Dissolution of pulp tissue by aqueous solution of chlorhexidine digluconate and chlorhexidine digluconate gel

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Abstract

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Aim To evaluate the activity of various root canal irrigants on bovine pulp tissue.

Methodology The irrigants tested were: 0.5, 1.0 and 2.5% sodium hypochlorite; 2% aqueous solution of chlorhexidine digluconate; 2% chlorhexidine digluconate gel (Natrosol™); and distilled water as control. Bovine pulp fragments were weighed and placed in contact with 20 mL of each tested substance in a centrifuge at 150 r.p.m. until total dissolution. Dissolution speed was calculated by dividing pulp weight by dissolution time. Statistical analysis was performed using the Kruskal–Wallis test.

Results Distilled water and both solutions of chlorhexidine did not dissolve the pulp tissue within 6 h. Mean dissolution speeds for 0.5, 1.0 and 2.5% sodium hypochlorite solutions were 0.31, 0.43 and 0.55 mg min^{-1}, respectively. The solvent ability of chlorhexidine solutions was similar to that of distilled water. The results for sodium hypochlorite solutions, chlorhexidine solutions and distilled water were statistically different ($P > 0.01$).

Conclusions Both chlorhexidine preparations and distilled water were not able to dissolve pulp tissue. All sodium hypochlorite solutions were efficient in dissolving pulp tissue; the dissolution speed varied with the concentration of the solution.

Keywords: chlorhexidine, irrigants, pulp.

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Introduction

The success of root canal treatment relies on thorough chemomechanical procedures (Stewart 1955). The persistence of residual pulp tissue, infected dentine or bacteria in the root canal system may be responsible for treatment failure (Callahan 1894, Grossman & Meiman 1941, Moorer & Wesselink 1982). The use of irrigating solutions facilitates debridement of the root canal space and is important for the success of root canal treatment (Cunningham & Balekjian 1980).

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Several studies have been conducted in search for an irrigant that meets four major properties: antimicrobial activity, non-toxicity to periapical tissues, water solubility and capacity to dissolve organic matter (Byströn & Sundqvist 1985, Jeansonne & White 1994, Kuruvilla & Kamath 1998).

Sodium hypochlorite has been used as a therapeutic agent since the 1930s for root canal infection (Walker 1936). It is as an excellent organic solvent and an effective antimicrobial agent (Byströn & Sundqvist 1985, Nakamura *et al.* 1985).

Chlorhexidine has been widely used in dentistry because of its broad-spectrum antimicrobial activity and substantivity. For these reasons, chlorhexidine has been recommended for root canal treatment. Chlorhexidine digluconate is formulated as an aqueous solution or gel preparation. The liquid form is most frequently used in dentistry, but chlorhexidine gel has recently been investigated for use in endodontics (Ferraz *et al*. 2001). In spite of a large number of papers on the antimicrobial activity and substantivity of chlorhexidine, a review of the literature did not yield any study on the property of chlorhexidine gel to dissolve organic matter.

The aim of this study was to evaluate the dissolution of pulp tissue by a 2% aqueous solution of chlorhexidine digluconate and by a 2% chlorhexidine gel in comparison with sodium hypochlorite at different concentrations.

Materials and methods

Tissue preparation

Eight incisors were extracted from fresh bovine mandibles and stored at $-20^{\circ}C$ until required. The teeth were thawed to room temperature, and two longitudinal grooves were cut on the proximal surfaces with a diamond bur. The teeth were split in half. Pulp tissue was removed and washed in distilled water. Each pulp sample was divided into four pieces of similar volume, resulting in five fragments for each of the six groups below.

Preparation of solutions

Sodium hypochlorite solutions were prepared by dissolution of a concentrated solution. Final concentrations were titered and adjusted to pH 9.0 by addition of boric acid; pH was measured by a pH meter. An external laboratory (Essencial Pharma, Itapetininga, Brazil) prepared the chlorhexidine digluconate irrigants. Distilled water and sodium hypochlorite solutions were prepared at the Endodontic Laboratory, Dental School, São Paulo University. All irrigants were kept under refrigeration until required.

Dissolution process

The fragments were weighed on an analytical balance and placed in contact with 20 mL of each irrigant in a beaker, as follows:

- Group 1: distilled water (control group)
- Group 2: 0.5% sodium hypochlorite
- Group 3: 1% sodium hypochlorite
- Group 4: 2.5% sodium hypochlorite
- Group 5: aqueous solution of 2% chlorhexidine digluconate
- Group 6: 2% chlorhexidine digluconate gel

The beakers were placed in an agitator at 150 r.p.m. until the fragments were totally dissolved, or for 6 h.

Time of dissolution was assessed from the time when the fragment was placed in contact with the irrigant to the moment when its total disappearance was observed by an impartial evaluator. Dissolution speed was calculated as weight of fragment divided by dissolution time.

Statistical analysis was performed with the Kruskal–Wallis test.

Results

Weight and time of dissolution of each pulp tissue specimen are shown in Table 1. All sodium hypochlorite solutions dissolved the pulp tissue. Chlorhexidine preparations, as well as distilled water, were not able to dissolve the fragments in a period of 6 h.

Dissolution speed was calculated as weight of fragment divided by time spent for its dissolution (mg min^{-1}). The means for the five samples in each group are shown in Fig. 1.

Mean dissolution speeds for 0.5, 1.0 and 2.5% sodium hypochlorite solutions were 0.31, 0.43 and 0.55 mg min^{-1}, respectively; these means were significantly different ($P > 0.01$). Likewise, results for chlorhexidine preparations

Table 1 Weight and time of dissolution of each pulp tissue sample in contact with each irrigant solution

		Sample 1		Sample 2		Sample 3		Sample 4		Sample 5	
		Weight (mg)	Time (min)	Weight (mg)	Time (min)	Weight (mg)	Time (min)	Weight (mg)	Time (min)	Weight (mg)	Time (min)
I	Distilled water	18.7	[a]	38.3	[a]	19.1	[a]	31.6	[a]	21.4	[a]
II	0.5% NaOCl	29.1	76.95	35.1	110.88	39.6	124.81	34.3	102.66	20.9	85.5
III	1.0% NaOCl	31.5	74.2	38.3	91.63	32.3	72.38	30	59.25	20.7	52.75
IV	2.5% NaOCl	25.8	57.88	34.3	63.1	30.6	47.75	32.6	45.8	17.7	40.88
V	2% gel chlorhexidine	24.7	[a]	22.6	[a]	42.0	[a]	22.8	[a]	25.0	[a]
VI	2% aqueous chlorhexidine	29	[a]	33.6	[a]	31.6	[a]	39.7	[a]	24.7	[a]

[a]Indeterminate time surpassing 6 h.

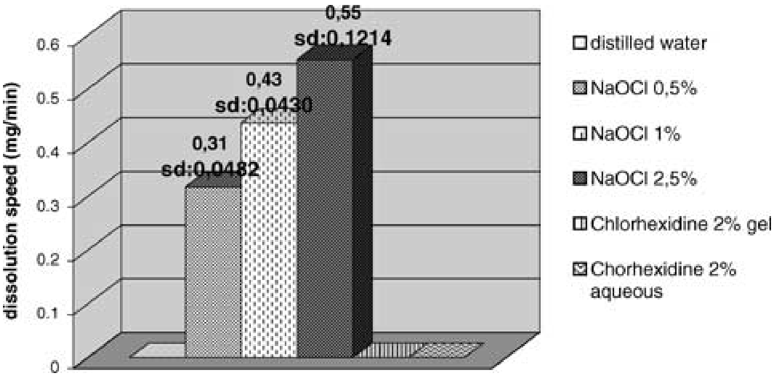

Figure 1 Mean dissolution speed of pulp tissue by the six endodontic irrigants.

and distilled water were statistically different from the results for the three hypochlorite solutions ($P > 0.01$).

Discussion

Callahan (1894) and Grossman (1941) demonstrated the importance of the solvent ability of an endodontic irrigant and emphasized that the elimination of pulp tissue from the root canal was important for the ultimate success of root canal treatment. Bovine pulp tissue was selected because of its similarity to human pulp tissue (Koskinen *et al.* 1980).

Moorer & Wesselink (1982) showed that tissue dissolution was dependent on three factors: frequency of agitation, amount of organic matter in relation to amount of irrigant in the system and surface area of tissue that was available. The present study was standardized using constant mechanical agitation and the same volume of irrigant for all samples. Furthermore, pulp samples were of similar weight (mean 29.25 mg).

The results were reported for dissolution speed, not for time of dissolution, to compensate for the variability in fragment weight. The results obtained for sodium hypochlorite are consistent with those reported in other studies that also investigated the association of solution concentration and solvent ability (Moorer & Wesselink 1982, Nakamura *et al.* 1985).

Aqueous solutions of sodium hypochlorite dissociate into HOCl and NaOH. As the substance reacts with the organic matter, HOCl and NaOH concentrations decrease. Moorer & Wesselink (1982) calculated the interaction of NaOCl with organic matter by measuring the consumption of chlorine available in the solution (HOCl and OCL^{-}). Considering that the HOCl and NaOH availability is closely associated with the pH of the solution, all sodium hypochlorite dilutions were adjusted to pH 9.0, which, in addition, promotes a higher stability of the solution (Siqueira 2000).

By electron microscopy, McComb & Smith (1975) demonstrated that the use of sodium hypochlorite produces walls virtually free of debris throughout the whole canal, including the apical region, and compared these results with canal instrumented with water irrigation.

It is important to remember that mechanical preparation, either with manual or rotatory instruments, removes most of the organic matter and promotes debridement, increasing the contact surface of the remnant soft tissue with the irrigant solution. Therefore, the solution would act in small portions of tissue and, with large volumes of irrigant solution, the dissolution speed would be enhanced.

Some authors demonstrated the efficiency of low concentrations of sodium hypochlorite solutions when irrigating root canals. Spangberg *et al.* (1973) showed that solutions at 0.5 and 1% concentrations were safer and as effective for canal disinfection.

Chlorhexidine is the chemical form of a bis-biguanide that is used and commercialized as a gluconate salt. It is, next to fluoride, the preventive agent that has been most intensively researched in dentistry (Fardal & Turnbull 1986). It is active against a wide range of Gram-positive and Gram-negative organisms, fungi, facultative anaerobes and aerobes (Henessey 1973). The action of chlorhexidine is the result of its adsorption to the microorganism cell wall, resulting in leakage of intracellular components (Gjermo 1974).

The activity of chlorhexidine used as a canal irrigant or intracanal medication has been studied and it is recommended for endodontic use because of its antimi crobial activity and presumed non-toxicity and intracanal substantivity. Many authors have demonstrated the antimicrobial action of chlorhexidine (Henessey 1973, Jeansonne & White 1994, D'Arcangelo *et al.* 1999, Ferraz *et al.* 2001); other authors reported its inability to dissolve organic matter (Jeansonne & White 1994, D'Arcangelo *et al.* 1999, Marley *et al.* 2001), but little evidence is available.

Conclusions

According to the methodology applied: (i) both chlorhexidine preparations and distilled water were not able to dissolve pulp tissue; (ii) all sodium hypochlorite solutions proved to be efficient in dissolving pulp tissue; and (iii) the dissolution speed varied proportionally to the concentration of the sodium hypochlorite solution; the more concentrated the solution, the higher the speed of dissolution.

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