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Effects of Calcium Hydroxide and Sodium Hypochlorite on the Dissolution of Necrotic Porcine Muscle Tissue

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The dissolving effect of the endodontic medicaments calcium hydroxide ($Ca(OH)_2$) and sodium hypochlorite (NaOCl) on necrotic tissue was studied. Pieces of necrotic porcine muscular tissue were placed in either a 0.5% NaOCl solution (Dakin's solution), $Ca(OH)_2$ mixed with water, or a NaOCl solution following pretreatment in $Ca(OH)_2$ for various time intervals. The tissue pieces placed in 0.5% NaOCl were not completely dissolved after 12 days. When NaOCl solution was changed every 30 min, the tissue was completely dissolved after 3 h. Pieces placed in $Ca(OH)_2$ exhibited a marked swelling and a jelly-like consistency. The increase in weight was maintained for 24 h, after which a decrease was noted. After 12 days, the tissue was completely dissolved. Pretreatment with $Ca(OH)_2$ for 30 min caused the tissue to dissolve in NaOCl within 90 min. Pieces that were pretreated for 24 h or 7 days dissolved within 60 min. As a control, tissue pieces were kept in isotonic saline solution and these were not dissolved after 12 days.

Apparently, long-term treatment with $Ca(OH)_2$ can dissolve necrotic tissue and pretreatment with $Ca(OH)_2$ can enhance the tissue dissolving effect of NaOCl.

A major goal in endodontic treatment is to obtain a clean root canal. Toward this end the instrumentation is generally supported by the use of a solution for irrigation. Root canals have a complex anatomy and there are areas that may not be accessible for instrumentation. Therefore, agents capable of dissolving necrotic tissue are used (1).

Sodium hypochlorite was successfully used in wound treatment during World War I (2) and later it was introduced in endodontic therapy as an antibacterial tissue-dissolving irrigant (3–6). It is extensively used and its tissue dissolving capability has been studied under varying test conditions (7).

Favorable results have been obtained when using calcium hydroxide as an intracanal dressing between visits (8, 9). Also, it has been found that the use of calcium hydroxide as a long-term intracanal dressing enhances the prognosis for treatment of young human teeth (10). The reason for this beneficial effect is not thoroughly understood. However, it has been demonstrated in monkey teeth that the use of calcium hydroxide results in considerably cleaner root canals compared with root canals where calcium hydroxide had not been used (11). As strong bases are able to denaturate and hydrolyze proteins (12), calcium hydroxide may contribute to a breakdown of intracanal soft tissue remnants thus rendering a cleaner root canal. In clinical practice, calcium hydroxide and sodium hypochlorite are often used in the same root canals. The purpose of this study was to assess in vitro, the separate and combined effects of calcium hydroxide and sodium hypochlorite on the dissolution of necrotic tissue.

MATERIALS AND METHODS

Porcine muscular tissue that had been kept refrigerated in 100% humidity for 2 wk was divided into pieces, 2 × 1 × 1 mm (Fig. 1*A*). The weight of the pieces was adjusted to 0.020 using a Sauter balance-type 404 (August Sauter K.G., Ebingen, Germany). The pieces were kept in 100% humidity to avoid drying. All experiments were performed at room temperature. Ten tissue pieces were used in each of the following seven groups.

In group 1 the tissue pieces were placed and kept in 20 ml of a paste of calcium hydroxide and water (0.6 g per ml).

In groups 2 and 3 the tissue pieces were kept in 20 ml of 0.5% sodium hypochlorite solution. In group 2 the tissue pieces were kept in the same solution and in group 3 the solution was changed every 30 min until the pieces were completely dissolved. The solution was freshly prepared from equal parts of a 1% sodium hypochlorite solution and a 2% sodium bicarbonate solution.

In groups 4 to 6 the tissue pieces were placed into 20 ml of a paste of calcium hydroxide and water. After 30 min (group 4), 24 h (group 5), or 7 days (group 6), the pieces were transferred into 20 ml of a 0.5% sodium hypochlorite solution.

The control (group 7) tissue pieces were kept in 20 ml of isotonic saline.

The tissue pieces placed into calcium hydroxide, 0.5% sodium hypochlorite solution, and isotonic saline were weighed after 30, 60, and 90 min and then after 10 and 24 h.

Prior to weighing, the pieces kept in calcium hydroxide were rinsed in saline and all pieces were blotted on a filter paper. After 24 h the experiment continued for the undissolved tissue until it was dissolved or for 12 days.

RESULTS

A marked swelling occurred in the tissue pieces that were placed in calcium hydroxide (group 1). After 30 min in calcium hydroxide paste, the average weight of the tissue pieces was 0.042 g. The tissue was whitish and had a jelly-like consistency (Fig. 1*B*). The weight was maintained for 24 h. Thereafter, the tissue pieces continuously decreased and could not be detected by means of a sieve after 12 days.

An initial swelling occurred also in groups 2 and 3 (tissue pieces that were placed in 0.5% sodium hypochlorite). After 60 min the average weight of the tissue pieces was 0.038 g and after 90 min 0.018 g (Fig. 1*C*). When kept in the same solution (group 2), the tissue pieces maintained this weight at 10 h and were not completely dissolved after 12 days. The tissue pieces that were transferred to freshly mixed solution of sodium hypochlorite every 30 min (group 3) reached a weight maximum after 60 min and were completely dissolved after 180 min.

The tissue pieces that were pretreated in calcium hydroxide paste for 30 min were completely dissolved within 90 min in the sodium hypochlorite solution (group 4). A partially dissolved specimen that was in the NaOCl for 30 min is seen in Fig. 1*D*. The pieces that were pretreated in calcium hydroxide for 24 h (group 5) or 7 days (group 6) were completely dissolved within 60 min when placed in 0.5% sodium hypochlorite.

The control tissue pieces (group 7) underwent a slight swelling and the weight increased to an average of 0.022 g. This weight was maintained for 12 days. The results are summarized in Table 1.

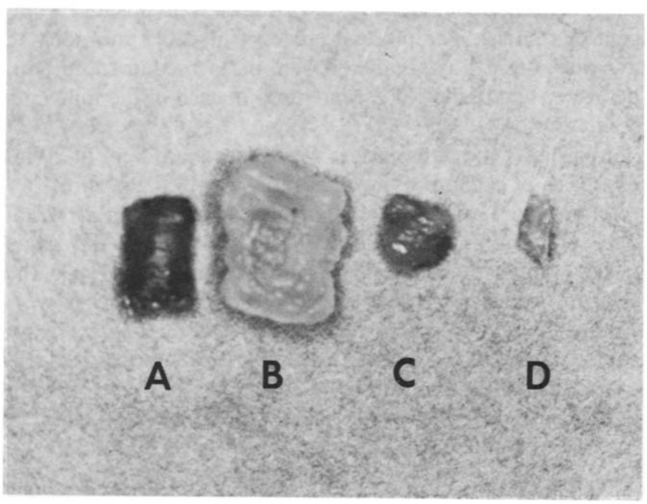

FIG 1. *A*, A piece of porcine muscular tissue, 2 × 1 × 1 mm. Pieces like this were used for the experiments. *B*, A piece of tissue similar to that shown in *A* that was kept in a calcium hydroxide-water paste for 30 min. The tissue is swollen, whitish, and jelly-like. *C*, A piece of tissue similar to that shown in *A* that was kept in a 0.5% sodium hypochlorite solution for 90 min. *D*, A piece of tissue similar to that shown in *A* that was kept in a sodium hypochlorite solution for 30 min after pretreatment in a calcium hydroxide-water paste for 30 min.

DISCUSSION

In this in vitro study of tissue dissolution, muscular tissue was used for practical reasons in order to get a sufficient amount of tissue. The dissolution rates of human pulpal connective tissue and porcine muscular tissue may not be similar, but the differences between dissolution rates of tissues influenced and not influenced by calcium hydroxide or sodium hypochlorite are conceivably the same.

The tissue solvent capacity of the 0.5% solution of sodium hypochlorite was considerably enhanced by a continuous exchange of fresh solution. This finding is in agreement with Dakin's method of constant flooding with a fresh solution (2) that was found to be most efficient in the treatment of infected wounds. It has also been demonstrated that mechanical agitation as well as fluid flow will enhance the dissolving capacity of sodium hypochlorite (7). Thus, the mechanical instrumentation of root canals appears to be a factor that can increase the dissolving effect of sodium hypochlorite. It has been stated that an increased degree of solvent action can be achieved by higher concentrations, and solutions up to 5.25% have been recommended for endodontic irrigation (13–15). However, studies including the cytotoxicity and antibacterial effect of various concentrations have demonstrated that the use of a 0.5% solution of sodium hypochlorite implies the least interference with vital tissue (16–20). When the tissue pieces were transferred to isotonic saline, a slight swelling in combination with a weight gain was noted. This in agreement with Hand et al. (13), who have demonstrated an uptake of liquid by necrotic tissue exposed to isotonic saline or water.

The effect of calcium hydroxide on vital pulp tissue has been widely studied, whereas its effect on necrotic tissue has not been investigated previously. The copious swelling of the tissue that took place in the mixture of calcium hydroxide and water resulted in a weight increase more than twice the original weight. Conceivably, a denaturation and hydrolyzation of the tissue by the hydroxide (12) leads to a destruction

TABLE 1. **Time values for dissolution of 0.02 g of porcine muscular tissue during a 12-day control period**

Group	Treatment Procedure	Time for Dissolution
1	Calcium hydroxide	12 days
2	0.5% Sodium hypochlorite	Not dissolved
3	0.5% Sodium hypochlorite fresh every 30 min	180 min
4	Calcium hydroxide, 30 min and 0.5% sodium hypochlorite	90 min
5	Calcium hydroxide, 24 h and 0.5% sodium hypochlorite	60 min
6	Calcium hydroxide, 7 days and 0.5% sodium hypochlorite	60 min
7	Isotonic saline	Not dissolved

of the tissue architecture in combination with a splitting of protein molecules, thus making the tissue more accessible to the sodium hypochlorite. This may explain the faster dissolution of tissue in sodium hypochlorite after pretreatment with calcium hydroxide. Also, the finding in this study that calcium hydroxide per se was able to completely disintegrate necrotic tissue may be of clinical value.

The tissue pieces used in this study were completely surrounded by sodium hypochlorite or calcium hydroxide and probably more accessible than tissue remnants in a root canal. However, Heide and Kerekes (11) have found that the use of calcium hydroxide as a long-term intracanal dressing gives clean root canals. It could be speculated that the calcium hydroxide may not only disrupt the structure within the tissue, but in the root canal it may also separate the tissue from the dentinal walls. Previously, it has been shown that teeth treated with an intracanal dressing of calcium hydroxide show no growth after bacterial sampling (21). Conceivably, calcium hydroxide, besides from its antibacterial effect, can dissolve tissue remnants that otherwise would have become substrate for bacteria.

CONCLUSIONS

In this study, using necrotic porcine muscle tissue, it was found that:

1. Calcium hydroxide was capable of dissolving tissue.
2. The tissue dissolving effect of sodium hypochlorite was enhanced by pretreatment with calcium hydroxide.

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