Chlorine dioxide was 4.67, as measured by a pH meter. (LI-120 pH meter, Elico Ltd.)
3. Group C: - (negative control): Pulp specimens were placed in 5 ml of saline.

Pulp specimens in each group were placed in individual closed vials containing the test solution, and all the vials were placed on a vibrator thus agitating the test solution to simulate fluid movement during root canal instrumentation. The pulp tissue was weighed after 30 min. Before weighing, the specimens were washed with distilled water to remove precipitate, blotted on a filter paper, dried and then weighed. All samples were weighed by a single investigator who was unaware of how each was to be treated.^{11,12} The percentage of tissue weight loss in each specimen was calculated and was implemented to analyze statistically. The percentage of tissue weight loss after subjecting to the solutions was calculated

Fig. 2 – Pulp tissue obtained after splitting the tooth.

Fig. 3 – Pulp specimen placed on digital analytical balance.

and the values were analyzed statistically by using one-way analysis of variance and Tukey HSD test.

Results

One-way analysis of variance indicated that Group A (2% Sodium hypochlorite) dissolved the pulp tissue more efficiently than Group B (5% Chlorine dioxide) and the difference was statistically significant (0.05 level). Tukey HSD test indicated that both Group A (2% Sodium hypochlorite) and Group B (5% Chlorine dioxide) dissolved the tissue pieces more effectively than the saline control ($p < .05$). Saline did not dissolve any of the specimens.

Mean percentage of weight loss of the human pulp tissue samples after exposure to test solutions and ANOVA summary are shown in Table 1.

Discussion

Previous studies^{13,14} have suggested an inverse relationship between pH of a solution and time taken by the solution to dissolve the tissue. In the current study, the pH of chlorine dioxide was 4.67, much lower than the pH of sodium hypochlorite (pH = 12), it might be one of the reasons of chlorine

dioxide being less effective in dissolving pulp tissue. In a similar study,¹¹ the pH of chlorine dioxide was raised up to 12 by using sodium hydroxide and it was concluded that chlorine dioxide is at par with sodium hypochlorite for dissolving bovine pulp tissue, but according to Deininger et al⁷ chlorine dioxide exhibits biocidal efficacy only over a pH range of 3–9. Moreover the addition of sodium hydroxide will result in making the aqueous solution of chlorine dioxide ineffective by breaking it down into sodium chlorate, sodium chlorite and water.

Sodium chlorate is a known non-selective herbicide¹⁵ and its presence in any root canal irrigant is questionable. The pH control of aqueous solution of chlorine dioxide can be best achieved during the time of manufacture.

Currently, the minimal inhibitory concentration of chlorine dioxide for use in endodontics is not known, previous studies used a concentration of 13.8%¹¹ but experimental results indicate that chlorine dioxide is unstable above the concentration of 9.5% ($[\text{ClO}_2]/[\text{air}]$)¹⁶ and also at a concentration of 13.8% the solution will be extremely pungent thus adding one major disadvantage. To minimize the damage caused to the fragile human pulp tissue samples while drying, weight reduction was calculated after the exposure of pulp tissue for a continuous period of 30 min.

Conclusion

Within the limitations of this study, it can be concluded that 5% chlorine dioxide is capable of dissolving human pulp tissue but is not as efficacious as 2% sodium hypochlorite. Further research needs to be done on determining optimum concentration of aqueous solution of chlorine dioxide, its effect on dentin, smear layer removing capability when used at a lower pH and its compatibility with various obturating and restorative materials used in Endodontics.

Intellectual contribution of authors

Study concept: Capt Sandeep Singh.

Drafting & Manuscript revision: Capt Sandeep Singh, Brig (Retd) Ramen Sinha.

Statistical analysis: Capt Sandeep Singh, Amber Ather.

Study supervision: Col S K Kar, S N Limaye.

Conflicts of interest

All authors have none to declare.

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Table 1 – Mean percentage of weight loss of the human pulp tissue samples.

Test solutions			Weight change (%) ± SD		
2% Sodium hypochlorite			−29.9 ± 5.674		
5% Chlorine dioxide			−17.1 ± 8.008		
ANOVA Summary					
Source	SS	df	MS	F	p
Treatment	817.9205	1	817.9205	16.98	0.000642

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