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**Research Article** 

# Comparison of Pro Root Mineral Trioxide Aggregate and Calcium Enriched Mixture Cement Microleakage as Root End Filling Material: An in Vitro Analysis of Dye Penetration

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Background: Sealing ability is an essential factor for retrograde filling material in successful endodontic apical surgery. The aim of this in vitro study was to evaluate dye microleakage of Mineral Trioxide Aggregate (MTA) and Calcium Enriched Mixture (CEM cement) as root end filling material.

Objectives: The aim of this in vitro study was to compare sealing ability of these two root end filling materials using the dye penetration method.

Materials and Methods: Eighty-six single rooted teeth were randomly divided into two study (n = 40) groups and two positive and negative control (n = 3) groups. After decoronation closely to the cementum enamel junction, the root canals were shaped by the crown down technique and obturated by gutta-percha and AH26 sealer with the lateral condensation method. Furthermore, 3 mm of root ends were resected and 3 mm root-end cavities were prepared by ultrasonic microsurgical tip. Root-end cavities were filled with each of the mentioned materials. Methylene blue dye was used for determination of dye leakage. Chi-2 analysis and t-test were used to compare groups.

Results: The mean apical penetration of dye in the MTA group was 0.94 mm while in the CEM cement group this was 1.17 mm. Eighty percent of the MTA group and 75% of the CEM cement group didn't have any dye leakage. The P level was greater than 0.05 (0.592). Conclusions: There was no significant statistical difference between MTA and CEM cement in sealing ability (P>0.05).

Keywords: Mineral Trioxide Aggregate; Calcium Enriched Mixture Cement; Dental Leakage; Root Canal Filling Materials

# 1. Background

Bacteria play an important role in creating apical lesions, thus the main purpose of endodontic treatment is to remove bacteria and their byproducts from root canals (1). When it is impossible to remove bacteria and their irritants completely by an orthograde treatment and retreatment, then endodontic surgery is indicated (1, 2). One of the important factors that guarantees endodontic surgery is apical sealing ability of the root-end filling material (3).

Until now, a variety of materials have been suggested as root-end filling material. However, it has been mentioned that Mineral Trioxide Aggregate (MTA) has an excellent biocompatibility and sealing ability (3, 4). Many clinicians believe that it is the gold standard of endodontic materials (3). Recently, Calcium Enriched Mixture (CEM cement) has been suggested as a root-end filling composition (3, 5). It plays a role in periapical tissue regeneration. Also it has better biocompatibility and antibacterial effect than that of MTA. Also, it seems that its manipulation is easier than MTA, such as appropriate setting time, decreased working time and increased flow. However, we need to know more about its sealing ability (3).

There are different methods for evaluating endodontic materials microleakage such as bacteria leakage, dye or ink, radio isotopes, electrochemical and fluid filtration techniques (6). However, it seems to be that dye penetration is the easiest and most popular method (1, 6).

## 2. Objectives

The aim of this *in vitro* study was to compare sealing ability of these two root end filling materials using the dye penetration method.

#### 3. Materials and Methods

This experimental in vitro study was performed on 86 single rooted and single canalled extracted teeth, which were randomly divided to two study groups of 40 each and two control groups of three each. The surface of teeth

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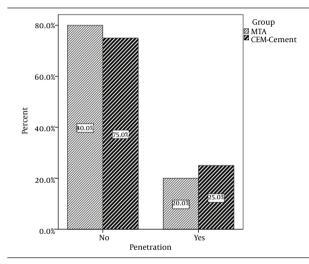
were cleaned and stored in 5.25% NaOCl for one hour. Next, they were washed and stored in normal saline until the time of the study. The teeth were decoronated close to the CEJ (cemento enamel junction) level in a way that the remaining sections were similar in size. Working length of canals was determined by the K-file #15 as 1 mm shorter than the apical point of teeth. Root canals were prepared by the crown down technique and FlexMaster file, in 0.06 tapers and size # 30. Root canals were dried with paper points (Aryadent, Tehran, Iran), and obturated with guttapercha (Diadent, Seoul, Korea) and AH26 sealer (Dentsply, DeTrey, Konstanz, Germany) by the lateral condensation method. Next, 3 mm of the apical part of roots were sectioned perpendicular to the long axis of teeth with highspeed hand piece and # 008 diamond fissure bur under a water spray. Furthermore, 3 mm deep and 1 mm diameter root-end cavities were prepared by ultrasonic instrument and root end cavity of each group was filled with one of the studied materials: ProRoot MTA (Densply Tulsa Dental, Tulsa, OK) and CEM cement (BioniqueDent, Tehran, Iran). The two materials were prepared according to the manufacturer's directions. ProRoot MTA was mixed with a 3:1 powder: liquid ratio and incrementally placed and compacted in the root-end cavity. Root-end surfaces were cleaned and covered with damp cotton pellets. The CEM cement powder and liquid (BioniqueDent, Tehran, Iran) were mixed according to the manufacturer's instructions and delivered to the dried canals using a sterile amalgam carrier and gently adapted to the apical portion of the canals using pre-fitted endodontic pluggers (M-series, Dentsply Maillefer, Tulsa, USA). All MTA and CEM cement retro-fillings were checked after 24 hours to ensure complete setting of the material. Lateral surfaces of roots were entirely coated with two layers of nail polish to prevent penetrating liquid through dentinal tubules and accessory canals. Positive control group: after preparation of root canals, canals stayed empty to permit dye penetration yet external surfaces were covered by two layers of nail polish. Negative control group: after teeth preparation, all root canals were filled with sticky wax and the entire surfaces of teeth were covered by two layers of nail polish to prevent dye penetration.

We used 2 mL-micropipettes (Supa Co., Tehran, Iran). The end of the pipette was cut through which teeth were inserted. The spaces between teeth and micropipettes were sealed with sticky wax. Retrofill materials were exposed to 1% methylene blue for 72 hours, and dye penetration between root-end filling material and apical part of tooth walls were evaluated by a stereomicroscope at  $40 \times$  magnification (Olympus, Tokyo, Japan). Teeth with complete dye penetration were considered as a complete leakage. Thus, they were excluded from the study and others were sectioned longitudinally and were evaluated by the stereomicroscope again. Amount of dye penetration was measured using a millimeter scale. Finally data were statistically analyzed using the Chi-2 and t-test. The statistical significance level was set at P < 0.05.

#### 4. Results

All positive controls had complete dye penetration, while the negative control group represented no dye leakage. Ten (25%) samples of the CEM cement group and eight (20%) samples of the MTA group had dye leakage (Figure 1). The P value was greater than 0.05 (0.592), which indicates no significant statistical difference between the two groups. The Chi-2 analysis test revealed a value of 0.287 and its degree of freedom was one.

The mean dye penetration of the CEM cement group was 1.175  $\pm$  1.3 mm while for the MTA group this was 0.937  $\pm$  1.23 mm. Comparison of dye penetration in both groups is presented in Figure 2. There was no significant difference in mean dye leakage according to millimeter between the two groups (P = 0.404). Using a stereomicroscope (40  $\times$ ) dye microleakage in the MTA and CEM cement group was assessed (Figures 3 - 6).





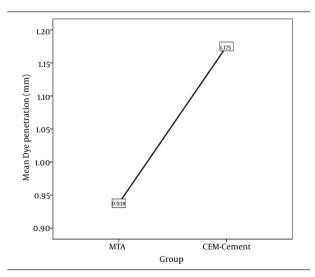
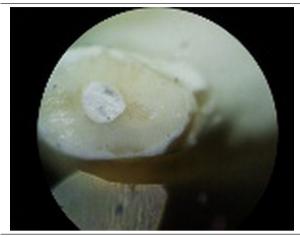


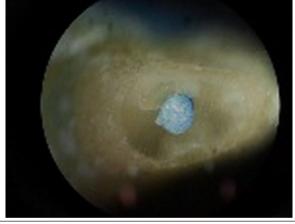
Figure 2. Mean of Dye Penetration in Calcium Enriched Mixture Cement and Mineral Trioxide Aggregate Group Based on Millimeters

Figure 3. Apical View of Calcium Enriched Mixture Cement Sample With No Dye Penetration



Stereomicroscope 40 ×.

Figure 4. Apical View of Mineral Trioxide Aggregate Sample With Dye Penetration



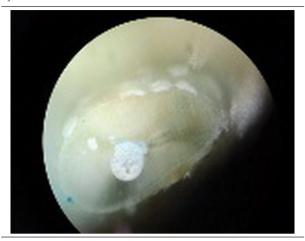
Stereomicroscope 40 ×.

Figure 5. Apical View of Mineral Trioxide Aggregate Sample With No Dye Penetration



Stereomicroscope 40 ×.

Figure 6. Apical View of Calcium Enriched Mixture Cement Sample With Dye Penetration



Stereomicroscope 40×.

#### 5. Discussion

Periradicular surgery is commonly considered as the treatment of choice when nonsurgical treatment has failed or if existing restorative or prosthetic treatment would be endangered by orthograde treatment (7). The success of endodontic surgery depends on the regeneration of periodontal attachment apparatus including, cement, periodontal ligament and alveolar bone. For this reason, it is important to seal an exposed resected root with a biomaterial with sealing ability and biocompatibility, which permits regeneration of normal periodontium on outer root surfaces (7). The biocompatibility of MTA in contact with cells and tissues was confirmed and new cementum formation was detected close to MTA (8). Due to the complexity of root canal system and variety of microbial colonies, which are infected root canals, we cannot warranty root canals of completely no microbial colonies. Thus, if we have tissue fluid leakage into root canals, we prepare good conditions for the growth of microbial colonies and this would be the reason of failure (9). Due to the good properties of CEM cement such as biocompatibility, antibacterial and low cytotoxic effect, flow ability and good clinical handling, CEM cement is considered as an excellent root-end filling material (1).

The results of our study indicate that the microleakage of CEM cement is comparable with MTA, thus it has good apical sealing ability. The dye leakage method determines material adaptation along the canal walls. Due to the small amount of dyes, it may demonstrate bacterial byproduct penetration (10). In dye microleakage studies, dye did not completely penetrate between the sealing materials and teeth, even after two weeks. One reason for such a phenomenon might be the presence of air bubbles that can inhibit the penetration process (1, 11). Also, it has been shown that the type of dye, methylene blue or fuchsine, or even the vacuum conditions don't change the amount of dye penetration (12, 13). In the present study, we placed dye on top of the root end filling material to take advantage of gravity force.

Differences between the results of dye penetration methods might be due to the different principles of each study (14). Also, methods of investigation of microleakage are very sensitive and it is possible for two investigators or even one investigator at two different times to reach different results, which are in disagreement. Unfortunately most of these kinds of studies are not reproducible thus it is difficult to relate the results of these *in vitro* studies to clinical procedures. For example clinical success of the cold lateral condensation technique, which has lots of microleakage *in-vitro* studies, is about 90% (15).

In the present study, 20% of the MTA group showed dye microleakage, while this was 25% for the CEM cement group. Hasheminia et al. investigated microleakage of MTA and CEM cement as retrograde filling materials in 2010. The difference between MTA and CEM cement microleakage was not significant (16). We obtained similar results in our study.

Kazem et al. studied microleakage of eight retrofill materials by using dye penetration and bacterial leakage methods in 2007. The materials studied in this research were gray MTA, white MTA, Pro Root MTA, amalgam, IRM (intermediate restorative material), GI (glass ionomer), CEM cement and Portland cement. Gray MTA showed less bacterial microleakage than the others. Also it was shown that gray MTA dye penetration is less than amalgam and IRM. There was no significant difference between MTA and CEM cement's microleakage, which was similar to the current study (5).

Ashraf et al. in 2013 demonstrated dye penetration and bacterial leakage of Pro Root MTA and Resilon retrograde filling materials. They found no significant differences in microleakage of MTA and Resilon, which were 20% and 15%, respectively (17). We also obtained MTA dye microleakage of 20%, which was similar to their results.

Asgary et al. compared sealing ability of MTA, IRM and CEM cement as root end filling materials using the dye microleakage method in 2008. Tukey's test revealed no significant differences between CEM cement and MTA. Usage of these two biomaterials are recommended and their sealing ability are superior to IRM (18). The results of our study confirmed these findings.

According to the study of Razmi et al. in 2009, which compared sealing ability of two root-end filling materials (MTA and CEM cement) following retro-preparation with ultrasonic or Er, Cr: YSGG laser, laser/CEM (cement) cement group showed significantly lower mean apical dye penetration. There were no statistically significant differences between laser/MTA, ultrasonic/MTA and ultrasonic/CEM cement groups. According to this study, we can conclude that CEM cement could be used instead of MTA for the reason of lower setting time and easier manipulation than MTA (19). We also found no significant differences between MTA and CEM cement group, which revealed they could be used in similar conditions. Thus MTA and CEM cement are nearly similar in sealing ability and we can use CEM cement instead of MTA as root end filling material in periradicular endodontic surgery.

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#### **Authors' Contributions**

Study concept and design: Farhad Faramarzi. Analysis and interpretation of data: Maryam Vossoghi. Drafting of the manuscript: Mitra Vossoghi. Statistical analysis: Mitra Vossoghi. Administrative, technical and material support: Bahareh Shams. Study supervision: Farhad Faramarzi. Critical revision of the manuscript for important intellectual content: Maryam Vossoghi and Farhad Faramarzi.

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