Perforation Repair Comparing Two Types of Mineral Trioxide Aggregate

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The purpose of this study was to evaluate the ability of two types of mineral trioxide aggregate (MTA) to seal furcal perforations in extracted human molars using an anaerobic bacterial leakage model. Forty, human, maxillary and mandibular molars were randomly divided into two experimental groups of 18, with two teeth used as positive controls and two teeth without perforations used as negative controls. Experimental group 1 was repaired with gray-colored MTA and group 2 with off-white-colored MTA. A dual-chamber, anaerobic, bacterial-leakage model was assembled. Brain-heart infusion broth with yeast extract, hemin, and menadione was used as the culture broth for Fusobacterium nucleatum. Two of 18 gray-colored MTA samples leaked and three off-white-colored MTA samples leaked. There was no significant difference between the two types of MTA in preventing leakage of F. nucleatum past furcal perforation repairs.

Perforations are procedural accidents that can have an adverse affect on the outcome of endodontic treatment. Sinai (1) found that the prognosis for a tooth with a perforation depends on the location of the perforation, the time the perforation is open to contamination, the possibility of sealing the perforation, and accessibility of the main canal. Seltzer (2) stressed the importance of sealing perforations immediately in studies on monkeys. He demonstrated that unsealed perforations are exposed to microbial contamination, resulting in further damage to the periodontium and ultimately destruction of bone and downward epithelial migration.

In a review article, Alhadainy (3) discussed the ideal properties of a perforation repair material: non-toxic, biocompatible, excellent sealing qualities, nonresorbable, radiopaque, and bacteriostatic. Balla et al. (4) correlated inflammation in the furcation area with the inadequate sealing ability or cytotoxicity of the repair material. Additionally, the repair material should be esthetically pleasing. Many materials have been suggested for use in furcation perforation repairs including glass ionomer, calcium hydroxide, Super-EBA cement, amalgam, tricalcium phosphate, compositebonded restorations, decalcified freeze-dried bone, and mineral trioxide aggregate (MTA) (3-10).

Different leakage models have been used in the past to assess the ability of materials to seal furcation perforations. These include the fluid-filtration model (6), dye-leakage model (10), and bacterial-leakage model (11). Weldon et al. (6) used a fluid-filtration technique to assess the ability of MTA, Super-EBA, and a combination of MTA and Super-EBA to seal furcation perforations and found no significant differences at 1 month. Daoudi and Saunders (10) studied the use of MTA and resin-modified, glass-ionomer cement to seal furcation perforations using a dye-leakage model. The authors found that perforations repaired with MTA leaked significantly less to the dye tracer than Vitrebond (10). Nakata et al. (11) used an anaerobic, bacterial-leakage model to assess the sealing ability of MTA and amalgam. The investigators found that teeth with furcation perforations repaired using MTA allowed the passage of Fusobacterium nucleatum significantly less than teeth repaired with amalgam.

To date, no controlled, clinical trials have been published documenting the use of MTA as a material suitable to repair furcation perforations. However, two case reports have been published demonstrating that MTA may be suitable for closing the communication between the pulp chamber and the underlying periodontal tissues (12). Thus, MTA seems to be an acceptable material to use in the sealing of perforations.

When using gray MTA, one is instructed to limit the material to the confines of the canal and/or pulp chamber area, not above the crestal bone level, because the material by nature of its component parts may lead to discoloration (13). Recently, a new type of MTA has become available as an off-white powder. Presumably, a benefit of this new type is its application in esthetically sensitive areas. The purpose of this study was to compare the ability of two types of MTA for sealing furcation perforations in extracted human molars using an anaerobic, bacterial-leakage model.

MATERIALS AND METHODS

Forty, extracted, human, maxillary and mandibular molars were collected following the protocol of the Oregon Health & Science University institutional review board. The molars had minimal restorations or caries and roots that were not fused. All teeth were stored in 0.2% thymol in physiologic saline. The occlusal surface of the crowns and the apical 5 mm of the roots were removed using high-speed tapered diamonds. A standardized 5-mm \times 5-mm

endodontic access opening was made in each tooth with high-speed carbide burs to hold the tip of a 5-ml irrigation syringe (Becton Dickinson & Co., Franklin Lakes, NJ). Pulp tissue and debris were removed and the pulp cavity irrigated with sterile saline. Cyanoacrylate cement (Krazy® Glue, Elmer's Products, Inc., Columbus, OH) was used to seal the root ends. Two coats of nail polish (Maybelline, Westfield, NJ) was applied over the entire tooth surface and allowed to dry at room temperature. A mixture of three parts sawdust and one part plaster was mixed with water to make a matrix that simulated the bony socket. The teeth were placed into the unset matrix to make an impression. After the matrix was set, the teeth were removed.

To ensure each perforation was centered between the roots, a #330 high-speed carbide bur was used to perforate the chamber floor from the external surface while the tooth was hand held. Each perforation was enlarged with a #80 file passing 5 mm beyond the root surface. The teeth were randomly divided into two groups of 18 teeth. The original, gray-colored formulation of MTA (Pro-RootTM MTA, Dentsply Tulsa Dental, Tulsa, OK) was used to repair the perforations in group 1, and the new, off-white-colored variety of MTA (ProRootTM MTA) was used to repair the perforations in group 2. Two teeth were perforated but not repaired and serve as negative controls. After treatment, the teeth were replaced in their individual matrices.

Saline-moistened Colla-plug (Calcitek, Carlsbad, CA) was packed into the perforation to provide a matrix for packing MTA (14). An attempt was made to fill the area apical to the perforation flush with Colla-Plug to provide an intimate matrix against which to pack MTA. The amount of Colla-plug packed into the perforation was variable among samples. For both groups, MTA was mixed with sterile water to a thick, creamy consistency and placed into the perforation with a Dovgan carrier (G. Hartzell & Son, Concord, CA). Both materials were condensed with a doubleended endodontic plugger #5/#7. The MTA was placed using magnification of $\times 2.75$ (Orascoptic/Sybron Dental Specialties, Middleton, WI). Both groups were left in the matrices with 100% humidity and allowed to set for 72 h at 37°C.

A dual-chamber, anaerobic, bacterial model was assembled using a 5-ml irrigation syringe and tooth as the upper chamber and a 20-ml scintillation vial (Wheaton, Millville, NJ) as the lower chamber. The syringe was secured via a hole drilled through the cap of the 20-ml scintillation vial. The tooth was attached with cyanoacrylate cement to the tip of the syringe to complete the upper chamber and the joint sealed with two coats of nail polish. A plastic cap from a disposable syringe was used to cover the tube opening of the upper chamber.

The entire apparatus was sterilized with a low-temperature, hydrogen peroxide, gas plasma system (Sterrad® Sterilization System, Johnson & Johnson, Irvine, CA). Brain-heart infusion (BHI) broth supplemented with yeast extract (5 g/L), hemin (5 mg/L), and menadione (10 mg/L) was aseptically placed into the lower chamber to a level above the tooth furcation. The vials were placed in an anaerobic chamber containing 85% N₂, 5% CO₂, and 10% H₂ for 48 h. This was done to eliminate any oxygen in the system, reduce the media before inoculation, and check for sterility of the system. Two to three milliliters of BHI broth turbid with *F. nucleatum* was pipetted into the upper chamber syringe reservoirs. Every 2 weeks excess BHI broth was removed from the upper chamber syringe reservoirs and replaced with 2 to 3 ml of fresh BHI broth turbid with *F. nucleatum*. The vials were incubated in the anaerobic chamber at 37°C and observed every 2 to 3 days for

turbidity of the broth in the lower chamber, indicating bacterial growth from penetration of bacteria past the perforation repair. The leakage evaluations were performed for 60 days. The vials were identified by code unknown to the operator throughout the experiment. Once turbidity was detected, a sample of the broth was Gram stained to verify the presence of *F. nucleatum*. Leakage of *F. nucleatum* was compared between the two groups and statistically analyzed using the Chi-square test with significance set at p < 0.05.

RESULTS

The positive controls showed turbidity 1 to 2 days after inoculation. The negative controls did not leak for the experimental duration of 60 days. The entire contents of the upper reservoir of one sample from group 1 leaked into the lower reservoir by day 2. After microscopic examination, this was determined to be caused by a previously undetected vertical root fracture. Two additional specimens from group 1 and three specimens from group 2 showed leakage that was detected between 15 and 21 days. The difference between the two materials was not statistically significant (p = 0.6787). Gram stains of the BHI broth from both the upper and lower chambers confirmed the presence of *F. nucleatum*.

DISCUSSION

Both gray and off-white MTA are marketed as ProRootTM MTA. According to the manufacturer, MTA is a powder consisting of fine, hydrophilic particles that in the presence of water creates a colloidal gel, solidifying within 4 h. The principal components of the gray-colored formula are tricalcium silicate, bismuth oxide, dicalcium silicate, tricalcium aluminate, tetracalcium aluminoferrite, and calcium sulfate dihydrate. Notably, the off-white-colored formula lacks the tetracalcium aluminoferrite. The lack of this iron-containing compound may account for its off-white appearance. Both formulae are 75% Portland cement, 20% bismuth oxide, and 5% gypsum by weight (13).

Many researchers have evaluated endodontic filling materials in vitro by using methods based on tracers. The tracers most often used are dyes, radioisotopes, bacteria, or bacterial by-products. Isotopes and dye molecules are much smaller than bacteria and most bacterial by-products and, thus, do not simulate the type of leakage that may occur clinically. Our results are consistent with those obtained in an in vitro study using *Staphylococcus epider-midis* by Torabinejad et al. (15) in which only 2 of 10 MTA, root-end samples leaked during the experimental period of 90 days. In their study, one sample allowed leakage at 25 days and the other at 41 days. However, at the conclusion of their 120-day study, Fischer et al. (16) found that 4 of 10 MTA, root-end samples did not allow passage of *Serratia marcescens*. In their study, the first MTA sample to leak was on day 49.

Because leakage of bacteria into the periapical tissues is believed to be a major cause of periradicular pathosis and the majority of bacteria associated with infections of endodontic origin are strict anaerobes, the detection of anaerobic bacteria is a more clinically relevant demonstration of microleakage associated with an endodontic filling material (17–20). Scheerer et al. (21) found that MTA did not allow passage of strict anaerobic bacteria (*Prevotella nigrescens*) for the experimental duration of 47 days. Nakata et al. (11) found that MTA showed no detectable leakage of *F. nucleatum* for the experimental period of 45 days. The results obtained in this study demonstrated a greater incidence of leakage compared with the results of Nakata et al. and Scheerer et al.

Scheerer et al. (21) used a model using 3 mm of MTA as a root-end filling material. In our study, varying thicknesses of MTA were used to repair furcation perforations. The differences in thickness of MTA in our study could account for the greater incidence of leakage. Our study used teeth with furcations of varying thickness, which also may explain the increased incidence of leakage. During the experimental set-up of our study, it was noted that the MTA could be easily over-filled, covering a portion of the pulpal floor. This would allow for more bulk of the MTA and in effect create an artificially greater gap to be traversed by the bacteria. When repairing the artificial perforations in our study, special care was taken to limit the MTA to the confines of the defect and to not allow any extension of the slight differences in the results of our study compared with Nakata et al. (11).

In conclusion, this study demonstrated the use of an anaerobic bacterial leakage model testing teeth with furcation perforations repaired with two types of MTA. There was no significant difference between the gray type of MTA and the off-white type of MTA in allowing the passage of *F. nucleatum*.

The views expressed in this article are those of the author and do not reflect the official policy or position of the United States Air Force, Department of Defense, or the U.S. Government.

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