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Original Article

Assessment and Comparison of Human Pulp Tissue Dissolution Capacities of Endodontic Irrigating Solutions: An *In Vitro* Study

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Abstract

Background: Over the past few decades practice of endodontics has been transformed by newer technology and materials and increased the success rate of endodontic treatment. Disinfecting and cleaning by chemomechanical procedures are the prerequisites for successful root canal treatment. Organic tissue dissolution is considered as one of the most vital 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 treatment.

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) (2.5–5.25%), chlorine dioxide (5%), and peracetic acid (5%).

Materials and Methods: Sixty 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 weight loss in percentage was calculated and statistically analyzed.

Results: Both 5.25–2.5% NaOCl showed mean value of 100%. Whereas, mean reduction for 5% ClO 2 was 55.30 ± 1.87%, whereas the same for 5% peracetic acid was 65.70 ± 1.69%.

Conclusion: NaOCl showed the best tissue dissolution capacity, followed by 5% peracetic acid.

Key words: Chlorine dioxide, Dental pulp, Endodontic irrigant, Peracetic acid, Sodium hypochlorite

INTRODUCTION

The field of endodontics has modernized with the developments of several techniques and materials over the past decades.[1] Disinfecting and cleaning by chemomechanical procedures are the fundamentals for successful root canal treatment. With advanced instrumentation technology, though root canal shaping can be efficiently achieved, effective cleaning of the entire root canal system, still, remains a challenge.[2,3] The attainment of endodontic therapy depends mainly on the efficiency of the chemomechanical preparation done during the process.[4] Any tissue residue left in the canal may prove to be a source for the surviving bacteria which ultimately result in endodontic failure.[5] The antimicrobial substances used as endodontic irrigant should possess tissue dissolution property,[6] enhance bacterial eradication,[7,8] and at the same time have low systemic toxicity.[9] Therefore, endodontic irrigation plays a most important role in completely debriding the canal.

Sodium hypochlorite (NaOCl) solution is one of the irrigant of choice because of its antimicrobial activity and pulp dissolution capacity.[10] The prewarming of NaOCl is an easy, and the most commonly used method to increase the reaction rate and enhance the dissolution of

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organic residues. About, a temperature range of 50–60°C is suggested for warming the NaOCl.[11] NaOCl also have cytotoxic property. Hence, the need for finding an alternative to NaOCl has been raised in various studies.

Chlorine dioxide is chemically similar to chlorine or hypochlorite but is less toxic and it is one of the familiar household bleach. It is reported to be tuberculocidal, bactericidal, virucidal, and fungicidal.[12] It is mostly commonly used for sewage water disinfection, industrial process water treatment, industrial air treatment. However, there is lack of literature regarding human pulp tissue dissolution capacity of ClO_2.[12] Peracetic acid is one of the most commonly used disinfectants in hospitals and industries areas.[13] Preliminary studies have shown that it can remove the smear layer,[14] which has been attributed to its acetic acid content.

It has antibacterial, sporicidal, antifungal and antiviral properties.[15] However, some issues related to its caustic effect at higher concentration have been raised.[15] In most of the previous studies, tissue solubility of numerous irrigants has been measured using bovine mucosa and bovine pulp tissue. Very few studies have been done on solubilizing effects of various irrigants on human pulp tissue.

Therefore, the aim of this *in-vitro* study is to compare the human pulp tissue dissolution by different concentrations of, NaOCl, chlorine dioxide and peracetic acid.

MATERIALS AND METHODS

This is *in-vitro* study conducted to compare the human pulp tissue dissolution by different concentrations of, NaOCl, chlorine dioxide and peracetic acid. The study protocol was approved by the Scientific Advisory committee and Institutional Ethics Committee. Sixty freshly extracted premolars were obtained from the outpatient department of the Department of Oral and Maxillofacial Surgery and from private clinics. Teeth extracted for orthodontic reasons were considered. Any tooth with restoration, caries, endodontic treatment, or wasting disease was excluded from the study. After extraction, the tooth was cleaned with a brush and stored in saline. The teeth were divided into four groups with 15 teeth in each group ($n = 15$), Group I: NaOCl 5.25%, Group II: NaOCl 2.5%, Group III: ClO2 5%, and Group IV: Peracetic acid 5%.

For retrieving the pulp tissue, the teeth were divided into two halves. Two longitudinal grooves were made on the proximal surface of the tooth with the help straight bur. They were split with the help of chisel and mallet. Pulp specimens were washed with help of distilled water to remove any remnants of blood remnants and refrigerated for 1 h for 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 submerged 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 using 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's *post-hoc* test significant difference test was 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 mean value of 100% [Table 1]. Whereas, mean reduction for 5% ClO_2 was 55.30 ± 1.87%, whereas the same for 5% peracetic acid was 65.70 ± 1.69%. 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

Chemomechanical preparation is essential procedure for the success of root canal instrumentation. Pulp tissue

Table 1: Mean percentage reduction in different groups

Test Solution	n	Mean± SD
5.25% NaOCl	15	100.00 ±0.00
2.5% NaOCl	15	100.00 ±0.00
5% Chlorine dioxide	15	55.30±1.87
5% Peracetic acid	15	65.70±1.69
Total	60	80.25±28.45

(NaOCl- Sodium Hypocloride, SD- Standard deviation)

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

Comparison between groups	Mean difference	p
5.25% NaOCl versus 2.5% NaOCl	0.000	1.000
5.25% NaOCl versus 5% Chlorine dioxide	65.800	<0.001
5.25% NaOCl versus 5% Peracetic acid	55.540	<0.001
2.5% NaOCl versus 5% Chlorine dioxide	45.600	<0.001
2.5% NaOCl versus 5% Peracetic acid	55.540	<0.001
5% Chlorine dioxide versus 5% Peracetic acid	-15.100	<0.001

(NaOCl- Sodium Hypocloride)

dissolution capability is one of the desired properties of an endodontic irrigant. The principal properties expected from an ideal irrigation solution are antimicrobial activity, water solubility, low toxicity to periradicular tissues with tissue solvent ability.[16] Over the years, several chemicals and their combinations have been studied for potential use as endodontic irrigant. From various literatures, it can be seen that NaOCl, though toxic, named "gold standard" as it is an excellent antimicrobial agent and solvent of organic tissue.

Therefore, NaOCl was used as a positive control group.[17] Various different types of tissues have been used in earlier studies such as bovine pulp,[18,19] umbilical cord,[20] pig palatal mucosa,[21] and rat dermal connective tissue.[22] However, these tissues do not simulate the conditions present clinically within the human root canal. Hence, human pulp tissue was better to considered for this study. Tissue dissolution capacity depends on various factors, such as amount of irrigant, amount and area of the tissue being tested, and the frequency of agitation.[23]

In our study, the study showed that 5% NaOCl was the most effective pulp tissue solvent among all irrigants. This is because of the ionization of NaOCl to liberate hypochlorous acid (HOCl) and hydroxyl ions in an aqueous environment.[24] When hydroxyl ion levels decrease as a result of the saponification and amino acid neutralization reactions, the pH also decreases, thereby favoring the formation of HOCl molecules.[12] The chloramination reaction is then initiated which results in degradation and hydrolysis of amino acids.[12]

In our study, it was found that 100% in Groups I and II, 55.3% in Group III and 65.7% 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.[18,23,25] 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).[26,27] 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).

In our study, 2.5% NaOCl and 5% NaOCl showed no significant difference in pulp dissolution at 30 min. Whereas study done by Taneja *et al.*[12] found that 5% NaOCl was less effective than 5% NaOCl at 30 min. Hand *et al.*[22] have also suggested that 5.25% NaOCl was a more effective solvent than 2.5% NaOCl.

In our study, significant difference was found between Groups II and III, Group II and Group IV, signifying NaOCl to be a better tissue solvent. Group IV showed significantly better tissue dissolution than Group III, suggesting peracetic acid also to be a better tissue solvent than ClO_2. ClO_2 has been shown to be toxic at higher pH, besides having antimicrobial effect.[28] The percentage of peracetic acid tested in this study was within the safe limits as suggested by Kühlfluck and Klammt.[29] Moreover, smear layer removal capacity of peracetic acid adds to its advocation as a possible root canal irrigant.

CONCLUSION

It can be concluded that both 2.5–5.25% NaOCl are a more effective pulp tissue solvent that chlorine dioxide and peracetic acid.

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