Sealing Ability of Different Root End Filling Materials: An *In Vitro* Study

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Abstract

The aim of this *in vitro* study was to analyze the ability of three materials to seal the root end effectively. Totally 80 extracted mandibular and maxillary single-rooted incisors were provided for this study. The samples were separated into four groups (Group 1: Ca(OH)₂, Group 2: MTA-Angelus, Group 3: ProRoot-MTA, Group 4: Control Group-No apical barrier). Materials were placed as 2 mm apical barriers. The root segments were obturated with sealer with gutta-percha and were covered with double-layered nail varnish up to apical 1 mm area. The apicals of all root segments were stored in methylene blue dye for 48 hours. Dye penetrations were measured using stereomicroscope. Fisher reality and chi-square test using for statistical analysis. According to microscopy results, there wasn't any important differences between the groups. Based on the findings it can be concluded that both MTA-Angelus and ProRoot-MTA have nearly the same sealing ability when used in a one-apical barrier technique.

Keywords: Apical Barrier; Apexification; Mineral Trioxide Aggregate; Root Filling; Sealing Ability

Introduction

The clinician faces an enormous difficulty in endodontic treatment of root apex incomplete pulp diseased teeth. There is no apical stop which creates the most important challenge. It is necessary to use a specific dressing material to neutralize the bacteria and their products and to stimulate the apexification process by forming a mineralized apical barrier so that the subsequent condensation gutta percha can be properly achieved [1].

The traditional treatment procedure aims to increase the apical hard tissue barrier formation repeating intracanal placement of calcium hydroxide. Restorative dentistry make a lot use of calcium hydroxide preparations by means of stimulating hard tissue formation in many endodontic situations; thus an interim root dressing, which is a therapeutic cavity liner [2].

Calcium hydroxide may stimulate a calcified barrier either as a pulp-capping agent or as a material put in the root canal adjacent to healthy pulpal or periodontal tissue. In the pulp, an external layer of necrosis develops which may go up to 2 mm depth as the material has high pH (up to 12.5). The inflammatory reaction is not severe further from this layer. If all bacteria is kept out of the operative field during the placement process of the material, the formation of a hard tissue may occur. On the other hand it is not calcium hydroxide that

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originates the calcium ions forming the barrier but the bloodstream. The most significant part of calcium hydroxide is seen as the hydroxyl group because of its establishment of alkaline environment that stimulates repair and active calcification. The osteoclast release lactic acid which may cause the mineral elements of dentine to dissolve and the alkaline pH induced prevents it by naturalizing. Moreover, the alkaline pH may trigger alkaline phosphatases, an significant enzyme in the development of hard tissue [3].

Despite the advantages of the calcium hydroxide, it has some disadvantages such as longer time of treatment, longer follow up of the patients at certain intervals, unpredictable apical closure, root fracture arising from the presence of thin roots and more sensitive to coronal microleakage and in apexification treatment [4].

The one apical barrier technique with MTA is shown as an alternative treatment option against treatment with calcium hydroxide. It is suggested that MTA be used to form apical barrier on account of its obturation features, biocompatibility, qualification to set in the existence of humidity and ability to activate hard tissue formation [5].

Some factors can endanger the achievement of endodontic treatment [6]. Persistent microorganisms are the main factors associated with chronic apical periodontitis and consequent endodontic failures [7]. One of the indications for peri-radicular surgery is when the conventional treatment is not able to eliminate the microorganisms of the apical tissue. Retrofilling should seal the apex against dissemination of bacterial products into peri-radicular tissues with root canal system [6].

To avoid the possible contaminants going into peri-radicular tissue following a surgical root canal procedure, a durable and leak proof apical seal is needed and this could be achieved with retrofilling materials. Gutta-percha, silver amalgam, glass ionomer, zinc oxide eugenol cements (IRM, Super EBA, Rickerts), calcium hydroxide cements (Selapex, Sealer 26) composite resins and recently MTA are among the materials used as root canal filling materials [8].

MTA is on the dentistry market in various names. MTA-Angelus and ProRoot MTA have similar composition. Duarte, et al. have showed that two together materials release calcium and supply alkaline environment [5,9]. MTA is used as repair material in root perforations, apical barrier in immature teeth with open apices, pulp capping, root-end filling material and pulpotomy treatment protocols [10]. MTA is advantageous because of its antimicrobial activity, biocompatibility, and inhibition of microbial leakage, not be cytotoxic, and support for hard tissue formation with its ability to induce the release of cytokines from bone cells [11,12].

Aim of the Study

The aim of this in-vitro study was to evaluate root-end sealing ability of three materials: Calcium Hydroxide, MTA Angelus and ProRoot MTA.

Materials and Methods

This in-vitro study contained a total of 80 single rooted human teeth prepared for use in experiments. Teeth with resorption, caries, and root fracture were excluded from study group. The teeth were cleaned of calculus, soft tissues and adhered bone and other debris with a ultrasonic scaling after extraction. To prevent the transmission of infection, the teeth had been preserved in 5.25% sodium hypochlorite (NaOCl) for one week and then placed in normal saline until use in the current study.

Ethical Approval

The study was completed compatible with the Helsinki Declaration presented in 1975 which is revised in 2000. Ethical approval of the study protocol was obtained from the Research Ethical Board of Istanbul University Faculty of Medicine, Istanbul, Turkey (number: 2012/1738-1298).

Preparation of specimens

The specimens were decoronated with micromotor handpiece (W&H Den-talwerk Bürmoos GmbH, Austria) and diamond discs (Diamant 0.15mm, Dentaurum, Germany) in order to the roots standartized approximately 10 mm root length [5]. The roots of the teeth were

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resected 2 mm from the root end to make a 90° angle to the long axis. A cylindrical carbide bur was used under water cooling in a high speed cycle at the root end resection. After intracanal tissues were extirpated, the crown down technique with K-files was used to prepare root canals. Apical preparation was operated at the same working length up to a size 40 K-file. The preparing root canal method was completed with 1.2 and 3 Gates-Glidden drills for the coronal shaping.

After using all endodontic instruments, 2 ml of 2.5% sodium hypochlorite (NaOCl) was used for root canal irrigation. In order to remove smear layer, 2 ml of 17% EDTA was administered for three minutes, thereafter a 2 ml distilled water was used as the last irrigation solution.

All steps were identical for each samples. Subsequently specimens were randomly distributed in four groups of 20 samples each.

- Group 1: Calcium hydroxide (Sultan Chemists Inc., Englewood, NJ, USA)
- Group 2: MTA- Angelus (Angelus Soluções Odontológicas, Londrina, Brazil)
- Group 3: ProRoot MTA (Dentsply Endodontics, Tulsa, OK, USA)
- Group 4: Control group – no apical barrier.

Materials were placed as 2 mm apical barriers and manipulated as required manufacturer’s directives. Each group had a mixture of MTA and distilled water in 3:1 ratio on a glass pad. Then the mixture was placed as apical barrier. The mixed material was condensed using a plugger into the root canal of the open apex tooth. After all materials was condensed as a root end filled materials, all the root segments were filled using the lateral condensation technique with AH-Plus sealer and gutta-percha (De Trey; Konstanz, Germany).

Radiographs were taken of all the specimens to corroborate the installation of the apical barriers and the obturation of each root segment. Each sample was stored for one week in 100% humidity and 37°C environment.

Measurement of sealing ability

The external root surfaces of all the root segments, yet the apical 1mm and their apical ends were double covered by coat of nail varnish. Then the all samples were put aside perpendicularly in methylene blue dye for 48 hours at room temperature.

After 48 hours the paint was removed from the samples. The all root segments were washed under tap water. The samples were divided according to their length with diamond discs and linear dye penetration was measured from apical root surface using stereomicroscope at 20X magnification. Figure 1 shows the microscopic photographs of each group.

Figure 1: The microscopic photographs of all groups.

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SPSS for Windows, version 10.0 was used for each statistical analysis. The findings were analyzed statistically by ki-kare test, Fisher reality test and OR (odds ratio) for comparison. P-values less than 0.05 were considered significant.

Results and Discussion

Results

According to microscopy results, the change in distribution of leakage (p = 0.419) between all groups was not considerable. Respectively, in Group 1, 2, 3 and 4 (control group): 6, 10, 11, 9 of the 20 samples showed no dye leakage. The risk of leakage according to control group, 1.9 in Group 1; 0.82 in Group 2; 0.67 in Group 3. The highest risk of leakage was found in Group 1 and the lowest risk of leakage was found in Group 3 (Table 1) (Figure 2).

<table>
<thead>
<tr>
<th>Group</th>
<th>Leakage(+)</th>
<th>Leakage(-)</th>
<th>OR (%95GA)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>14</td>
<td>70%</td>
<td>6</td>
</tr>
<tr>
<td>Group 2</td>
<td>10</td>
<td>50%</td>
<td>10</td>
</tr>
<tr>
<td>Group 3</td>
<td>9</td>
<td>45%</td>
<td>11</td>
</tr>
<tr>
<td>Group 4</td>
<td>11</td>
<td>55%</td>
<td>9</td>
</tr>
</tbody>
</table>

χ²: 2.82 p = 0.419

Table 1: Odds ratio of groups.

According to post-hoc analysis between groups, the distinction was not statistically important (p > 0.05) (Table 2).

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### Table 2: The risk assessment of groups.

<table>
<thead>
<tr>
<th>Comparison of groups</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1/Group 2</td>
<td>0.332</td>
</tr>
<tr>
<td>Group 1/Group 3</td>
<td>0.201</td>
</tr>
<tr>
<td>Group 1/Group 4</td>
<td>0.514</td>
</tr>
<tr>
<td>Group 2/Group 3</td>
<td>0.751</td>
</tr>
<tr>
<td>Group 2/Group 4</td>
<td>0.746</td>
</tr>
<tr>
<td>Group 3/Group 4</td>
<td>0.752</td>
</tr>
</tbody>
</table>

**Discussion**

The root development and closure of the apex takes place as far as 3 years after the tooth erupts. During this period, the treatment of pulpal injury is not easy for the clinician. Two treatment protocols are conducted considering the vitality of affected pulp, apexogenesis or apexification [1]. Apexogenesis is a vital pulp therapy. It is aimed to maintain the development of the root physiologically in the treatment of apexogenesis [6]. Apexification is a method of inducing a calcified barrier at the apex of a nonvital tooth with incomplete root formation [5].

The dental caries and trauma can affect the development of teeth. In both cases the dental pulp can be non-vital. Dental pulp is non-vital before the root develops and the apical foramen closes, anatomical root development is changed entirely. The gutta percha may not be fully condensed since the apical barrier is incomplete in these cases. In traditional treatment protocols, it is recommended to place calcium hydroxide into the root canal at certain periods to form apical hard tissue barriers. This chemical material has several disadvantages for example it may make recall management of the patient difficult or create a delay in the treatment [5]. In this study, the most leakage was seen in the calcium hydroxide group.

The seeking of another material instead of calcium hydroxide, the preferred treatment for a long time, has been going on. MTA, an alternative, contains bismuth oxide and Portland cement, consisting of dicalcium silicate, tricalcium silicate, tricalcium aluminate and tetracalcium aluminoferrite. After being set, MTA has the same pH (12.5) with calcium hydroxide [2,7]. This material is osteoconductive, which might be a help in the healing and adapting process of the periapical tissue. There are studies examining the process of apexification of immature teeth either using MTA or calcium hydroxide [4]. In this study we compared calcium hydroxide and two MTA materials. Because MTA materials are more durable materials in terms of content, leakage amounts are less.

El-meligy and Avery’s studies on apexification treatment with MTA or calcium hydroxide were conducted with 15 children who have at the least two necrotic permanent teeth. They carried radiographic and clinical evaluations after 3, 6 and 12 months of treatment. The results show that 2 out of 15 children treated with calcium hydroxide had permanent peri-radicular inflammation and sensitivity to percussion in 6 months and 12 months yet no children treated with MTA had the same problem [13,14].

Moretti., et al. randomly selected among 23 children who are aged from 5 to 9 and have 45 mandibular primary molar teeth with dental caries and treated them with zinc oxide-eugenol paste (control treatment), calcium hydroxide and MTA. During the 24 month follow up, the children who received MTA or control treatment had successful clinical and radiographic results and 29% of the children in MTA group showed dentine bridge formation. However among the ones who received calcium hydroxide treatment, 64% of them had unsuccessful clinical and radiographic results and internal resorption was a common radiographic indication [15].

Bonte., et al. choose randomly among children who needed apexification because of nonvital permanent incisors and used either calcium hydroxide or MTA. 50% of the children treated with calcium hydroxide showed a mineralized barrier, while 82% of the ones treated with MTA showed a mineralized barrier as well (p < 0.07) after 12 months. Root fractures were observed in 4 of the 15 teeth in calcium hydroxide treated group yet none in the MTA group [16].

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Menezes., et al. investigated seventy six dogs teeth which received direct pulp protection and pulpotomy treatment with Portland Cement, White Portland cement, ProRoot MTA and MTA Angelus. To conduct histological analysis, samples were taken from sacrificed animals 120 days after treatment. Being used as pulp-capping materials each material showed similar outcomes. Each sample preserved pulp vitality and a hard tissue bridge was formed during healing process [14].

Shahi., et al. compared the sealing properties of 4 different root end filling materials: Gray Portland cement (PC), White Portland cement (PC), gray MTA and White MTA by using dye leakage test. After all teeth were prepared, 96 single–rooted human teeth were randomly categorized into 6 groups as 4 experimental groups (n:20) and 2 control groups (negative control groups and positive control groups n:8). Experimental materials were applied to the prepared root canal cavities in the experimental groups. A stereomicroscope at 16x magnification was used to measure penetration of dye. Any dye penetration was not demonstrated by the negative control groups but the positive control groups showed dye penetration in the complete root-end cavity. However, between the groups the difference was not statistically important (p > 0.05) [8]. There was no significant difference between the materials in this study.

Lolayekar., et al. randomly assigned 50 maxillary and mandibular incisor teeth to receive ProRoot MTA, MTA Angelus and control group (no material). After the all materials were manipulated and teeth were prepared, root sealing ability of three groups was evaluated. The control group had showed complete dye penetration whereas the microleakage at different rates was observed in the experimental groups. On the other hand ProRoot MTA and Angelus MTA hadn’t differ significantly from each other [5]. According to the control group, although the amount of leakage of MTA materials was less, no statistical significance was found.

In this study we tested the sealing ability of calcium hydroxide, MTA Angelus and ProRoot MTA as apical barriers. The degree of microleakage was calculated with a dye penetration method, as the method is cheaper, has high degree of dyeing and is lighter than bacterial toxins in molecular terms [8].

Besides its positives attributes, it has some potential problems. Particle molecular size, pH and chemical reactivity of the dye can influence its degree of penetration. In addition, it was shown that alkaline materials such as MTA and calcium hydroxide can discolorate the methylene blue dye, which may lead to unreliable finding [16].

Conclusion
In conclusion, the present study demonstrated that in terms of microleakage MTA Angelus, ProRoot MTA and calcium hydroxide have shown similar results. Based on above findings it can be concluded that all experimental groups have similar sealing ability when used in a one-step apical barrier technique.

MTA is known as a material that requires a lot smaller amount of time when it is used to form the apical barrier, therefore it makes the treatment less long. When the time of treatment is shorter, the success rate of MTA in clinical situations is higher as the patient complies better with treatment completion.

Conflict of Interest
The authors declares that there is no conflict of interest regarding the publication of this paper.

Bibliography


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