Article published online: 2021-11-01

ORIGINAL ARTICLE

Comparison of human pulp tissue dissolution capacities of different irrigating solutions: An *in vitro* study

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ABSTRACT

Background: Organic tissue dissolution is considered as one of the most important and desirable property of endodontic irrigant, any soft tissue remnant, harboring bacteria, left in the canal after endodontic therapy may be the cause of failure. **Aim:** The present study aimed at assessing and comparing the human pulp dissolution (thereby eliminating the bacteria) capacity of some potential endodontic irrigants viz., sodium hypochlorite (NaOCl) (2.5% and 5.25%), chlorine dioxide (5%) and peracetic acid (5%). **Materials and Methods:** Forty human pulp specimens from extracted premolars were taken and weighed. They were immersed in test solution for 30 min, dried on filter paper and weighed again. The percentage weight loss was calculated and statistically analyzed. **Conclusion:** It can be concluded that NaOCl showed the best tissue dissolution capacity, followed by 5% peracetic acid.

Key words
Chlorine dioxide, dental pulp, peracetic acid, sodium hypochlorite

Ethics: The guidelines of the Institutional Ethical Committee were followed.

INTRODUCTION

The success of endodontic therapy depends mainly on the effectiveness of the chemo-mechanical preparation done during the process. Mechanical preparation alone is ineffective in completely debriding the canal because of the complex anatomy of the root canal system.[1] Any tissue remnant left in the canal may prove to be a source for the surviving bacteria and result in endodontic failure.[2] Therefore, endodontic irrigation plays a major role in completely debriding the canal. The antimicrobial substances used as endodontic irrigant should possess tissue dissolution property,[3] enhance bacterial elimination[4,5] and at the same time have low systemic toxicity.[6]

Access this article online	
Quick Response Code:	Website: www.ejgd.org
	DOI: 10.4103/2278-9626.154168

Sodium hypochlorite (NaOCl) solution is considered the irrigant of choice because of its antimicrobial activity and pulp dissolution capacity.[7] It also possesses cytotoxic property. Hence, the need for finding an alternative to NaOCl has been raised in various studies.[3,8,9]

Chlorine dioxide (ClO_2) has currently emerged and gained attention as a possible alternative to NaOCl. It is chemically similar to NaOCl, but is less toxic since it does not form chlorinated hydrocarbons on contact with the organic matter.[3] It has also been reported as tuberculocidal, bactericidal, virucidal and fungicidal.[9] However, studies concerning human pulp tissue dissolution capacity of ClO_2 are lacking.

Peracetic acid is one of the most commonly used disinfectants in hospitals and industries.[10] It has antibacterial, sporicidal, antifungal and antiviral properties.[11] Preliminary studies have shown that it can remove the smear layer,[12] which has been attributed to its acetic acid content. However, some issues related to its caustic effect at higher concentration have been raised.[11]

A comprehensive comparative study dealing with the tissue dissolution capacities of ClO_2, NaOCl and peracetic acid has not been performed. Hence, the purpose of this study was to evaluate organic tissue dissolution capacity of the above-mentioned irrigants.

MATERIALS AND METHODS

Forty freshly extracted premolars were collected from the outpatient department of the Department of Oral and Maxillofacial Surgery. Only the teeth extracted due to orthodontic reasons were considered. Prior ethical approval was taken from the Institutional Ethical Committee. Any tooth with restoration, caries, endodontic treatment or wasting disease was excluded. After extraction, the tooth was cleaned with a brush and stored in saline. The teeth were divided into four groups with 10 teeth in each group (n = 10), as given below:

Group I: NaOCl 5.25% (Vishal Dentocare Pvt. Limited, Ahmedabad, India)
Group II: NaOCl 2.5% (Vishal Dentocare Pvt. Limited, Ahmedabad, India)
Group III: ClO_2 5% (Acuro Organics Limited, New Delhi, India)
Group IV: Peracetic acid 5% (Acuro Organics Limited, New Delhi, India).

For retrieving the pulp tissue, the teeth were split into two halves. Two longitudinal grooves were made on the proximal surface of the tooth with the help of long neck straight bur. They were split with the help of chisel and mallet. Pulp specimens were washed in distilled water to remove any remnants of blood and refrigerated for 1 h to assist in sectioning. Pulp tissue samples were standardized to weight range of 0.023–0.028 g by cutting them with the help of No. 15 BP blade. The samples were weighed on an analytical balance (MX-7301A Anamed, India). After weighing, the samples were immersed in test solution (10 ml) in a small plastic vial and kept for 30 min. They were placed on a vibrator (Unident, India) at 3000 rpm for 1 min to achieve proper immersion and simulate conditions present in the pulp canal during biomechanical preparation. After 30 min, the solution was filtered through a filter paper (Whatman) and left overnight for drying. The weight of the residual pulp tissue was measured. Readings were noted, and percentage weight reduction was calculated. Statistical analysis was performed using SPSS version 15.0. Analysis of variance, followed by Tukey honestly significant difference test were used to compare differences between groups. The confidence level of the study was kept at 95%. Hence a $P < 0.05$ indicated a statistically significant intergroup difference.

RESULTS

Both 5.25% and 2.5% NaOCl showed elimination [Table 1]. Mean reduction for 5% ClO_2 was 45.40 ± 1.51%, whereas the same for 5% peracetic acid was 55.60 ± 1.58%. Statistically, there was a significant difference among groups ($P < 0.001$). On comparing the data, no difference was observed between 5.25% and 2.5% NaOCl. However, both 5% ClO_2 and 5% peracetic acid solution displayed significantly lower mean value than 5.25% and 2.5% NaOCl groups ($P < 0.001$). On comparing 5% ClO_2 with 5% peracetic acid solution, mean reduction was found to be significantly lower in 5% ClO_2 than 5% peracetic acid group ($P < 0.001$) [Table 2].

DISCUSSION

Pulp tissue dissolution capability is one of the desirable properties of an endodontic irrigant. A wide selection of irrigant solutions with varying properties and advantages are now available. A limited number of studies have been performed on the human pulp tissue dissolution capability of these new irrigating solutions. The present study aimed at assessing the pulp tissue dissolution (thereby eliminating the harbored bacteria) capacity of two potential endodontic irrigants – ClO_2 and peracetic acid with the most commonly used and established irrigant, NaOCl.

Human pulp tissue was used to assess and quantitatively compare the test solutions. Various other tissues have been used in previous studies like bovine pulp,[13,14] umbilical cord,[15] pig palatal mucosa,[16] rat dermal connective tissue.[17] However, these tissues do not simulate the conditions present clinically within the human root canal. Hence, human pulp tissue was considered for this study. Tissue dissolution capacity depends on various factors, like the amount of irrigant, amount and area of the tissue being tested and the frequency of agitation.[18] Keeping these factors into consideration, the human pulp tissue samples of similar weight were used, the amount of test solution was fixed and the continuous agitation

Table 1: Mean percentage reduction in different groups

Test solution	*n*	Mean±SD	Minimum	Maximum
5.25% NaOCl	10	100.00±0.00	100	100
2.5% NaOCl	10	100.00±0.00	100	100
5% chlorine dioxide	10	45.40±1.51	43	48
5% peracetic acid	10	55.60±1.58	53	58
Total	40	75.25±25.35	43	100

NaOCl - Sodium hypochlorite, SD - Standard deviation

Table 2: Between group comparison (Tukey's HSD test)

Comparison between groups	Mean difference	SE	*P*
5.25% NaOCl versus 2.5% NaOCl	0.000	0.488	1.000
5.25% NaOCl versus 5% chlorine dioxide	54.600	0.488	<0.001
5.25% NaOCl versus 5% peracetic acid	44.440	0.488	<0.001
2.5% NaOCl versus 5% chlorine dioxide	54.600	0.488	<0.001
2.5% NaOCl versus 5% peracetic acid	44.440	0.488	<0.001
5% chlorine dioxide versus 5% peracetic acid	−10.200	0.488	<0.001

NaOCl - Sodium hypochlorite, SE - Standard error, HSD - Honestly significant difference

was done on the vibrator at 3000 rpm for 1 min to simulate intra-canal fluid movement during irrigation. After splitting the tooth, pulp tissue was removed using spoon excavator, washed in distilled water to remove any remnants of blood. The pulp specimen was then refrigerated for 1 h to assist in sectioning.[9] Digital analytical balance used for weighing the samples was calibrated every time to increase the degree of precision. Before weighing, the tissue samples were blotted dry using a filter paper.

Mean percentage weight loss was calculated for each solution tested by a simple mathematical formula viz., weight loss (in mg)/initial tissue weight (in mg) × 100. This gave an accurate quantitative means of assessment of pulp tissue dissolution capacity of the tested solutions. Mean percentage weight reduction has been listed in Table 1. It was found to be 100% in Groups I and II, 45.4% in Group III and 55.6% in Group IV. Group I (5.25% NaOCl) served as control because complete dissolution was anticipated in this group as various studies done in the past have demonstrated this effect.[13,18,19] This effect of NaOCl has been attributed to its proteolytic activity. Group II (2.5% NaOCl) showed similar results. Various studies have demonstrated similar results for 2.5% NaOCl, but for a longer time tested (2 h).[20,21] We found similar results in 30 min, which may be explained on the basis of tissue specimen used (human pulp tissue than the bovine pulp tissue and palatal mucosa).

Antimicrobial properties of ClO_2 have been reported by Eddy *et al.*,[22] along with its tissue dissolution capacity by Cobankara *et al.*[3] Peracetic acid is also one of the strongest disinfectants with antimicrobial property and smear layer removal effect,[11] but a comprehensive quantitative study assessing human pulp tissue dissolution was missing. In our study significant difference was found between Groups II and III, Group II and Group IV, suggesting NaOCl to be a better tissue solvent. Group IV showed significantly better tissue dissolution than Group III, suggesting peracetic acid to be a better tissue solvent than ClO_2. ClO_2 has been shown to be toxic at higher pH, besides having antimicrobial effect.[23] The percentage of peracetic acid tested in this study was within the safe limits as suggested by Kühlfluck and Klammt.[24] Moreover, smear layer removal capacity of peracetic acid adds to its advocation as a possible root canal irrigant.

CONCLUSION

Within the limitations of the study, it can be concluded that both 2.5% and 5.25% NaOCl are an effective pulp tissue solvent. However, 5% peracetic acid may prove to be a suitable alternative.

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How to cite this article: Jain A, Shrivastava TV, Tabassum S, Bahuguna R. Comparison of human pulp tissue dissolution capacities of different irrigating solutions: An *in vitro* study. Eur J Gen Dent 2015;4:64-7.

Source of Support: Nil, **Conflict of Interest:** None declared.