

In Vitro Evaluation of a Calcium Phosphate Cement Root Canal Filler/Sealer

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An in vitro dye leakage study was performed to compare the apical leakage of a fill with injectable calcium phosphate cement (CPC) filler/sealer and a master silver cone with leakage from a fill of Sealapex sealer and laterally condensed gutta-percha. Ten instrumented, extracted, single-rooted human teeth were obturated with either laterally condensed gutta-percha and Sealapex as the sealer or with a single master cone and the CPC paste sealer. Additional teeth were included in the study to serve as controls. The teeth were placed in 1% poly-R dye solution (pH 7.0) for 5 days. After the teeth were longitudinally sectioned apical leakage of dye was measured. There were no significant differences between the CPC and Sealapex groups. The single cone CPC procedure provided an adequate apical seal against dye penetration. Should retreatment become necessary the single cone may be removed to provide access for instrumentation.

A calcium phosphate cement (CPC) (1) comprised of equimolar amounts of tetracalcium phosphate (TTCP) and dicalcium phosphate anhydrous (DCPA) was shown to be biocompatible (2, 3) and osteoconductive (4). Clinical studies (5–9) have demonstrated that CPC is efficacious in repairing bone defects in humans. In vitro studies (10) and animal models (11, 12) have indicated that it is also useful in endodontics as a filler/sealer in root canal treatment. In addition to being used as a sealer/filler in vitro studies have shown that CPC can also seal a furcation perforation (13) and could be used as an apical barrier for apexification (14). These results suggest that CPC has potential to promote the healing of bone in endodontic treatment.

Recently the use of an injectable filler/sealer in root canal treatment has gained more acceptance because of its convenience and flexibility. Injectable CPC filler/sealer has several distinctive advantages in addition to its biocompatibility that eliminate the problem of filler/sealer extrusions. CPC adapts closely to root canal walls and sets to a hard mass. Unlike other sealers (15) moisture in the canal will not decrease the strength of CPC; the

mechanical strength of CPC actually increased when placed in saliva or plasma-like fluids (16).

Because of its rigidity one concern in using CPC for endodontic applications is the problem of retreatment. Glass ionomer cement shares the same problem, which was resolved with the insertion of a gutta-percha master cone to create a path for retreatment if required (17). The objective of this study was to determine the sealing ability of CPC when used as filler/sealer in conjunction with a master cone. Because the consistency of CPC was too stiff for the complete insertion of a master gutta-percha cone, a silver point was used instead. The rationale was that if adequate sealing was obtained, this procedure has the potential to be clinically useful because the silver point could be withdrawn to create a path for filler removal in case retreatment becomes necessary.

MATERIALS AND METHODS

Fifty grams of TTCP was ground in a 250 ml agate jar with 120 agate balls (10 mm diameter) and 60 g of cyclohexane for 24 h in a ball mill (Retsch PM4, Brinkman, NY). After cyclohexane was completely evaporated TTCP was sifted through a fabric sieve with 5 μ m openings (Spectrum/mesh, Houston, TX). Commercially obtained DCPA was ground in ethanol (volume fraction = 95%) in the agate jar with 120 agate balls for 24 h to obtain a median particle size of 1.0 μ m. A CPC mixture that contained equimolar amounts of the ground TTCP (mass fraction = 73%) and DCPA (mass fraction = 27%) was prepared. The powder component of the experimental CPC filler was prepared by combining 80% of this mixture and 20% barium sulfate to produce the necessary radiopacity. Each 10 g of the liquid contained 3.3 g of glycerin and 6.7 g of a 0.25 mol/L aqueous sodium phosphate solution that also contained 0.03 g of carboxymethyl cellulose (CMC) (Sigma, St. Louis, MO). Glycerin served as a lubricant that aided the injectibility of the filler paste (10). Sodium phosphate was used to make the cement harden more rapidly, and CMC made the cement more cohesive and resistant to washout (18). The CPC root canal filler paste was prepared by combining 2.65 g of the powder with 1 g of the cement liquid.

Twenty-four, single-rooted human anterior teeth were used. A #15 file was introduced into the canal, and the working length for each root was established at 1 mm short of the apical foramen. Sequential files, beginning at size 15, were inserted into the apical portion of the canal using a reaming motion at the predetermined working length. Flaring of the coronal portion of the canal was

accomplished with a step-back flaring technique. Canals were irrigated with 5.25% NaOCl for each file. To maintain patency of the apical foramen, a #15 file was passed through the apical foramen after the use of each file size. The teeth were stored in 0.1% sodium azide solution until obturation.

Twenty-four instrumented teeth were divided into four groups: positive control, negative control, conventional gutta-percha obturation group, and CPC experimental group. Four teeth were obturated with laterally condensed gutta-percha without sealer. Two of these were used as a positive control, and the other two were completely covered with two coats of nail polish and served as a negative control. The remaining 20 teeth were randomly divided into two groups of 10 each. In the gutta-percha group root canals were coated with Sealapex (Kerr, Romulus, MI) applied by a lentulo spiral and then laterally condensed with gutta-percha. In the CPC experimental group the teeth were obturated with injectable CPC canal filler paste with a syringe, and a single master silver point was then inserted. After obturation the excess gutta-percha was burned off in the laterally condensed group and the excess silver point was removed with a bur in the CPC group. Crowns were sealed with sticky wax. The teeth were then placed in plastic tubes containing tissue paper moistened with a saliva-like solution, which contained 1.2 mmol/L CaCl_2 , 0.72 mmol/L K_2HPO_4 , 30 mmol/L KCl, and 50 mmol/L HEPES at pH 7.4, and incubated at 37°C in a 100% humidity incubator for 3 days. The teeth were then immersed in the saliva-like solution for another 3 days to allow the specimens and the filler/sealer materials to be in equilibrium with the aqueous environment.

Apical leakage was estimated by a dye penetration test. The apical one-third of each root was immersed in pH-neutral 1% poly-R dye (Sigma) solution for 5 days in a 37°C, 100% relative humidity chamber. The teeth were then removed from the dye solution, rinsed with water, and longitudinally cut into halves with a 4-inch diameter diamond wafering blade (Buehler, Waukegan, Lake Buff, IL) under running water. Each section was then examined under a stereomicroscope at $\times 25$ magnification. Apical leakage was measured to the nearest 0.01 mm from the apex to the most coronal extent of dye penetration. A *t* test was used to determine whether there was a significant difference between the Sealapex–gutta-percha and CPC groups at a level of significance of 5%.

RESULTS

The two negative controls did not show any dye penetration, whereas the two positive controls revealed full dye penetration. The mean (SD; $n = 10$) distances of poly-R dye penetration in the Sealapex–gutta-percha and CPC–silver point groups were, respectively, 0.84 mm (0.65 mm) and 0.75 mm (0.67 mm), which were not significantly different ($p > 0.05$).

DISCUSSION

Methylene blue has been most commonly used in leakage tests, but it is a very small molecule (relative molecular weight = 357) and, therefore, readily penetrates the root tissue. Consequently the external root surfaces must be coated to prevent dye penetration and to remain dry during the experimental procedures, and this condition is less clinically relevant (19). Poly-R was selected for the dye penetration test because it

has a particle size of $\sim 0.02 \mu\text{m}$, which is smaller than oral bacteria ($> 0.3 \mu\text{m}$). Poly-R does not penetrate into root tissue and, as a result, the root surfaces of the specimens need not be coated for the dye penetration test. Thus, the present in vitro experimental conditions mimic the in vivo situation in that the roots remained moist and the canal filling materials were in contact (through dentin tubules) with the aqueous environment in each step of the treatment and test procedures.

After the filling procedure the obturated specimens were incubated in a 37°C, 100% humidity chamber to allow the CPC paste to harden. This step, although not included in most root canal sealing tests in the literature (20), is consistent with the conditions in the oral environment.

Variances in the dye penetration results reported in this study, although relatively large, are comparable with those reported in previous studies (20). The factors that contributed to the large variances are unknown. Differences, however, in the morphology of the root canals, which could affect adaptation of the sealer/filler to the canal walls, may have been a factor. Nevertheless in this study the variance of CPC group was similar to that of the gutta-percha group.

The diametral tensile strength of the hardened CPC used in this study was 2.4 MPa, which is considerably lower than the typical value of 9 to 12 MPa reported in the literature (18). This is probably because in the present study we used a CPC powder with smaller TTCP particles ($< 5 \mu\text{m}$ compared with $17 \mu\text{m}$) and a lower powder-to-liquid ratio (2.65 compared with 4.0). The present results, however, would suggest that CPC with this lower strength was adequate for use as filler/sealer.

The results of the present study showed that satisfactory apical sealing was obtained with the use of a single master point and CPC filler/sealer in root canal treatment. Thus the procedure met an important requirement for the endodontic treatment. Removal of the master silver point would provide an access opening for retreatment. The CPC filler/sealer was found to be softer and less tough than dentin and can be readily removed with endfiles from the straight canals used in the present study.

Recent preliminary test results showed that a CPC mixture that contains TTCP particles with a narrower range of particle sizes (between 5 and $13 \mu\text{m}$) was sufficiently soft to allow insertion of a gutta-percha master cone in straight canals. The sealing ability of this material has yet to be tested. Future improvements in the properties of CPC and in master cone design and materials should make this procedure clinically useful.

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Certain commercial equipment, instruments, or materials are identified in this article to foster understanding. Such identification does not imply recommendation or endorsement by the National Institute of Standards and Technology or the ADAHF, nor does it imply that the materials or equipment identified are necessarily the best available for the purpose.

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