Color Changes of Teeth after Treatment with Various Mineral Trioxide Aggregate–based Materials: An *Ex Vivo* Study

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Abstract

Introduction: Mineral trioxide aggregate (MTA) materials have been used for many years as a pulp therapy material. The most widely used product, ProRoot MTA (Dentsply, Tulsa, OK), has a major drawback in that it causes tooth discoloration. Alternatives have recently been developed such as ENDOCEM Zr (MARUCHI, Wonju, Korea) and RetroMTA (BioMTA, Seoul, Korea). The purpose of this study was to compare the discoloration of these various MTA-based materials. **Methods:** Discoloration of discs prepared from 4 different MTA-based materials (ProRoot MTA, MTA Angelus [Angelus, Londrina, PR, Brazil], ENDOCEM Zr, and RetroMTA) were observed at 15 and 30 minutes after exposure to light at an intensity of 1000 mA/cm^2. In a tooth model, 12 premolars were used per each group to retrofill the pulp chamber with MTA-based materials. The degree of discoloration was measured over a 16-week period using a digital spectrophotometer. **Results:** Distinct color changes were observed for discs made from ProRoot MTA and MTA Angelus, but no clear change was observed for those made from either ENDOCEM Zr or RetroMTA. In the tooth model, more distinct, time-dependent color changes were observed for teeth filled with ProRoot MTA and MTA Angelus than for those filled with ENDOCEM Zr and RetroMTA. **Conclusions:** Less discoloration was observed with ENDOCEM Zr and RetroMTA (which contain zirconium oxide) than with ProRoot MTA and MTA Angelus (which contain bismuth oxide) in both of the test models used. *(J Endod 2015;41:737–741)*

Key Words

Bismuth oxide, discoloration, ENDOCEM Zr, mineral trioxide aggregate, RetroMTA, zirconium oxide

Mineral trioxide aggregate (MTA) was approved as a root canal treatment material by the US Food and Drug Administration in 1998; since then, it has been used in various areas of the root canal treatment process, such as apexogenesis, pulpotomy, direct pulp capping, and root perforation repair, because of its excellent biocompatibility and sealing ability (1–5). There have been many researches and clinical case reports on the use of MTA or MTA-like materials as pulp treatment materials in deciduous teeth and immature permanent teeth (6–8).

Despite its reported advantages, the use of MTA has the following drawbacks: it is difficult to handle, expensive, difficult to remove, and discolors over time (9). Of major importance esthetically, the discoloration that occurs produces results that are not esthetically good, which is particularly problematic when direct pulp capping or pulpotomy are required in the anterior jaw area (10, 11).

The first MTA material to be developed for use in teeth, gray MTA (GMTA), was a dark color and was found to cause tooth discoloration; therefore, white MTA (WMTA) was developed, which had similar clinical properties to GMTA but produced more esthetically pleasing results (12). The difference between WMTA and GMTA lies in their respective contents of metal oxides (eg, iron, aluminum, and magnesium oxides), which are the main cause of discoloration (13). However, WMTA also causes some tooth discoloration because it still contains various metal oxides, although the content is low, together with bismuth oxide, which was added to improve the radiopacity of the material (10, 11, 14). The degree of discoloration that occurs depends on the content of the components of metals such as bismuth, iron, aluminum, and magnesium oxides. Therefore, within a range that does not change the properties of MTA, the content of bismuth oxide could be reduced to minimize any color change. ENDOCEM Zr (MARUCHI, Wonju, Korea) and RetroMTA (BioMTA, Seoul, Korea) are products that have recently become available commercially in which bismuth oxide has been replaced by zirconium oxide as an alternative radiopacifier. Although the manufacturers claim that this product does not cause discoloration either *in vivo* or *ex vivo*, as yet there have been no studies to confirm this. Based on this discussion, we established the following null hypothesis: ENDOCEM Zr and RetroMTA (which contain zirconium oxide) show less discoloration than MTA Angelus (Angelus, Londrina, PR, Brazil) and ProRoot MTA (Dentsply, Tulsa, OK) (which contain bismuth oxide) in an *ex vivo* model.

Therefore, the aim of the present study was to compare the degree of tooth discoloration that occurs over time in specimens and extracted teeth filled with ENDOCEM Zr, RetroMTA, MTA Angelus, and ProRoot MTA.

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Materials and Methods

Disc Discoloration Experiment

Discs were fabricated by mixing distilled water with ProRoot MTA, MTA Angelus, ENDOCEM Zr, or RetroMTA according to the manufacturer's recommendations using a rubber mold (8 mm × 1 mm). In addition, bismuth oxide powder (Duksan, Ansan, Korea) and zirconium oxide powder (Sigma-Aldrich, St Louis, MO) were put into 4-well plates and then soaked for 15 minutes in glycerin (Duksan). The disc and bismuth/zirconium specimens were then irradiated with light for 15 minutes or 30 minutes using a VALO light irradiator (Ultradent, South Jordan, UT) in standard power mode (1000 mW/cm^2). The specifications of the ProRoot MTA, MTA Angelus, ENDOCEM Zr, and RetroMTA used in the present study are given in Table 1.

Preparation of Extracted Teeth for the Discoloration Experiment

Sixty premolars (30 maxillary premolars and 30 mandibular premolars) that had been extracted for orthodontic purposes from 20 people aged 10–29 years (11 males and 9 females) were used. Teeth with dental abnormalities, restorations, or more than moderate dental caries were excluded. The experiment was performed under guidelines approved by the Institutional Review Board of the Dental Hospital, Yonsei University. Written, informed consent for their teeth to be used was obtained from all of the subjects and their parents (form # 2-2013-0036).

After cutting the teeth horizontally about 2 mm below the cementoenamel junction, the pulp tissue was removed using a barbed broach (Mani, Tokyo, Japan), and any organic materials on the surface of the teeth were physically removed by curettage; persistent organic material was removed by soaking the teeth for 10 minutes in 2.5% sodium hypochlorite solution (Duksan). The teeth were then washed several times with normal saline and stored at 4°C until required for the experiment.

The teeth were assigned to 1 of 5 groups (n = 12 teeth per group) named according to the MTA product that would be used to fill their pulp chamber: control (no MTA material), ProRoot MTA, MTA Angelus, ENDOCEM Zr, and RetroMTA. The materials were prepared as instructed by the manufacturer and then used to retrofill the teeth to the level of the cementoenamel junction in the pulp chamber. After the early setting phase, the lower area was sealed with light-cured compomer base (Ionosit-Baseliner; DMG, Hamburg, Germany). In the control group, the pulp tissue was removed from the teeth, which were then only sealed with Ionosit-Baseliner compomer.

Molds were fabricated for each tooth using rubber impression material (Aquasil Soft Putty, Dentsply). Holes with diameters of about 6 mm were created in the molds using a biopsy punch (SFM Medical Devices, Wächtersbach, Germany) so that the extent of discoloration could be measured at the same site. The hole was located so that it revealed an area between the midbuccal third and cervical third to facilitate observation of any MTA-induced internal discoloration (Supplemental Figure S1 is available online at www.jendodon.com).

Measurement of Tooth Color

All of the specimens were put into 12-well plates (BD Falcon, Franklin Lakes, NJ) and stored next to a window in artificial saliva (Tariba; Hanlim Pharm, Seoul, Korea), which was changed every 2 weeks. The tooth color was measured using the VITA Easyshade Compact device (Vita Zahnfabrik, Bad Sackingen, Germany) at the following time points: 0 (baseline), 1, 2, 4, 8, 12, and 16 weeks. The color measurement was made in triplicate for all experimental groups using the $L^*a^*b^*$ value, where L^* is the light intensity, a^* is the red-green parameter, and b^* is the yellow-blue parameter. Using these $L^*a^*b^*$ values, the differences between the color measured at the initial time point (time 0) and those measured at the various time points (ΔE) were calculated as follows:

$$\Delta E = \left[(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2\right]^{1/2}$$

where ΔL, Δa, and Δb are the changes in L^*, a^*, and b^*, respectively, between the initial time point and the next time point being compared.

Statistical Analysis

All statistical analyses for differences in color change among the products between the initial time point and the 6 other time points after the initiation of light treatment were conducted using SPSS Statistics (versions 21.0; SPSS IBM, Armonk, NY). One-way analysis of variance ($P < .05$) was performed, and the Scheffé test (Bonferroni correction, $P < .01$) was used as a post hoc test.

Results

Disc Discoloration Experiment

After 15 and 30 minutes of light exposure, the discs made from MTA Angelus and ProRoot MTA discs were darkly discolored, whereas there was no color change for the RetroMTA and ENDOCEM Zr discs. The discoloration was darker for the ProRoot MTA disc compared with the MTA Angelus disc. In addition, although there was a clear color

TABLE 1. Mineral Trioxide Aggregate–based Materials Used in the Study

Material	Manufacturer	Ingredients
ProRoot MTA	Dentsply, Tulsa, OK	Portland cement 75% Calcium sulfate dehydrate (gypsum) 5% Bismuth oxide 20%
MTA Angelus	Angelus, Londrina, PR, Brazil	Calcium oxide 55%–60% Silicon dioxide 17%–20% Aluminum oxide 2%–4% Bismuth oxide 18%–22%
ENDOCEM Zr	MARUCHI, Wonju, Korea	Calcium oxide 27%–37% Silicon dioxide 7%–11% Aluminum oxide 3%–5% MgO, Fe_2O_3 3%–5% Zirconium dioxide 43%–46%
RetroMTA	BioMTA, Korea	Calcium carbonate 60%–80% Silicon dioxide 5%–15% Aluminium oxide 5%–10% Calcium zirconia complex 20%–30%

change in the bismuth oxide powder with curing time, the zirconium oxide powder did not display any discoloration (Fig. 1).

Tooth Discoloration Experiment

One-way analysis of variance revealed significant differences between each of the 5 groups after 8 weeks of light exposure ($P < .05$). Furthermore, the post hoc test revealed a statistically significant difference among the ProRoot MTA, MTA Angelus, and control groups at 8, 12, and 16 weeks ($P < .01$, Fig. 2). In addition, at 16 weeks, RetroMTA differed significantly from ProRoot MTA and MTA Angelus, whereas ENDOCEM Zr did not differ significantly from ProRoot MTA or MTA Angelus. As shown photographically in Figure 3, prominent discoloration was observed in the ProRoot MTA and MTA Angelus groups at 8, 12, and 16 weeks. Analysis of longitudinal sections made after 16 weeks of light exposure revealed darkening of the outer surface of teeth treated with Angelus MTA and the contacting dentinal tubules rather than of the material itself. No discoloration was found anywhere in similar sections of teeth treated with RetroMTA (Supplemental Figure S2 is available online at www.jendodon.com).

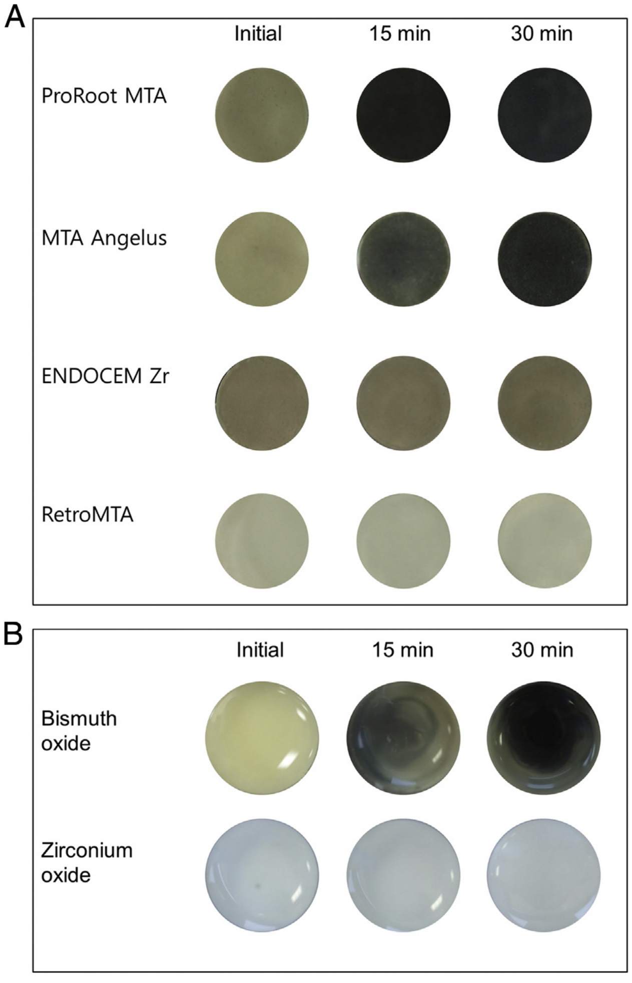

Figure 1. Discoloration of 4 discs made of different types of commercially available MTA products (as labeled) at different time points after exposure to light. (*A*) Images of MTA discs in glycerin exposed to light for 0 minutes (initial) and 15 and 30 minutes. (*B*) Images of bismuth oxide and zirconium oxide powder in glycerin exposed to light for 0 minutes (initial) and 15 and 30 minutes.

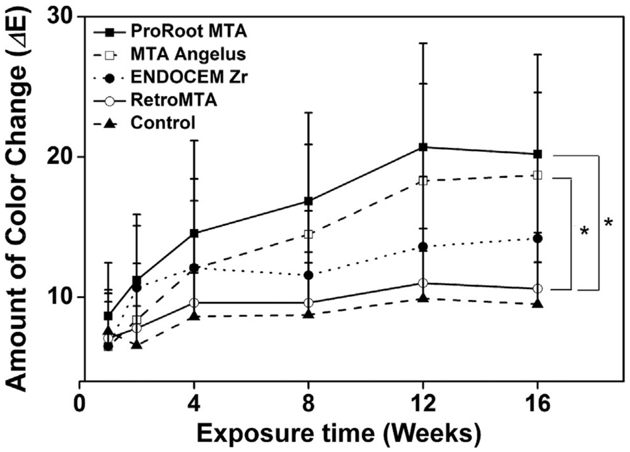

Figure 2. Change in ΔE values in groups of teeth retrofilled with 1 of 4 MTA-like materials (as labeled) and a control group during 16 weeks of light exposure. ΔE values indicate the differences in color compared with the initial color and were calculated using 3 different measurements of $L^*a^*b^*$ values. Data are mean and standard deviation values. *Statistically significant differences among the groups at 8, 12, and 16 weeks of light exposure (analysis of variance test, $P < .05$; post hoc Scheffé test, $P < .01$).

Discussion

This study conducted discoloration experiments on a variety of MTA materials that are indicated clinically for pulp treatment, with a view to determining which would be the best for use in esthetically important areas of the jaw. It was found that RetroMTA and ENDOCEM Zr exhibited less discoloration over the 16-week experimental period than the conventionally used Angelus MTA and ProRoot MTA. Despite the advantages of MTA as an excellent pulp treatment material, it does cause discoloration and therefore should be used with caution for the treatment of incisors, which are located in an esthetically important area of the jaw (15, 16).

When exposed to light in an oxygen-free environment, bismuth oxide, which is reportedly the dominant cause of MTA discoloration (15–17), dissociates into dark-colored crystals of metallic bismuth and oxygen. First, it is known that bismuth oxide is activated by both visible and ultraviolet (UV) light in a similar way to when it is heated. The UV-visible diffuse spectrum for nanocrystallite bismuth oxide spans from wavelengths shorter than 300–500 nm, with a maximum at 400 nm (18). In this study, the disc and bismuth/zirconium specimens were irradiated with light using a VALO light irradiator, which spans from 395–480 nm and is within the UV-visible diffuse spectrum for nanocrystallite bismuth oxide. Therefore, irradiation with curing light has been used to accelerate the rate of WMTA discoloration. Also, it should be possible to prevent discoloration by inhibiting the formation of this metallic bismuth element through an increase in oxygen pressure. The oxygen-free environment that is created by the placement of a restoration over the MTA after pulp treatment may encourage the discoloration of the MTA beneath the restoration. Glycerin was used in the present study to reduce the exposure of the test MTAs to oxygen, thus mimicking the conditions that exist during tooth restoration. The oxygen transmission rate of glycerin is low (19) so it acts as a reducing agent, "stealing" the oxygen element from bismuth oxide (20) and thus rapidly inducing discoloration. In the present study, ENDOCEM Zr and RetroMTA, which contain zirconium oxide as a radiopaque material, exhibited less discoloration than the MTAs that contain bismuth oxide (ie, ProRoot MTA and MTA Angelus). Moreover, the discoloration obtained with ProRoot MTA was slightly darker than with MTA Angelus; this finding is in line with the

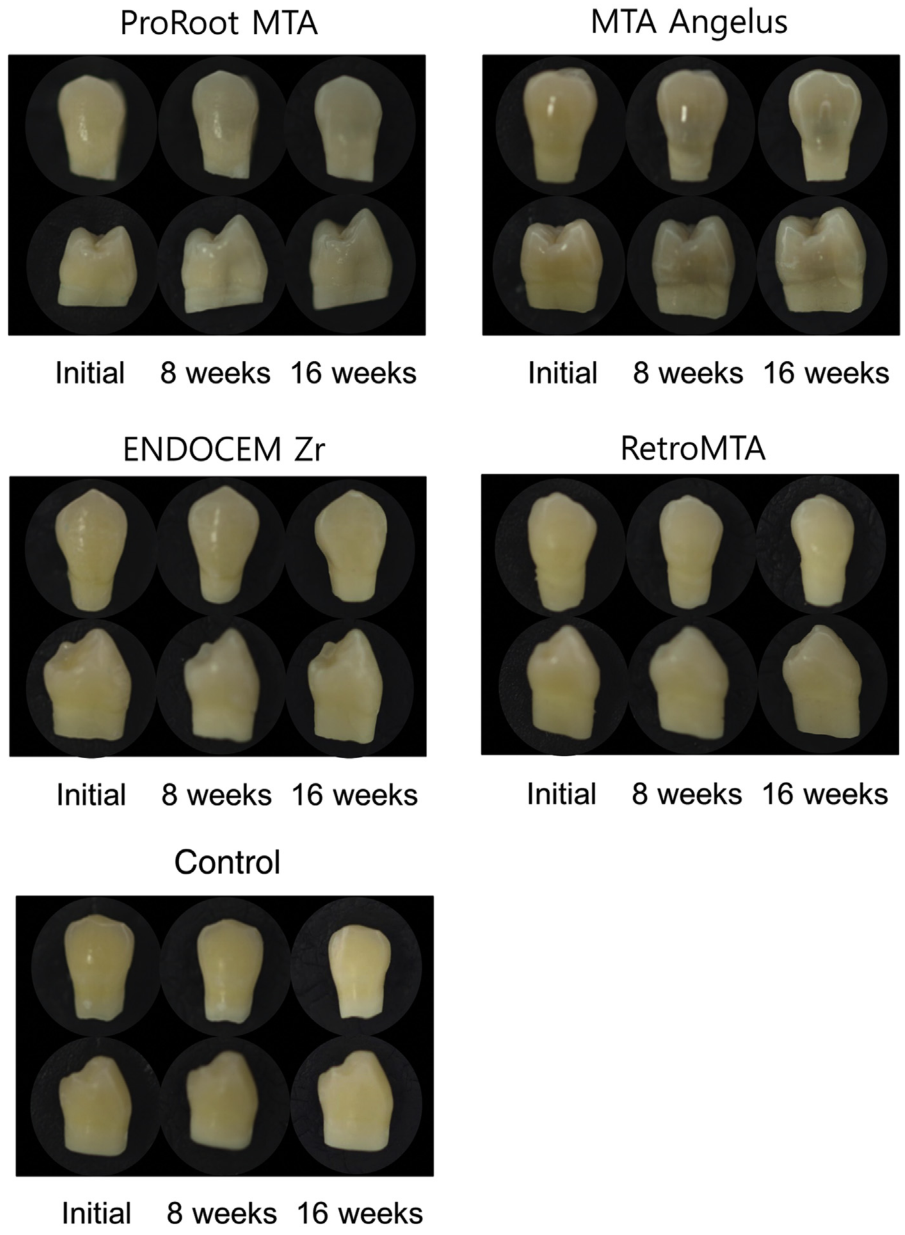

Figure 3. Photographs of discoloration observed at 0, 8, and 16 weeks of light exposure in teeth retrofilled with ProRoot MTA, MTA Angelus, ENDOCEM Zr, and RetroMTA and in control teeth (no MTA filling). The teeth retrofilled with ProRoot MTA and MTA Angelus exhibited a relatively more distinct color change than those retrofilled with ENDOCEM Zr and RetroMTA and the control group.

bismuth oxide content being lower for MTA Angelus than for ProRoot MTA (21).

Paradoxically, overoxidation of bismuth oxide can also lead to discoloration. Discoloration occurs when materials containing bismuth oxide and WMTA come into contact with sodium hypochlorite solution and become oxidized (17). When bismuth oxide is oxidized, its oxygen becomes unstable and reacts with carbon dioxide in the air, producing bismuth carbonate, which is sensitive to light and causes discoloration.

However, there are other potential causes of the discoloration. For example, 1 study found that the degree of discoloration of MTA differed according to the type of light source used, such that the discoloration rate increased as the brightness and wavelength range of the light source increased (16). Furthermore, other studies have shown that the blood in the pulp can deepen the color change of the MTA and that MTA must therefore be applied only after perfect hemostasis or complete removal of the pulp to reduce this discoloration (15, 22).

Research has also been conducted to find ways to overcome MTA discoloration. The application of a dentin bonding agent before filling the pulp cavity with MTA prevented the MTA component from penetrating the dentinal tubules, resulting in less discoloration (23). In another study, bleaching was tested as a method of removing discoloration, but this was unsuccessful as long as the MTA remained in place (24). Ultimately, because the discoloration is induced primarily by the metal components contained within the MTA, such as aluminum, iron, magnesium, and bismuth oxide, it can currently only be prevented by reducing the contents of these metal materials to a degree that does not alter the properties of this pulp as a therapeutic material (12).

Alternative materials such as zirconium oxide and tantalum oxide have been manufactured and used in recent years to reduce the contents of the heavy metals in MTAs and to replace bismuth oxide (25, 26). Unfortunately, there have been few reports on the recently developed MTA-like materials used in the present study. ENDOCEM Zr is a zirconium oxide–enriched calcium silicate–based cement that is fast setting and has reduced solubility (27). RetroMTA was developed as a bioceramic cement that contains a hydraulic calcium zirconia complex and has a reduced setting time of 150 seconds (http://www.biomta.com).

The development of dental materials has thus far focused only on the functional aspects; however, the focus of future studies should be the development of materials with ideal characteristics in terms of esthetics. To that end, the goal of the present study was to determine the relative tendency toward discoloration of 4 currently available MTA materials and to determine whether, as claimed by the manufacturers, the more recently developed materials (ENDOCEM Zr and RetroMTA) are esthetically superior to the conventionally used MTAs (ProRoot MTA and MTA Angelus). Extracted teeth were sectioned for use as test models in this study, and although this *ex vivo* condition is different from the *in vivo* situation in real patients, the study design provides an adequate model for the comparison of discoloration in MTA-based materials (11, 23).

In conclusion, the newly introduced ENDOCEM Zr and RetroMTA, which contain zirconium oxide, exhibited less discoloration compared with the existing ProRoot MTA and MTA Angelus, which contain bismuth oxide.

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The authors deny any conflicts of interest related to this study.

Supplementary Material

Supplementary material associated with this article can be found in the online version at www.jendodon.com (http://dx.doi.org/10.1016/j.joen.2015.01.019).

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