Chemical Analysis of the Formation of Calcium Carbonate and its Influence on Calcium Hydroxide Pastes in Connective Tissue of the Dog - Part II

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The objective of this research was to chemically analyze calcium hydroxide pastes added to three hydrosoluble vehicles having different acid-base characteristics by implanting polyethylene tubes in subcutaneous connective tissue in dogs, evaluating the formation of calcium carbonate over a period of 7, 30, 45, and 60 days. The three vehicles were saline, anesthetic, and polyethylene glycol 400. Determination of the formation of calcium carbonate was evaluated by volumetry of neutralization using hydrochloric acid for titration.

Key Words: calcium hydroxide, intracanal dressing, calcium carbonate.

Introduction

The chemical dynamics of calcium hydroxide as demonstrated by ionic disassociation characterize its properties. The inhibiting effect of calcium hydroxide on bacterial enzymes which leads to its antibacterial property and its activating effect on tissue enzymes such as alkaline phosphatase producing its mineralizing characteristic illustrate the biological qualities of hydroxyl and calcium ions on both bacteria and tissue. These effects show that its enzymatic properties are due to its elevated pH (Estrela et al., 1994, 1995).

Calcium hydroxide is obtained from the calcination of calcium carbonate until its transformation into calcium oxide. When calcium oxide is hydrated, the reaction between calcium hydroxide and carbon dioxide leads to the formation of calcium carbonate (Greenwood and Earnshaw, 1984).

The properties of calcium hydroxide resulting from ionic liberation are directly influenced by carbon dioxide which, by forming a weak oxide acid, could provoke partial neutralization of the dressing which is basic. The resulting calcium carbonate does not have the same biological properties as does calcium hydroxide.

Some studies have reported concern about the effect of carbon dioxide from atmospheric air on the quality of calcium hydroxide (Maisto and Capurro, 1964; Seltzer and Bender, 1979; Holland et al., 1979, 1982; Cohen and Lasfargues 1988; Pertot et al., 1992). The need for research that evaluates the influence of tissue carbon dioxide on calcium hydroxide pastes has motivated this study. The objective was to verify the chemical dynamics that occur in the ionic disassociation of calcium hydroxide pastes with respect to the formation of calcium carbonate when used in different vehicles in the connective tissue of the dog.

Material and Methods

Twenty-four polyethylene tubes (number 10, Levine) were used in this study. The tubes were 60 mm long and had internal diameters of 3 mm, one of the ends of each tube being sealed by heat and compression.

The tubes were divided into 4 groups of 6 each. Each group of six tubes was further subdivided into three groups of two each. The tubes were then completely filled with 1.2 g of calcium hydroxide paste (Quimis, USA) with the following vehicles: saline solution, anesthetic solution (2% xylocaine with vasoconstrictor) and polyethylene glycol 400. The pastes were prepared to the same viscosity, 3501 centi-Poise at 0.1 rpm, as determined by a Brookfield digital rheometer (cone-plate type), the resulting pastes being compared clinically to the consistency of standard toothpaste.

The four groups of tubes were implanted into the subcutaneous tissue of a two-year-old dog of undetermined breed, two groups on either side of the thorax, separated by 10 cm. The dog was pre-anesthetized iv with 0.1 mg/kg acepromazine (0.2% Acepram) and, after 15 minutes, anesthetized with 3% sodium thiopental (Thionembutal), applied by an oral-tracheal catheter. Anesthesia was maintained with 2% halothane (Fluothane).

After periods of 7, 30, 45, and 60 days the tubes were removed in groups of six, being carefully transported for chemical analysis in completely sealed inert flasks containing nitrogen atmosphere.

Chemical analysis was done using the volumetric method of neutralization. Hydrochloric acid was used as the titrate, the final point of the reaction being observed visually by the use of methyl orange and phenolphthalein indicators (Basset et al., 1981). For each observation period, 7, 30, 45, and 60 days, a sample was removed from each pair of tubes according to the paste vehicle used, and transferred with quantitative care to a liter of distilled water in which it was dissolved. Of this solution, 25 ml was titrated with hydrochloric acid at a concentration of 0.0109 mols per liter using a phenolphthalein indicator. Another 25 ml of the same solution was titrated with the same acid, using a methyl orange indicator. The titrations were done in cold solutions to avoid as much as possible losses of carbon dioxide. Each sample was titrated three times for each indicator used.

We will first consider the volume of acid used in titration with the phenolphthalein indicator and call it v. We note at this point that all of the hydroxide will be neutralized and the carbonate converted to hydrogen carbonate. Let us now consider the volume of acid used in titration with the methyl orange indicator. This volume will be called V. Once again there is complete neutralization, this time of all the alkali present. If we subtract the volume v of the acid used in first titration from the volume V of the acid of the second titration we arrive at the volume of acid necessary to neutralize half of the carbonate. The volume of acid necessary to neutralize all of the carbonate is thus 2(V-v).

It is now possible to calculate the carbonate which is formed. At the point of equivalence the number of mols n of a solution can be calculated multiplying the concentration in mols per liter by the volume. In the present case we have $nCO32- = 2 \times MH+ \times VC$. Because all of this carbonate is present in the 25 ml withdrawn from the solution, the concentration in mols per liter (CM) corresponds to $CM = 40 \times nCO32-$. Since the concentration of the acid is standardized and corresponds to 0.0109 mols per liter, $CM = 0.872 \times VC$.

To convert this concentration in grams per liter we must remember that the molecular weight of CaCO3 is 216.88. Therefore, every mole of CaCO3 corresponds to 216.88 g. The concentration in grams per liter of carbonate is thus calculated by the formula: $C (g/l) = 189.12 VC$.

Results

Table 1 shows the formation of carbonate ions (calculated) in grams per liter.

Discussion and Conclusions

The antibacterial and mineralizing capacity of calcium hydroxide is due to its ionic disassociation and is directly related to its elevated pH. The effect of pH on enzymatic activity of anaerobic bacteria has permitted Estrela et al. (1994) to present the hypothesis that calcium hydroxide produces a definite and irreversible

bacterial enzymatic inactivation at extreme conditions of pH and a temporary or reversible inactivation when there is a return of the ideal pH for enzymatic action.

Hydroxyl ions can alter the metabolism of bacteria enzymes through the pH gradient which exists in the cytoplasmic membrane because the velocity and intense ionic action leads to a lesive and toxic effect (Estrela et al., 1995). Several studies have shown the antibacterial effect of calcium hydroxide on different respiratory types of bacteria (Bystron et al., 1985; Safavi and Nichols, 1993, 1994; Estrela et al., 1996).

Since the vehicles associated with calcium hydroxide present acid/base and/or hydrosoluble/oily characteristics, they all exert certain influences on the velocity of ionic disassociation. Estrela and Pesce (1996) have chemically analyzed the liberation of calcium and hydroxyl ions of calcium hydroxide pastes with vehicles of different acid/base and hydrosolubility characteristics by conductimeter analysis of their solutions. The liberation of hydroxyl ions from the aforementioned pastes can be demonstrated by the liberation of calcium ions, using as a parameter the direct relation between calcium hydroxide and hydroxyl ions and the molecular weight of calcium hydroxide. In calcium hydroxide the proportion of hydroxyl ions to calcium ions is 45.89 to 54.11. The calculated percentages of liberation of hydroxyl ions are thus 84.80% of the measured percentages of the liberation of calcium ions.

However, calcium hydroxide in contact with carbon dioxide also undergoes chemical change, being transformed into calcium carbonate. Some researchers have studied the loss in quality of the product as a result of its carbonation due to carbon dioxide gas in the atmosphere.

The results obtained from this study show that when saline vehicles are employed in calcium hydroxide pastes the rate of formation of calcium carbonate is practically unaltered after 30 days over a period of 60 days when there are no exchanges of paste under the same conditions of inflammation. When polyethylene glycol 400 was used as vehicle for calcium hydroxide paste, this was also observed after 45 days. The rate of calcium carbonate formation was also lower than the other two pastes during the initial 30-day period. The rate of calcium carbonate formation in the anesthetic-based paste was higher than the other two in the initial 7-day period; the rate remained relatively the same over the entire two-month period.

From these results, it can be deduced that, after the initial reaction of calcium hydroxide on the tissue, there are strong reasons to reduce the number of changes of the paste during its use as an intracanal dressing. This is especially true after the initial inflammatory symptoms have been removed.

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