Quantifying Coronal Primary Tooth Discoloration Caused by Different Pulpotomy Materials

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Introduction: *Bioceramic materials, gray and white mineral trioxide aggregate (GMTA, WMTA), have been shown to have high rates of success in various endodontic applications. A major drawback is their tendency to discolor teeth. Biodentine (BD), a recenlty developed bioceramic material, has been claimed not to discolor teeth. The aim of this study was to compare tooth discoloration after applying different pulpotomy base materials (BD, GMTA and WMTA).*

Methods: *Forty human fully developed primary incisors teeth were used in this study. Coronal access was achieved by a Tungsten Carbide drill, and the pulp chambers were accessed and chemo-mechanically debrided. Each material was placed in the pulp chamber, up to the cervical sectioning level. All specimens were incubated at 37°C and 100% humidity for 14 weeks and have been evaluated before the study and weekly. Color was assessed according to the CIE L*a*b* color space system.*

Results: *The ΔE (delta E) of all experimental groups (GMTA, WMTA and BD) were significantly different from the control group at all time points. Color changes in the GMTA and WMTA groups showed significantly higher discoloration compared to BD group in the cervical part of the crown, since week 1. WMTA group showed significant discoloration in the cervical part as of week 1, and gradually increased over time. BD group showed no significant discoloration over time. GMTA group showed the most significant discoloration at week 1 and week 14 ($P<0.05$).*

Conclusions: *both GMTA and WMTA pulpotomy materials may discolor tooth structure over time in an extracted primary anterior tooth model. When choosing bioceramic pulpotomy material, BD may be preferable, mainly in esthetic area.*

Key Word: *Biodentine, Discoloration, Pulputomy, Mineral Trioxide Aggregate, Pro Root*

INTRODUCTION

Pulpotomy is the accepted treatment procedure for primary teeth with exposed coronal pulps inflamed by bacteria due to caries, traumatic injury, or another iatrogenic cause[1]. The goal of pulpotomy is to remove infected coronal pulp tissue while preserving healthy radicular pulp, thereby promoting the integrity and retention of teeth until physiologic exfoliation[2]. Various pulpotomy medicaments aiming at devitalization, preservation, or regeneration of the remaining pulp tissue have been used to date. Among those medicaments are: formocresol, calcium hydroxide, gluteraldehyde, devitalising (N2) paste (paraformaldehyde), zinc oxide eugenol (ZOE), kripaste, Ledermix, electrosurgery, ferric sulphate, bioceramic materials, mineral trioxide aggregate (MTA), growth factors, lasers etc[3]. The ideal pulpotomy medicament would be bactericidal, biocompatible, promote the healing of the root pulp, and be compatible with the physiological process of root resorption[4]. Such a medicament or technique with all of those features remains unavailable. Formocresol has been challenged due to its potential deleterious effects[5], glutaraldehyde has not gained popularity due to its instability and short life span[6], ferric sulfate has evidence of causing internal resorption[7] calcium hydroxide's high PH wounds the pulp in

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a manner that initiates the inflammatory cascade[6]. Given the lack of clear evidence supporting the superiority of any particular treatment method[8] research has continued to seek alternative pulpotomy agents that can provide better clinical efficacy without secondary effects.

Tooth discoloration induced by endodontic materials is a common finding and may impair the esthetic outcome of endodontically treated teeth[11]. A major etiological factor for the occurrence of local intrinsic staining, especially in the cervical and middle thirds of the crown, is the presence of root canal filling materials in contact with the coronal dentin of the pulp chamber. Any change to the optical and chromatic properties of the dentinal structure is likely to cause an alteration in the outward appearance of the crown caused by its light transmitting and reflecting properties[12].

Mineral trioxide aggregate (MTA) is a biomaterial that has been investigated for endodontic applications since the early 1990s. MTA materials are derived from a Portland cement parent compound; It is a bioactive silicate, containing hydrophilic particles, such as tricalcium silicate, tricalcium aluminate, dicalcium silicate, tricalcium oxide, bismuth and iron[13]. MTA has been demonstrated to be a biocompatible material with ability to form hydroxyapatite when exposed to physiologic solutions and may provide better microleakage protection[14]. In both animal and human studies, MTA materials have been shown to have excellent potential as pulp-capping and pulpotomy medicaments, but studies with long-term follow-up are limited. Even though high success rates were reported, a number of cases of discoloration have also been noted.

Original MTA was first introduced in grey color, which caused severe discoloration after its application, and concerns were raised about the aesthetic competency of this material. The tooth colored formula (WMTA) was then introduced to compensate for this short coming[15,16]. The crystalline structure, as well as the chemical properties and mechanism of action is somewhat unchanged, although higher concentrations of aluminum oxide, magnesium oxide and iron oxide in Grey MTA (GMTA), result in a considerable staining potential[16,17] Determination of the effect of dentin bonding agent (DBA. To reduce the discoloration potential, the chemical composition of MTA was changed, and an improved formulation was later introduced as White MTA (WMTA). The most significant difference between the two types of MTA is the lack of iron ions in WMTA[18]. However, it has been reported that WMTA may cause discoloration as well[19,20]. Some authors state that the discoloration induced by MTA may be attributed to bismuth oxide, which is added to improve the radiopacity in both grey and white formulations[21]. However, no scientific evidence is available to support this statement. Because MTA is based on industrial Portland cement with bismuth oxide added as an radiopacifier, pure Portland cement was suggested as an alternative[22]. Newer bioceramics have been developed to address the drawbacks of gray and white MTA. Although various calcium-silicate based products have been launched to the market recently, one among these has been the focus of attention and the topic of a variety of investigations. This material is the Biodentine (BD, Septodont, Saint-Maur-des-Fossés, France), calcium-silicate based product, which became commercially available in 2009. BD indications are similar to MTA, and according to BD manufacturers does not cause tooth discoloration. Studies have shown that it has similar clinical features to MTA[23–26] BD is actually formulated using the MTA-based cement technology with improvements of some properties of these types of cements such as physical qualities and handling[21]. It is indicated for crown and root dentin repair treatment, indirect and direct pulp capping, pulpotomy, repair of perforations or resorptions, apexification, and root-end fillings[27,28].

The primary anterior teeth, particularly the maxillary anterior teeth, have a key impact on facial and oral aesthetics of children[29]. These can severely affect the quality-of-life of children, causing physical, social and psychological impairment[30,31] Despite these concerns, discoloration studies of anterior primary dentition and its impact have received relatively little attention in the literature, compared to the color of permanent dentition. Therefore, the aim of this study was to assess in vitro the color alterations of primary teeth associated with different pulpotomy materials by using a reflectance spectrophotometer.

MATERIALS AND METHOD

Prior to the investigation the study was approved by Tel-Aviv university research ethics board. Forty human fully developed primary incisors teeth were used in this study, recently extracted from male and female patients ranging in age from 2–6 years. The teeth were selected based on dimension, similarity in morphology, and absence of any crack or carious defects; minimum of two thirds of the original root length was required in order to be included in the study. Debris and soft tissue remnants on the root were removed with a sharp scalpel. All teeth were stored in phosphate-buffered saline until used. Coronal access was achieved by a tungsten carbide drill HS 330 (Dentsply Maillefer, Tulsa, OK, USA), pulps were extirpated with an excavator, and the internal axial walls of the pulp chambers were chemo-mechanically debrided with Hedström files (Dentsply Maillefer, Tulsa, OK, USA) and 2.5% sodium hypochlorite (10 mL) through the access. After final irrigation, the pulp chambers were washed with sterile saline (5 mL). After mixing the materials according to manufacturers' instructions, the pulpotomy base material was placed on the floor of the pulp chamber and padded against the pulp orifices using a Dovgan carrier (Quality Aspirators, Duncaville, TX, USA) through the access. A slight vertical pressure was applied with finger pluggers to fill the pulp chamber with the material, up to the cervical sectioning level. Ten negative controls were instrumented and kept unfilled.

The experimental teeth were randomly divided into the following experimental groups:

- WMTA Group (N=10): Mineral Trioxide Aggregate (Pro Root; Dentsply-Tulsa Dental, Johnson City, TN, USA) mixed according to the manufacturer instructions was placed inside the root using Dovgan carriers (Quality Aspirators, Duncanville, Texas).
- Biodentine Group (N=10): Biodentin (Septodont, Saint-Maur-des-Fossés, France) mixed according to the manufacturer instructions was placed inside the root using Dovgan carriers (Quality Aspirators, Duncanville,Texas).
- GMTA Group (N=10): MTA ANGELUS (GMTA, Angelus, Soluções em Odontologia, Londrina, PR, Brazil) mixed according to the manufacturer instructions was placed inside the root using Dovgan carriers (Quality Aspirators, Duncanville,Texas).
- Control Group (N=10): without pulpotomy base material.

The coronal access cavity was then filled with resin modified glass ionomer (EQUIA Forte Fil, GC Europe, Leuven, Belgium). All specimens incubated at 37°C and 100% humidity for 14 weeks and have been evaluated before the study and weekly for three months.

Color Evaluation Procedure

The evaluation procedure was done according to previous suggested model[33]. In brief, all teeth were polished with nonfluoridated and oil-free pumice, rinsed, and dried completely for 10 seconds. The reflectance spectrophotometer SpectroShade (LUA005, MHT Optic Research AG, Zurich, Switzerland; software version, 2.20) was used to evaluate to color alterations. According to a previous investigation, this instrument provides precise measurements during longitudinal evaluation of tooth color in vivo[34].

Color quantification was based on the CIE's L*, a*, and b* color system that uses 3 parameters to define color: the L* coordinate is a measure of lightness similar to value in the Munsell system[35] and ranges from 0 (black) to 100 (white); the a* and b* coordinates represent positions on red/green and yellow/blue axes, respectively. The readings of a* and b* combined provided the same information as hue and chroma in the Munsell system. The spectrophotometer was calibrated before each image-capture session with white and green ceramic tiles supplied by the manufacturer. The calibration process compensated for any deviation in the quantity of illumination output from the internal light source[36]. The spectrophotometric data of each tooth was recorded at 5 consecutive times by positioning, removing, and repositioning the intraoral camera on a black surface to prevent enamel-color alterations[37]. The resulting images were subsequently cross-referenced by using the built-in synchronized image program. Each tooth gone through spectrophotometric analysis of the crown area in its incisal, middle and cervical thirds of its buccal surface (Figure 1). All images were recorded by the same operator (HA).

The CIE color parameters (L*, a*, and b*) were measured and averaged for each material and the resultant color differences (ΔE) between the interval groups was calculated according to the following equation: $\Delta E=[(L * i - L * ii)2 + (a * i - a * ii)2 + (b * i - b * ii)2]1/2$. where i and ii represent the color measurements made before and after the treatment for different time period, respectively.

Statistical Analysis

Color assessment in relation to pulpotomy material, was made with 2-way mixed analysis of variance (ANOVA) for the DE differences and 2-way ANOVA for the time differences. It was found appropriate to analyze the data after applying the log transformation. For each area, mixed effects 2-way ANOVA nested model was fitted. Likelihood ratio test was then preformed to test each of the covariates effects. These statistical analyses were conducted with SPSS software (version 12.0, SPSS, Chicago, Ill., USA). The level of significance was set at $P < 0.05$.

RESULTS

The results relating to the ΔE of the materials over time are presented in Table 1 and Figure 2. The ΔE of all experimental groups (GMTA, WMTA and BD) were significantly different from the control group at all time points ($P<0.05$).

Color changes in the GMTA and WMTA groups, had no statistical significant differences, but both medicaments (GMTA, WMTA) showed significant higher discoloration compared to BD group in the cervical part of the crown, since week 1 ($P<0.05$).

WMTA group showed significant discoloration in the cervical part as of week 1 ($P<0.05$), and gradually increased over time till week 2 (Figure 2). BD group showed no significantly discoloration over time. GMTA group showed the significant discoloration at week 1 and week 14 ($P<0.05$). Figure 1 demonstrate dark discoloration that appears, mainly in the cervical part of the crown, after just 1 week (figure 1c) after medicament placement, which appears even more intense after 14 weeks (figure 1d).

DISCUSSION

This study investigated the potential of 3 bioceramics (GMTA, WMTA, and BD) to induce coronal discoloration in extracted primary teeth. The results indicate that two of the materials tested ,GMTA and WMTA, discolor tooth structure to a perceptible degree. BD though, showed no discoloration over time. Both GMTA and WMTA groups had significant discoloration in the first week and more overall color change over time. However, only GMTA showed another significate difference over time (at week 14).

The irrigation protocol in this study during canal preparation was NaOCl and a final irrigation with sterile saline to remove the smear layer. This protocol was chosen to facilitate the maximum penetration of the materials into the dentinal tubules. According to Davis et al[38], discoloration is less evident or takes longer when the smear layer is not removed, and materials in the canal penetrate more readily when the smear layer is removed. Irrigating with NaOCl has been shown to react with bismuth-containing materials causing a dark-brown discoloration[39], hence final saline rinse was made before pulpotomy material was placed in the chamber.

We used a spectrophotometer and the CIE L*a*b* space system to evaluate color change. The system approximates uniform distances between the color coordinates while covering the visual color space. Seghi et al[40] showed that a color change of 2 units of CIE L* a* b* was detected 100% of the time by observers, and 0.5–1 unit was detected 80% of the time, this finding may explain the result in our study, in which gray discoloration could be clearly seen in the GMTA group. The current study showed that all materials discolored to some degree. In contrast, the control group was below the perceptibility threshold at all time points, which lends credence to the validity of the findings.

Despite the harmonious color of WMTA with dental tissues, tooth discoloration was observed in the findings of this study, which was in accordance with the results of some other studies[11,16]. These findings limit applications of MTA in endodontic treatments such as pulp capping and pulpotomy in the esthetic zones[17]

In the present study, the color change was evident in the first week and increased through the third month; however, it remained stable.

Table 1: Changes in color (ΔE) expressed as mean ± standard deviation over 14 weeks.

	N	area	Immediately after placement	1 Week	2 Weeks	4 Weeks	8 Weeks	14 Weeks
Control	10	Cervical	0.39 ± 0.03[a]	0.37 ± 0.04[a]	0.39 ± 0.1[a]	0.41 ± 0.08[a]	0.44 ± 0.09[a]	0.36 ± 0.03[a]
		Middle	0.53 ± 0.14[a]	0.6 ± 0.08[a]	0.7 ± 0.05[a]	0.49 ± 0.02[a]	0.52 ± 0.07[a]	0.7 ± 0.1[a]
		Incisal	0.81 ± 0.08[a]	0.91 ± 0.07[a]	0.87 ± 0.06[a]	0.89 ± 0.08[a]	0.69 ± 0.06[a]	0.9 ± 0.03[a]
Biodentine	10	Cervical	1.86 ± 0.78	1.73 ± 1.02[b]	1.63 ± 0.69[b]	1.62 ± 0.56[b]	1.82 ± 0.63[b]	1.86 ± 0.7[b]
		Middle	2.17 ± 0.62	2.33 ± 0.63	2.47 ± 0.47	2.45 ± 0.55	2.23 ± 0.59	2.61 ± 0.76
		Incisal	2.33 ± 0.65	2.46 ± 0.84	2.36 ± 0.8	2.3 ± 0.76	2.27 ± 0.36	2.54 ± 0.56
GMTA	10	Cervical	2.09 ± 0.39	2.92 ± 0.62[c]	2.96 ± 0.33	2.82 ± 0.66	2.93 ± 0.32	3.48 ± 0.78[c]
		Middle	2.22 ± 0.67	2.22 ± 0.36	2.39 ± 0.36	2.05 ± 0.73	2.32 ± 0.57	2.41 ± 0.93
		Incisal	2.47 ± 0.28	2.61 ± 0.39	2.7 ± 0.29	2.08 ± 0.76	2.45 ± 0.34	2.55 ± 0.74
WMTA	10	Cervical	1.53 ± 0.58	2.46 ± 0.69[c]	2.85 ± 0.4	2.78 ± 0.46	2.91 ± 0.5	2.87 ± 0.58
		Middle	1.85 ± 0.43	2.02 ± 0.63	2.1 ± 0.41	2.1 ± 0.49	2.3 ± 0.61	2.09 ± 0.6
		Incisal	2.12 ± 0.79	1.98 ± 0.86	2.09 ± 0.5	1.95 ± 0.85	2.25 ± 0.67	2.05 ± 0.53

Within each time point, groups displaying different superscript letters indicate a significant difference. a - difference between the control to all other groups. b - difference between material and c - differences within groups in different time stops.

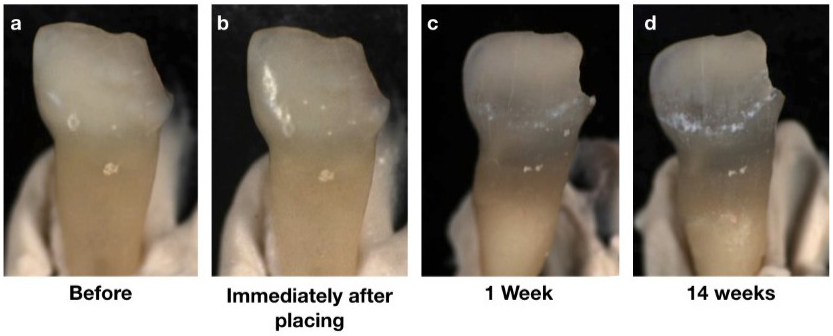

Figure 1: In the figure, central primary incisor, before treatment (a) and immediately after the medicament placement (b). Dark discoloration was noticed, mainly in the cervical part of the crown, after just 1 week (c), which appeared even more intense after 14 weeks (d).

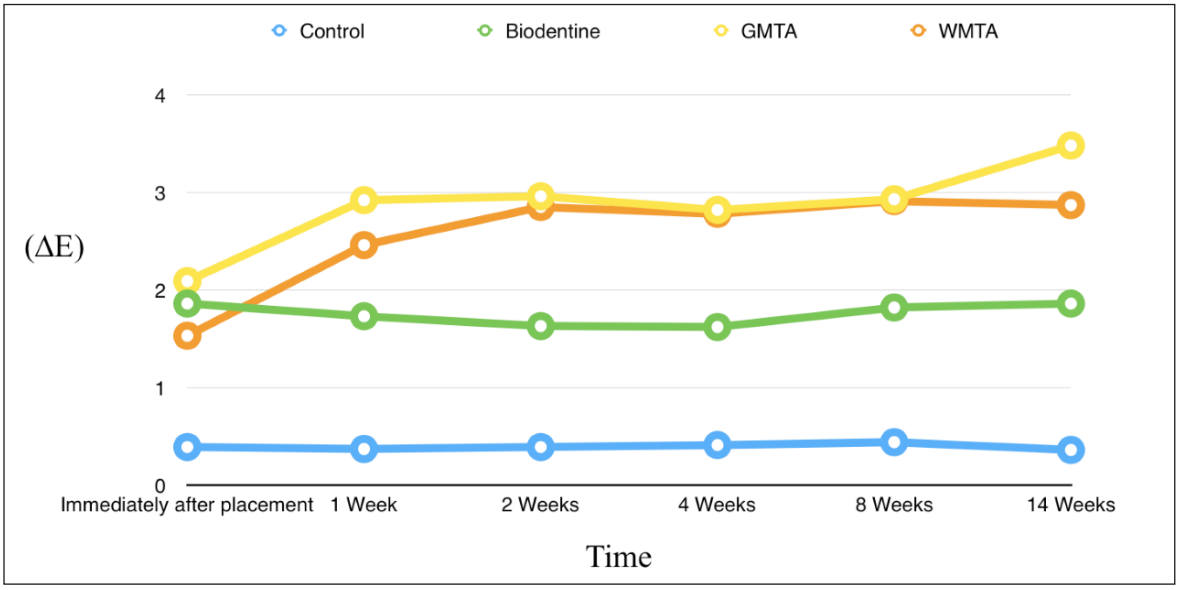

Figure 2: Color changes expressed as mean ± standard deviation over 14 weeks. The ΔE of all experimental groups were significantly different from the control group at all time points. Color changes in the GMTA and WMTA groups, showed significantly higher discoloration compared to BD group in the cervical part of the crown, since week 1 to week 14.

Variable amounts and durations of color change was reported with the same formulation of MTA, which would be the result of different thickness of the remaining tooth structure, colorimetric method of color measurement and material application methods[16,20,38]. The mentioned metal oxides, are present in the tooth-colored formula (WMTA) although to a very low degree, and can induce tooth discoloration[16]. Elements such as Fe, Mn and Cu, with d-electrons, are well known to have strong colors in oxide form. The d-electrons are readily excited by a visible spectrum light. Other oxides without such electrons (Ca, Si, Al, Mg and S) tend to be colorless or white, but the heavy ones such as bismuth has a yellow oxide[41]. Some authors state that the yellow oxide of bismuth used in both formulations of MTA for radiopacity, is the significant factor for tooth discoloration[21].

Research indicates that the most pronounced staining from dental materials occurs in the cervical third of the crown[42,43], which is why the area of measurement for this study was confined to the cervical third. During the experiments, it was observed that the WMTA and GMTA groups showed gray discoloration of the cervical root dentin, whereas the BD group had no discernible color change in the corresponding region. This finding is more in alignment with the current body of research that has shown MTA to cause gray discoloration of tooth structure[16,42,44] To the best of our knowledge, no previous research has shown BD to discolor tooth structure[39].

Many of the clinical applications of bioceramics involve proximity to and incorporation of blood. Studies have shown that the presence of blood compounds the staining propensity of gray or white MTA[11,42], and blood alone also has potent staining ability[45,46]. This study lacks in this matter, because no blood contamination was tested which may not reflect the clinical procedure in vivo. To evaluate the intrinsic staining potential of the materials with blood contamination, future studies are required.

We conclude that both GMTA and WMTA pulpotomy material may discolor tooth structure over time in an extracted primary anterior tooth model. When choosing bioceramic pulpotomy material, BD may preferable, mainly in esthetic area.

REFERENCES

1. Fuks AB. Current concepts in vital primary pulp therapy. Eur J Paediatr Dent. 2002;3(3):115-120. http://www.ncbi.nlm.nih.gov/pubmed/12870999. Accessed September 23, 2018.
2. AMERICAN ACADEMY OF PEDIATRIC DENTISTRY CLINICAL PRACTICE GUIDELINES 245. http://www.aapd.org/media/policies_guidelines/g_pulp.pdf. Accessed September 23, 2018.
3. Markovic D, Zivojinovic V, Vucetic M. Evaluation of three pulpotomy medicaments in primary teeth. Eur J Paediatr Dent ;6(3):133. 2005.
4. Vargas KG, Fuks AB, Peretz B. Pulpotomy Techniques: Cervical (Traditional) and Partial. In: Pediatric Endodontics. Cham: Springer International Publishing; 2016:51-70. doi:10.1007/978-3-319-27553-6_5
5. Ranly DM. Assessment of the systemic distribution and toxicity of formaldehyde following pulpotomy treatment: Part one. ASDC J Dent Child. 1985;52(6):431-434. http://www.ncbi.nlm.nih.gov/pubmed/3864800. Accessed November 27, 2019.
6. Praveen KN, Rashmi N, Vipin BK, Pujan MP. Pulpotomy Medicaments: Continued Search for New Alternatives- A Review. https://pdfs.semanticscholar.org/4116/5c5c4386eff7dad9b9680d7099d92ddd58e0.pdf. Accessed November 27, 2019.
7. Vargas KG, Packham B. Radiographic success of ferric sulfate and formocresol pulpotomies in relation to early exfoliation. Pediatr Dent. 27(3):233-237. http://www.ncbi.nlm.nih.gov/pubmed/16173229. Accessed November 27, 2019.
8. Smaïl-Faugeron V, Courson F, Durieux P, Muller-Bolla M, Glenny A-M, Fron Chabouis H. Pulp treatment for extensive decay in primary teeth. Cochrane Database Syst Rev. 2014;(8):CD003220. doi:10.1002/14651858.CD003220.pub2
9. HOLAN, G. Introduction to dental trauma : Managing traumatic injuries in the primary dentition. Pediatr Dent Infancy Through Adolesc. 2005:236-256. https://ci.nii.ac.jp/naid/10030290004/. Accessed September 23, 2018.
10. Cvek M. A clinical report on partial pulpotomy and capping with calcium hydroxide in permanent incisors with complicated crown fracture. J Endod. 1978;4(8):232-237. doi:10.1016/S0099-2399(78)80153-8
11. Lenherr P, Allgayer N, Weiger R, Filippi A, Attin T, Krastl G. Tooth discoloration induced by endodontic materials: a laboratory study. Int Endod J. 2012;45(10):942-949. doi:10.1111/j.1365-2591.2012.02053.x
12. van der Burgt TP, Mullaney TP, Plasschaert AJM. Tooth discoloration induced by endodontic sealers. Oral Surgery, Oral Med Oral Pathol. 1986;61(1):84-89. doi:10.1016/0030-4220(86)90208-2
13. Asgary S, Akbari Kamrani F, Taheri S. Evaluation of antimicrobial effect of MTA, calcium hydroxide, and CEM cement. Iran Endod J. 2007;2(3):105-109. http://www.ncbi.nlm.nih.gov/pubmed/24298290. Accessed July 28, 2018.
14. Torabinejad M, Hong CU, McDonald F, Pitt Ford TR. Physical and chemical properties of a new root-end filling material. J Endod. 1995;21(7):349-353. doi:10.1016/S0099-2399(06)80967-2
15. Belobrov I, Parashos P. Treatment of Tooth Discoloration after the Use of White Mineral Trioxide Aggregate. J Endod. 2011;37(7):1017-1020. doi:10.1016/J.JOEN.2011.04.003
16. Ioannidis K, Mistakidis I, Beltes P, Karagiannis V. Spectrophotometric analysis of coronal discolouration induced by grey and white MTA. Int Endod J. 2013;46(2):137-144. doi:10.1111/j.1365-2591.2012.02098.x
17. Akbari M, Rouhani A, Samiee S, Jafarzadeh H. Effect of dentin bonding agent on the prevention of tooth discoloration produced by mineral trioxide aggregate. Int J Dent. 2012;2012:563203. doi:10.1155/2012/563203
18. Song J-S, Mante FK, Romanow WJ, Kim S. Chemical analysis of powder and set forms of Portland cement, gray ProRoot MTA, white ProRoot MTA, and gray MTA-Angelus. Oral Surgery, Oral Med Oral Pathol Oral Radiol Endodontology. 2006;102(6):809-815. doi:10.1016/j.tripleo.2005.11.034
19. Maroto M, Barberia E, Planells P, Vera V. Treatment of a non-vital immature incisor with mineral trioxide aggregate (MTA). Dent Traumatol. 2003;19(3):165-169. doi:10.1034/j.1600-9657.2003.00106.x
20. Watts JD, Holt DM, Beeson TJ, Kirkpatrick TC, Rutledge RE. Effects of pH and Mixing Agents on the Temporal Setting of Tooth-colored and Gray Mineral Trioxide Aggregate. J Endod. 2007;33(8):970-973. doi:10.1016/J.JOEN.2007.01.024

21. Steffen R, van Waes H. Understanding mineral trioxide aggregate/Portland-cement: A review of literature and background factors. Eur Arch Paediatr Dent. 2009;10(2):93-97. doi:10.1007/BF03321608
22. Hwang Y-C, Kim D-H, Hwang I-N, et al. Chemical Constitution, Physical Properties, and Biocompatibility of Experimentally Manufactured Portland Cement. J Endod. 2011;37(1):58-62. doi:10.1016/J.JOEN.2010.09.004
23. Han L, Okiji T. Bioactivity evaluation of three calcium silicate-based endodontic materials. Int Endod J. 2013;46(9):808-814. doi:10.1111/iej.12062
24. Shokouhinejad N, Nekoofar MH, Razmi H, et al. Bioactivity of EndoSequence Root Repair Material and Bioaggregate. Int Endod J. 2012;45(12):1127-1134. doi:10.1111/j.1365-2591.2012.02083.x
25. Zhou H, Shen Y, Wang Z, et al. In Vitro Cytotoxicity Evaluation of a Novel Root Repair Material. J Endod. 2013;39(4):478-483. doi:10.1016/j.joen.2012.11.026
26. AlAnezi AZ, Jiang J, Safavi KE, Spangberg LSW, Zhu Q. Cytotoxicity evaluation of endosequence root repair material. Oral Surgery, Oral Med Oral Pathol Oral Radiol Endodontology. 2010;109(3):e122-e125. doi:10.1016/J.TRIPLEO.2009.11.028
27. Butt N, Bali A, Talwar S, Chaudhry S, Nawal R, Yadav S. Comparison of physical and mechanical properties of mineral trioxide aggregate and Biodentine. Indian J Dent Res. 2014;25(6):692. doi:10.4103/0970-9290.152163
28. Camilleri J. Investigation of Biodentine as dentine replacement material. J Dent. 2013;41(7):600-610. doi:10.1016/j.jdent.2013.05.003
29. Flores-Mir C, Silva E, Barriga MI, Lagravère MO, Major PW. Lay person's perception of smile aesthetics in dental and facial views. J Orthod. 2004;31(3):204-209. doi:10.1179/146531204225022416
30. Cortes MI de S, Marcenes W, Sheiham A. Impact of traumatic injuries to the permanent teeth on the oral health-related quality of life in 12-14-year-old children. Community Dent Oral Epidemiol. 2002;30(3):193-198. http://www.ncbi.nlm.nih.gov/pubmed/12000342. Accessed September 23, 2018.
31. Holan G, Needleman HL. Premature loss of primary anterior teeth due to trauma–potential short- and long-term sequelae. Dent Traumatol. 2014;30(2):100-106. doi:10.1111/edt.12081
32. Gherunpong S, Tsakos G, Sheiham A. The prevalence and severity of oral impacts on daily performances in Thai primary school children. Health Qual Life Outcomes. 2004;2(1):57. doi:10.1186/1477-7525-2-57
33. Karamouzos A, Athanasiou AE, Papadopoulos MA, Kolokithas G. Tooth-color assessment after orthodontic treatment: A prospective clinical trial. Am J Orthod Dentofac Orthop. 2010;138(5):537.e1-537.e8. doi:10.1016/J.AJODO.2010.03.026
34. Karamouzos A, Papadopoulos MA, Kolokithas G, Athanasiou AE. Precision of in vivo spectrophotometric colour evaluation of natural teeth. J Oral Rehabil. 2007;34(8):613-621. doi:10.1111/j.1365-2842.2007.01744.x
35. Nickerson D. Munsell Color System, Company, and Foundation.; 1976. https://munsell.com/wp-content/uploads/2011/03/munsell-color-history-dorothy-nickerson.pdf. Accessed September 23, 2018.
36. Chu SJ. Use of a reflectance spectrophotometer in evaluating shade change resulting from tooth-whitening products. J Esthet Restor Dent. 2003;15 Suppl 1:S42-8. http://www.ncbi.nlm.nih.gov/pubmed/15000904. Accessed September 23, 2018.
37. Russell MD, Gulfraz M, Moss BW. In vivo measurement of colour changes in natural teeth. J Oral Rehabil. 2000;27(9):786-792. http://www.ncbi.nlm.nih.gov/pubmed/11012854. Accessed September 23, 2018.
38. Davis MC, Walton RE, Rivera EM. Sealer Distribution in Coronal Dentin. J Endod. 2002;28(6):464-466. doi:10.1097/00004770-200206000-00012
39. Camilleri J. Color Stability of White Mineral Trioxide Aggregate in Contact with Hypochlorite Solution. J Endod. 2014;40(3):436-440. doi:10.1016/j.joen.2013.09.040
40. Seghi RR, Hewlett ER, Kim J. Visual and Instrumental Colorimetric Assessments of Small Color Differences on Translucent Dental Porcelain. J Dent Res. 1989;68(12):1760-1764. doi:10.1177/00220345890680120801
41. Asgary S, Eghbal MJ, Parirokh M, Ghoddusi J, Kheirieh S, Brink F. Comparison of Mineral Trioxide Aggregate's Composition with Portland Cements and a New Endodontic Cement. J Endod. 2009;35(2):243-250. doi:10.1016/j.joen.2008.10.026
42. Felman D, Parashos P. Coronal Tooth Discoloration and White Mineral Trioxide Aggregate. J Endod. 2013;39(4):484-487. doi:10.1016/j.joen.2012.11.053
43. Karabucak B, Li D, Lim J, Iqbal M. Vital pulp therapy with mineral trioxide aggregate. Dent Traumatol. 2005;21(4):240-243. doi:10.1111/j.1600-9657.2005.00306.x
44. Jang J-H, Kang M, Ahn S, et al. Tooth Discoloration after the Use of New Pozzolan Cement (Endocem) and Mineral Trioxide Aggregate and the Effects of Internal Bleaching. J Endod. 2013;39(12):1598-1602. doi:10.1016/J.JOEN.2013.08.035
45. Freccia WF, Peters DD. A technique for staining extracted teeth: a research and teaching aid for bleaching. J Endod. 1982;8(2):67-69. doi:10.1016/S0099-2399(82)80260-4
46. Marin PD, Bartold PM, Heithersay GS. Tooth discoloration by blood: an in vitro histochemical study. Dent Traumatol. 1997;13(3):132-138. doi:10.1111/j.1600-9657.1997.tb00026.x