pH and calcium ion release evaluation of pure and calcium hydroxide-containing Epiphany for use in retrograde filling

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ABSTRACT

Objective: Hydroxyl (OH^-) and calcium (Ca^{++}) ion release was evaluated in six materials: G1) Sealer 26, G2) White mineral trioxide aggregate (MTA), G3) Epiphany, G4) Epiphany + 10% calcium hydroxide (CH), G5) Epiphany + 20% CH, and G6) zinc oxide and eugenol. Material and Methods: Specimens were placed in polyethylene tubes and immersed in distilled water. After 3, 6, 12, 24, and 48 h, 7, 14, and 28 days, the water was assessed for pH with a pH meter and for Ca^{++} release by atomic absorption spectrophotometry. Results: G1, G2, G4, and G5 had the highest pH until 14 days ($p<0.05$). G1 presented the highest Ca^{++} release until 6 h, and G4 and G5, from 12 h through 14 days. Ca^{++} release was greater for G1 and G2 at 28 days. G6 released the least Ca^{++}. Conclusions: MTA, Sealer 26, Epiphany, and Epiphany + CH release OH^- and Ca^{++} ions. Epiphany + CH may be an alternative as retrofilling material.

Key words: Retrograde obturation. Hydrogen-ion concentration. Calcium hydroxide.

INTRODUCTION

Retrograde filling consists in the preparation and filling of an apical cavity with an appropriate material[14]. In the search for a biocompatible material capable of inducing mineralized apical tissue formation, calcium hydroxide (CH) has been included in the formulation of filling materials[5,11,19]. In order to stimulate mineralized tissue formation, the filling material should be able to increase the pH and release of calcium ions (Ca^{++})[9,12,13].

Sealer 26 is an epoxy resin-based cement containing CH that can be used for retrograde filling if prepared with a higher powder/resin ratio than when used as a root canal sealer[21,23,24]. Tanomaru-Filho, et al.[22] (2006) studied Sealer 26 (Dentsply, Ind. e Com. Ltda., Petrópolis, RJ, Brasil) in retrograde fillings in dogs' teeth and observed similar periapical repair with Sealer 26, ProRoot MTA (Tulsa Dental, Johnson, TN, USA), and Sealapex (Kerr Corp., Orange, CA, USA) with zinc oxide. Some studies have shown lower rates of pH increase and Ca^{++} release for Sealer 26 compared to other CH-containing cements[4,19]. However, in these studies, Sealer 26 was prepared using the powder/resin ratio recommended for its use as a root canal sealer.

MTA a retrograde filling material composed of tricalcium silicate, tricalcium aluminate, and other mineral trioxides, shows adequate sealing ability in retrograde fillings[27,29] and in the treatment of root perforations[15,17]. This material is also biocompatible when used in retrograde filling[23,28] and root canal perforations[30]. MTA promotes alkaline pH[6,18] and has a mechanism of action similar to CH[12].

Epiphany is a resin cement component of the Epiphany/Resilon system. Its presence allows greater adhesion between the endodontic filling materials and the root canal walls[7,26]. This cement has been evaluated for use in retrograde fillings. Maltezos, et al.[16] (2006) evaluated the seal promoted by the Resilon/Epiphany system, Pro Root MTA, and Super-EBA as retrograde filling materials, and observed less bacterial leakage for Resilon and MTA compared with Super-EBA. Those authors concluded that Resilon is a viable option as retrograde filling material.

Duarte, et al.[5] (2004) proposed the addition of CH to AH Plus cement in order to reduce its flowability. We suggest that CH can be added to Epiphany to attain thicker consistency, facilitating its use as a retrograde filling material. The addition of CH can alter the release of OH^- and Ca^{++} ions. Therefore, the present study aimed to evaluate the pH increase and calcium ion release of Epiphany cement and the Epiphany-CH association, compared to other cements used in retrograde filling.

MATERIAL AND METHODS

The following materials, containing either CH or calcium oxide, were evaluated (Figure 1).

Preparation of specimens

For pH and calcium ion release evaluation, 10 specimens were prepared from each material studied. Sixty polyethylene tubes measuring 1 cm in length and 1.5 mm in diameter were filled with the cements to be evaluated.

Sealer 26 (G1) was prepared in a 5:1 powder/resin ratio, according to Tanomaru-Filho, et al.[22] (2006). White MTA (G2) was prepared in a powder/liquid ratio of 0.33 g/1g[10]. Pure Epiphany (G3) was prepared using equal measurements of both pastes. After weighing the two paste components, 10% or 20% CH (CH) was added to the Epiphany sealer, resulting in the materials derived from Epiphany evaluated in G4 and G5, respectively. Zinc oxide and eugenol cement was prepared in a powder/liquid ratio according to Bernabé, et al.[2] (2005).

Immediately after preparing the materials and filling the tubes they were placed in lidded flasks (JProlab, São José dos Pinhais, PR, Brazil) containing 10 mL distilled water with neutral pH and kept in an oven at 37°C. After 2, 6, 12, 24, and 48 h, 7, 14, and 28 days, the water was assessed for pH and calcium ion release. The tubes containing the cements were placed in new flasks with 10 mL of distilled water for further analyses in the different time periods.

Analyses of pH and Calcium Release

pH readings

pH readings were performed with an UltraBasic pH Meter (Denver Instrument Company, Arvada, CO, USA), previously calibrated using substances with pHs 4, 7, and 10.

Ca^{++} Ion Release Readings

Ca^{++} release was monitored with an H1170 Hilger & Watts Atomspeck atomic absorption spectrophotometry (Rank Precision Industries Ltd. Analytical Division, London, UK) equipped with a calcium atom-specific hollow cathode lamp (wavelength =422.7 nm and current =20 mA), 0.7 nm slit and 5.0 cm burner length.

Ca^{++} ion release readings were compared to a standard curve obtained by diluting calcium at 1.0, 2.0, 3.0, 4.0, 5.0, 6.0, 7.0, 8.0, 9.0, 10.0, 15.0, 20.0, and 25.0 ppm (Calcium Standard Solution, Merck, Darmstadt, Germany) in ultra-pure water.

Statistical Analysis

Data were subjected to statistical analysis by ANOVA and individual comparisons were performed using the Tukey-Kramer method. A significance level of 5% was set for all analyses.

Material	Composition	Manufacturer
Sealer 26	Calcium hydroxide, bismuth oxide, tetramine hexamethylene, titanium dioxide, bisphenol epoxy resin.	Dentsply, Rio de Janeiro, RJ, Brazil
White MTA	Silicon dioxide , potassium oxide, aluminum oxide, sodium oxide, iron oxide, sulfur trioxide, calcium oxide, bismuth oxide, magnesium oxide, insoluble residues of calcium oxide, potassium and sodium per sulphates and crystalline silica.	Angelus, Soluções em Odontologia, Londrina, PR, Brazil
Epiphany	Bisphenol-A-glycidyl dimethacrylate, polyethylene glycol dimethacrylate, ethoxylated bisphenol-A dimethacrylate, urethane dimethacrylate, barium sulphate, silica, calcium oxide, bismuth, pigments.	Pentron Clinical Technologies, LLC., Wallingford, CT, EUA
Zinc oxide and eugenol	Powder: zinc oxide Liquid: eugenol	Dentsply, Rio de Janeiro, RJ, Brazil

Figure 1- Materials used in the study, their compositions and manufacturers

Table 1- Mean ± standard deviation of hydroxide ions (pH) released from the materials studied after different periods of time

	Sealer 26	White MTA	Epiphany	Epiphany + 10% CH	Epiphany + 20% CH	Endo Fill
3 h	10.47±0.18 (A)	9.918±0.22 (BC)	9.044±0.17 (E)	9.774±0.29 (CD)	10.09±0.1 (B)	7.286±0.28 (F)
6 h	9.617±0.22 (A)	9.139±0.24 (B)	8.184±0.13 (C)	9.162±0.17 (B)	9.522±0.09 (A)	7.609±0.31 (D)
12 h	9.120±0.14 (A)	8.745±0.14 (B)	7.527±0.15 (D)	8.009±0.13 (C)	9.372±0.35 (A)	7.232±0.17 (E)
24 h	8.425±0.1 (B)	8.628±0.19 (A)	7.116±0.1 (E)	7.651±0.09 (D)	7.887±0.06 (C)	7.043±0.19 (E)
48 h	8.185±0.16 (B)	8.555±0.1 (A)	7.455±0.15 (D)	7.600±0.27 (CD)	7.741±0.16 (C)	7.377±0.21 (D)
7 d	8.173±0.1 (A)	8.198±0.03 (A)	7.544±0.04 (C)	7.741±0.06 (B)	7.785±0.05 (B)	7.359±0.22 (D)
14 d	8.258±0.11 (A)	8.213±0.1 (A)	7.580±0.16 (C)	7.922±0.09 (B)	7.896±0.04 (B)	7.090±0.33 (D)
28 d	7.800±0.1 (AB)	7.845±0.22 (AB)	7.709±0.17 (B)	7.875±0.12 (AB)	7.976±0.13 (A)	7.270±0.18 (C)

(A,B,C,D) In each period, means values followed by different labels are statistically different ($p<0.05$).

Table 2- Mean ± standard deviation of calcium (mg%) released from the materials studied after different periods of time

	Sealer 26	White MTA	Epiphany	Epiphany + 10% CH	Epiphany + 20% CH	Endo Fill
3 h	0.118±0.13 (A)	0 (B)	0 (B)	0 (B)	0 (B)	0 (B)
6 h	1.513±1.13 (A)	0.782±0.56 (B)	0.487±0.02 (BC)	0.855±0.18 (AB)	1.041±0.04 (AB)	0.219±0.03 (C)
12 h	0.249±0.08 (BC)	0.102±0.06 (C)	0.021±0.02 (C)	0.381±0.33 (B)	0.573±0.05 (A)	0 (C)
24 h	0.730±0.07 (C)	0.777±0.13 (C)	0.852±0.5 (C)	1.21±0.36 (B)	1.706±0.09 (A)	0.145±0.04 (D)
48 h	0.389±0.19 (C)	0.494±0.09 (C)	1.293±0.34 (B)	1.426±0.10 (B)	2.081±0.68 (A)	0.052±0.11 (CD)
7 d	0.170±0.09 (D)	0.519±0.06 (C)	1.615±0.16 (B)	1.774±0.15 (B)	2.581±0.33 (A)	0.007±0.02 (D)
14 d	0.229±0.05 (B)	0.303±0.07 (B)	0.06±0.04 (C)	0.43±0.14 (A)	0.520±0.08 (A)	0 (C)
28 d	1.575±0.32 (A)	1.06±0.21 (B)	0 (D)	0.268±0.05 (C)	0.424±0.06 (C)	0 (D)

(A,B,C,D) In each period, means values followed by different labels are statistically different ($p<0.05$).

RESULTS

The mean pH values of the cements evaluated in the various experimental periods are shown in Table 1. Sealer 26 presented the highest pH value in the initial periods (3 and 6 h). After 12 h, Sealer 26 and Epiphany + 20% CH had equivalent results. From 7 through 14 days, White MTA and Sealer 26 had the highest pH values, followed by Epiphany with 10% and 20% CH. At 28 days, all materials had similar results, except for zinc oxide and eugenol (ZOE). In all the experimental periods ZOE had the lowest pH values ($p<0.05$).

The mean Ca^{++} ion release values for all cements evaluated are shown in Table 2. Ca^{++} release at 3 and 6 h was greater for Sealer 26. From 12 h through 14 days, Epiphany with 10% and 20% CH had the highest Ca^{++} release. After 28 days, Sealer 26 and White MTA presented the highest Ca^{++} release. In all experimental periods, ZOE had the lowest Ca^{++} release values.

DISCUSSION

The methodology we followed consists in filling standardized tubes with the materials to be tested and immersing in distilled water. The pH is then determined in the resulting solution[1]. Numerous authors have utilized similar methodology, immersing the plastic tubes containing cements in distilled, deionized, or Milli-q water. Duarte, et al.[6] (2003) used tubes measuring 10 mm in length and 1.5 mm in internal diameter. Santos, et al.[18] (2005) evaluated pH and calcium release using MTA and an experimental dental cement placed in tubes measuring 10 mm in length and 1 mm in diameter. Distilled water was used due to its purity and neutral pH[8].

Although several CH-based sealers are available in the market, the presence of CH in the composition of an endodontic sealer does not assure release of calcium and OH^- ions in the final product. After setting of the cement, the ions may not be released[19], or the CH may be inactivated by other components in the sealer.

The endodontic materials Sealer 26 and Epiphany contain CH or calcium oxide in their formulation. MTA includes calcium oxide in its composition. During its setting, hydration reactions take place, resulting in production of CH. When the set material is placed in a solution, it dissociates into OH^- and Ca^{++} ions, increasing both the pH and the calcium concentrations in the medium[20].

Regarding the pH of the cements evaluated, during the first 12 h, Sealer 26 and Epiphany with 20% CH showed the highest values among all materials studied. This may be due to the presence of 37% CH in the composition of Sealer 26, and due to its longer setting time. Epiphany cement, which contains calcium oxide, had 20% CH incorporated into its formulation. Similar results were reported by Duarte, et al.[4] (2000), who observed higher pH values for Sealer 26 in the initial experimental periods, when using the material in the powder/resin ratio recommended for it use as a root canal sealer. MTA had initial pH of 9.01; 3 h after mixing its pH was 8.28 and remained constant thereafter. These results are in accordance with those obtained by Duarte, et al.[6] (2003).

Tay, et al.[25] (2007) described the calcium phosphate phases produced after immersion of set Portland cement in phosphate buffered saline solution. The authors observed that the pH changes occurred in two stages, with release of OH^- ions during the precipitation of calcium phosphate. Moreover, these authors concluded that the biological action of MTA may be partially attributed to the ability to induce mineralized tissue formation by the components of Portland cement.

From 12 h through 14 days after mixing, Epiphany cement with 20% and 10% CH and pure Epiphany showed the greatest levels of calcium ion release, which may be related to the material's properties of water sorption and solubility. Donnelly, et al.[3] (2007) reported a higher degree of water sorption and solubility for Epiphany compared to Ketac-Endo, InnoEndo, EndoREZ, Kerr EWT, AH Plus, and GuttaFlow cements. These properties may have contributed to the high values observed for Ca^{++} release in the longer experimental periods.

Duarte, et al.[5] (2004), suggested the addition of CH to AH Plus cement in order to decrease its flowability. In the present study, addition of CH to Epiphany promoted better consistency for its use in retrograde fillings, accompanied by greater release of OH^- and Ca^{++} ions. Further studies are necessary for a more detailed evaluation of the properties of this material, including its biological effects.

CONCLUSIONS

Based on the methodology used in this study, it may be concluded that Sealer 26, White MTA, Epiphany, and Epiphany with CH presented Ca^{++} and OH^- ion release. Addition of CH to Epiphany may be an alternative as retrofilling material.

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