Dental Traumatology

Dental Traumatology 2013; **29**: 383–388; doi: 10.1111/edt.12011

Staining susceptibility of new calcium aluminate cement (EndoBinder) in teeth: a 1-year *in vitro* study

Lucas da Fonseca Roberi Garcia[1], Fabiano Gamero Aguilar[1], Hebert Luis Rossetto[2], Marcio Gomes Sabino[2], Fernanda de Carvalho Panzeri Pires-de-Souza[1]

[1]Department of Dental Materials and Prosthodontics, Ribeirão Preto School of Dentistry, University of São Paulo, Ribeirão Preto, SP, Brazil; [2]Binderware Biomaterials Ltd., São Carlos, SP, Brazil

Key words: calcium aluminate cement; color stability; mineral trioxide aggregate; staining susceptibility

Correspondence to: Prof. Dr. Fernanda de Carvalho Panzeri Pires-de-Souza, Departamento de Materiais Dentários e Prótese, Faculdade de Odontologia de Ribeirão Preto, USP, Avenida do Café, s/n, Bairro Monte Alegre, Ribeirão Preto, São Paulo CEP: 14040-904, Brazil
Tel.: +55 16 3602 3973
Fax: +55 16 3633 0999
e-mail: ferpanzeri@usp.br

Accepted 2 August, 2012

Abstract – *Aim*: To evaluate the staining susceptibility of new calcium aluminate cement (EndoBinder, EB) in teeth, with or without radiopacifying agent, in comparison with mineral trioxide aggregate (MTA). *Materials and methods*: Forty bovine incisors were used. After biomechanical preparation and filling, 2 mm of their fillings were removed for cervical plug fabrication ($n = 10$): EB without radiopacifier; EndoBinder + Bismuth Oxide (EBBO); Gray mineral trioxide aggregate (GMTA) and White mineral trioxide aggregate (WMTA). After teeth restoration, initial color readout was taken (Easyshade – CIE Lab) on the vestibular face. The teeth were stored in artificial saliva at 37°C for 1 year, and after time intervals of 30, 180, and 360 days, new color readouts were taken to determine color alteration (ΔE) in comparison with the initial readout (baseline). *Results*: The results (2-way ANOVA repeated measures, Bonferroni – $P < 0.05$) demonstrated that after 360 days, all groups presented ΔE above the clinically accepted limit ($\Delta E \geq 3.3$), however, without significant difference among them ($P > 0.05$). All groups presented decrease in ΔL values over the course of time, and the greatest variation occurred for WMTA, with significant difference in comparison with EB and EBBO ($P < 0.05$). *Conclusions*: Some negative features of MTA, such as the high incidence of dental structures staining justify the development of new materials. GMTA, WMTA, and EB with and without radiopacifying agent displayed color alteration (ΔE) after 360 days; however, the luminosity change (ΔL) was lower in EB in comparison with GMTA and WMTA.

Certain procedures performed in endodontic therapy, such as apexifications, pulpotomies, paraendodontic surgeries, treatment of internal resorptions, and root and furcal perforations require specific sealing cement to obtain successful treatment (1). Mineral trioxide aggregate (MTA) was initially developed as a material for retrograde filling and root and furcal perforations treatment (2); and due to its good performance, it has been used in various other applications, mainly in dental traumatology (3–8).

Mineral trioxide aggregate is basically composed of Portland cement (% by weight) (75.0), Bi_2O_3 (20.0), and dehydrated $CaSO_4$ (5.0) (9). Portland cement consists of SiO_2 (21.2), CaO (68.1), Al_2O_3 (4.7), MgO (0.48), and Fe_2O_3 (1.89) which, when water is added, forms a hydrated calcium silicate paste (10).

Despite the good performance of MTA as a sealing cement (7), certain of the negative features of the material must be considered, such as the low compressive strength (11); long setting time (12); handling characteristics (13); low flow ability (2); high solubility in a humid environment (14); presence and release of arsenic (15) and high incidence of staining dental structures (16, 17).

Initially, MTA was available commercially only in the gray form. The high concentration of iron oxide in its composition, responsible for the dark color of the material, limited its application to posterior teeth due to staining caused by the material in the dental tissues (10, 17). To solve this deficiency, a new version of MTA was developed, with a low iron oxide concentration. However, some studies have pointed out that this version also caused darkening of dental tissues (9, 16, 17).

These deficiencies justify the development of new materials with adequate biologic and physicomechanical properties (14). Thus, a new calcium aluminate-based cement (Patent Number PI0704502-6, 2007) called EndoBinder (Binderware, São Carlos, SP, Brazil) was developed to maintain the good performance of MTA, but without its negative features. EndoBinder is produced by the process of Al_2O_3 and $CaCO_3$ calcination at temperatures ranging from 1315 to 1425°C (18). The calcium aluminate formed is cooled and then triturated until an adequate particle size is obtained. This process of synthesis used to obtain EndoBinder provides a material with advantages in relation to MTA, among them, the control of the levels of impurities such as Fe_2O_3, which causes tooth darkening (16). As a radiopacifying agent, bismuth oxide (20% by weight) is added to the cement, to promote adequate radiopacity (19), in accordance with specification ISO 6876 (20).

Thus, the aim of this *in vitro* study was to evaluate the staining susceptibility of new calcium aluminate cement (EndoBinder) in teeth, with or without the addition of a radiopacifying agent (bismuth oxide), in comparison with Gray and White MTA (GMTA and WMTA). The null hypothesis tested was that there would be no difference in the staining ability of the tested cements, irrespective of the presence or absence of the radiopacifying agent in EndoBinder.

Material and methods

Sample preparation – teeth filling

Forty healthy bovine incisors were used, with root length standardized at 20 mm. Before biomechanical preparation, the teeth were immersed in a 0.5% chloramine solution for 48 h for disinfection, and then, washed in running water for 24 h. After this, they were radiographed to evaluate the root canals. Access cavity was performed on the palatine face of the teeth with spherical carbide bur no. 5 (KG Sorensen, São Paulo, SP, Brazil), mounted at high-speed appliance (Silent – MRS 400; Dabi Atlante, Ribeirão Preto, SP, Brazil), and finishing and divergence of cavity walls with Endo Z bur (Dentsply/Maillefer, Tulsa, OK, USA). The root canal was abundantly irrigated with 1% sodium hypochlorite solution and the working length determined by inserting a K-type file (Dentsply/Maillefer) compatible with the root canal diameter until it was visualized in the apical foramen and then withdrawn 1.0 mm. Due to the great root canal size, it was debrided only with Gates-Glidden burs #6, 5, 4, and 3 consecutively (Dentsply/Maillefer) mounted at low-speed appliance (MRS 400 – Dabi Atlante), respecting the working length. Irrigation was performed with 2.5 ml of 1% sodium hypochlorite at each change of instrument, drying with absorbent paper cones (Dentsply/Maillefer) and the teeth were filled with gutta-percha points (Dentsply/Maillefer) and Sealer 26 filling cement (Dentsply, Petrópolis, RJ, Brazil) by the classic lateral condensation technique (21). After 7 days (period equivalent to three times the setting time of the filling cement, as recommended by the manufacturer), 2 mm of the filling were removed with a preheated Hollenback 3S instrument (SS White/Duflex, Rio de Janeiro, RJ, Brazil) and remnants of Sealer 26 were cleaned out from the access cavity using 92.6% alcohol. The teeth were then randomly separated into four groups (n = 10) according to the type of cement used as cervical plug, as described in Table 1.

The cements were manipulated in accordance with the manufacturers' recommendations, with the proportion of 1 g of powder to 0.21 ml of distilled water being used for EndoBinder; and for MTA, one dose of powder to one drop of distilled water (18).

Table 1. Cements used in the study

Groups	Materials	Manufacturer
EB	EndoBinder without radiopacifying agent	Binderware, São Carlos, SP, Brazil
EBBO	EndoBinder + 20% (weight) Bi_2O_3	
GMTA	MTA Gray	Ângelus, Londrina, PR, Brazil
WMTA	MTA White	

MTA, mineral trioxide aggregate; EB, EndoBinder; EBBO, EndoBinder + Bismuth Oxide; GMTA, gray mineral trioxide aggregate; WMTA, white mineral trioxide aggregate.

After fabricating the cervical plug, the teeth were restored with composite Z250 (A3 shade – 3M ESPE Dental Products, St. Paul, MN, USA) according to the incremental technique, using a two-step adhesive system with prior acid etching (Adper Single Bond 2; 3M ESPE Dental Products) and light activation with a LED-type appliance (FlashLite 1401, Discus Dental – Culver City, CA, USA – light intensity $\geq$ 1100 mW cm^{-2} and wavelength in the band between 460 and 480 nm), for 20 s. After this, the samples were photographed (Canon EOS, Digital Rebel XTi, Lake Success, NY, USA) for comparison of the images before and after 360 days.

Color analysis

The initial color readout was performed in the center of the cervical region on the vestibular face of each tooth, using Spectrophotometer Easyshade (VITA Zahnfabrik, Bad Säckingen, Germany), a portable appliance with a digital pointer capable of reading color numerically (22). The observation pattern simulated by the Spectrophotometer followed the CIE $L^*a^*b^*$ system, recommended by CIE (Commission Internationale de l'Éclairage). This consists of two axes, a^* and b^* that have right angles and represent the dimension of tonality or color. The third axis is luminosity L^*, which represents the amount of light reflected from the object, and it is expressed in numeric values ranging from 0 (black) to 100 (white). As closer to zero, the darker the color of the object. In turn, lighter objects present numeric values close to 100. L^* axis is perpendicular to the plane a^*b^*. With this system, colors can be specified with the coordinates L^*, a^*, and b^*.

After the initial color readout, the teeth were stored in artificial saliva and kept in an oven at 37°C throughout the period of 1 year. Every 7 days, the saliva was replaced with fresh solution. After time intervals of 30, 180, and 360 days, new color readouts were performed for comparison with the values obtained in the initial readout (baseline).

The means for each coordinate were calculated and the color alteration (ΔE) between the values obtained in the initial and subsequent readouts was calculated with the use of the following formula: $\Delta E = [(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2]^{1/2}$, where, ΔE is the color alteration; $\Delta L = L_{Final} - L_{Initial}$; $\Delta a = a_{Final} - a_{Initial}$; $\Delta b = b_{Final} - b_{Initial}$; ΔL represents the difference in luminosity. Positive ΔL values mean the sample is lighter than the standard (baseline readout), and negative values mean the sample is darker than the standard. Δa represents the difference in the red–green parameter ($-a^*$ = green and $+a^*$ = red) and Δb the difference in the yellow–blue parameter ($-b^*$ = blue and $+b^*$ = yellow) (23). The values obtained were submitted to the normality test (Kolmogorov–Smirnov) and statistical analysis (2-way ANOVA repeated measures, Bonferroni's test at a level of significance of 5%).

Results

The values obtained for ΔL and their comparison are shown in Table 2. When comparing the values of the various groups, it could be observed that all the samples presented decrease in ΔL values over the course of time, which means loss of luminosity. This loss of luminosity may be interpreted as a trend toward darkening of the tooth, because the negative values for ΔL represent a trend toward black. At 360 days, EBBO presented the least variation in ΔL, with statistically significant difference in comparison with GMTA and WMTA, which presented the greatest variation ($P < 0.05$). Samples of groups GMTA and WMTA, after 360 days, presented staining in the cervical region, as can be observed in Fig. 1c,d.

The values obtained for ΔE and their comparison are shown in Table 3. Over the course of the time of analysis, an increase in the ΔE values was observed for all groups. At 180 days, EB presented the highest ΔE value, however, with the other groups also presenting ΔE values above 3.0. Whereas in the final period of analysis, WMTA presented the highest ΔE value, followed by EB, however, without statistically significant difference in comparison with the other groups ($P > 0.05$).

Table 2. ΔL values for groups and periods

Groups	Periods: 30 days	180 days	360 days
EB			
Median	1.05[a]	−1.32[a]	−3.47[ab]
Maximum	6.80	6.30	4.50
Minimum	−3.40	−7.40	−7.70
EBBO			
Median	−1.85[a]	−0.21[a]	−1.85[a]
Maximum	1.70	7.20	7.90
Minimum	−3.20	−5.80	−6.20
GMTA			
Median	−1.75[a]	−3.60[a]	−6.25[bc]
Maximum	1.20	2.20	−2.10
Minimum	−4.80	−9.40	−11.60
WMTA			
Median	−1.80[a]	−5.21[b]	−7.50[c]
Maximum	2.20	1.30	−3.20
Minimum	−4.30	−8.80	−16.10

EB, EndoBinder; EBBO, EndoBinder + Bismuth Oxide; GMTA, gray mineral trioxide aggregate; WMTA, white mineral trioxide aggregate.
Different letters in the columns indicate statistically significant difference (2-way ANOVA repeated measures, Bonferroni – $P < 0.05$).

Discussion

This study evaluated the staining ability of new calcium aluminate cement (EndoBinder) in teeth, with or without the addition of a radiopacifying agent (bismuth oxide), in comparison with Gray and White MTA. The staining ability of MTA is known (16, 17), despite its qualities (7). However, few studies in literature quantify this color change. Thus, this study compares the color alteration after use of MTA, in its two commercial forms, with a new calcium aluminate-based cement with potentially beneficial properties (18).

The dental structure staining of MTA is related to its gray color, and in an endeavor to solve this problem, the manufacturer developed a white version of the material (17). Both commercial forms of the material have similar compositions, however, with lower concentrations of Al_2O_3, MgO, and mainly FeO in the white version (10).

Nevertheless, *in vivo* studies have reported cases of dental structures staining when perforations were treated with WMTA (16, 24), a result similar to that found in this *in vitro* study. The hypothesis that not only iron oxide but also other constituents of MTA, such as bismuth oxide, used as radiopacifying agent may cause staining have been questioned (16,17,24), but not proved yet. Also, some manufacturers have reported that the use of pure Portland cement (without radiopacifying agent) provides better color stability than the commercial form of MTA. However, there are no studies which confirm such information (17).

Studies have demonstrated that human and bovine enamel are very similar in their composition, morphology, and physical properties, thereby bovine teeth have been used continuously to assess various properties of human enamel (25). In the same manner, the diameter and number of dentinal tubules per mm^2 have been reported to be similar for human molars and bovine incisors (25). Furthermore, the permeability of bovine dentin close to the cervical area has also been shown similar to coronal human dentin, allowing its use in this *in vitro* study (25).

It is known that the bismuth oxide present in MTA is not inert, as it affects its hydration mechanism, making its microstructure more fragile than that of Portland cement (26). The induction by MTA of reparative tissue and dentin bridges formation in exposed pulps is also reduced, due to the presence of bismuth oxide (27). This compound becomes part of the hydrated phase of the material, forming a structure composed by hydrated bismuth calcium silicate, ettringite, and monosulphate, which are leached with the calcium hydroxide formed from the hydration of calcium silicate (28). This process reduces the precipitation of calcium hydroxide in the hydrated paste, altering the reparative ability and decreasing physicochemical properties of MTA (28).

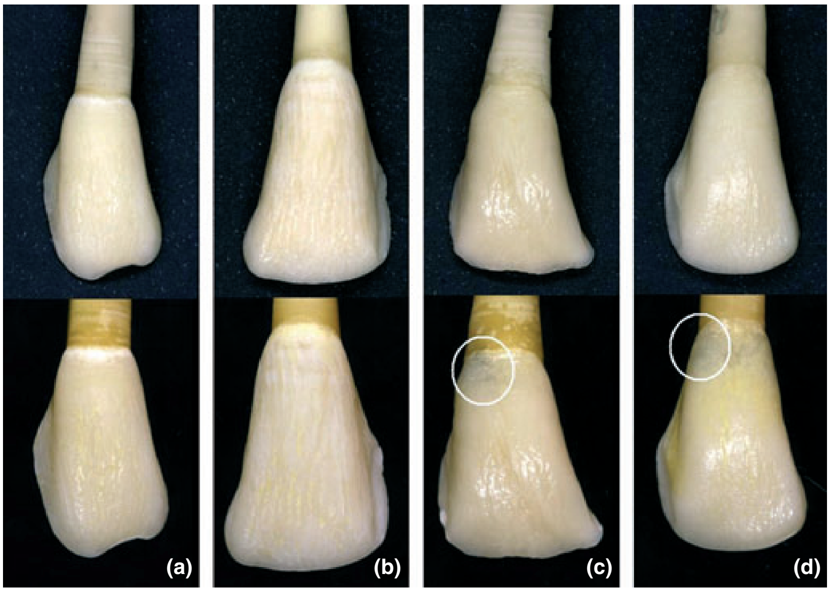

Fig. 1. Color alteration observed in the samples of the groups evaluated before (above) and after 360 days of analysis (below). (a) EndoBinder and (b) EndoBinder + Bismuth Oxide – cervical region close to the area of application of the cement without signs of staining; (c) Gray mineral trioxide aggregate (MTA) and (d) White MTA – staining in the cervical region (indication).

Table 3. ΔE values for groups and periods

	Per'		
Groups	30	180 days	360 days
EB			
Median	4.50[a]	7.67[a]	7.27[a]
Maximum	11.28	14.75	15.22
Minimum	0.91	2.24	4.20
EBBO			
Median	2.84[a]	6.33[a]	7.10[a]
Maximum	5.56	9.24	12.46
Minimum	1.74	2.96	4.68
GMTA			
Median	3.15[a]	5.83[a]	8.40[a]
Maximum	5.01	9.49	11.74
Minimum	1.39	3.81	2.71
WMTA			
Median	3.02[a]	6.02[b]	9.36[a]
Maximum	5.39	13.34	17.83
Minimum	0.66	3.36	5.92

EB, EndoBinder; EBBO, EndoBinder + Bismuth Oxide; GMTA, gray mineral trioxide aggregate; WMTA, white mineral trioxide aggregate.
Different letters in the columns indicate statistically significant difference (2-way ANOVA repeated measures, Bonferroni – $P < 0.05$).

Furthermore, the biocompatibility of bismuth oxide has been questioned, due to its degradation, which raises the levels of this compound in tissues adjacent to restorative materials, although its action on human cells is not completely understood up to now (29). However, recent studies have demonstrated that MTA, with the addition of bismuth oxide is biocompatible, proving that its use as radiopacifying agent is safe (9, 30).

Bismuth oxide is generally used as radiopacifying agent due to its high atomic number, providing endodontic cements with radiopacity values higher than that equivalent to the Al scale (31). In a recent study, Aguilar et al. (19) evaluated the radiopacity of EndoBinder associated with 20% by weight of different radiopacifiers and demonstrated that bismuth oxide presented better performance than zinc oxide (ZnO) and zirconium oxide (ZrO_2). This property is important in allowing evaluation of the quality of root canal filling, detection of perforations, and follow up of internal resorptions, differentiation between adjacent anatomic structures (enamel and dentin), periapical lesions, and recurrent caries (32).

The results of this study demonstrate that the staining caused by MTA is not related to bismuth oxide, because EndoBinder with and without radiopacifying agent presented a similar performance between them.

All groups evaluated in this study presented color alteration above the clinically acceptable limit ($\Delta E \geq 3.3$) after 360 days (33); result similar to that found by Lenherr et al. (34) in a recent study. When ΔL was evaluated separately, it could be observed that the groups GMTA and WMTA presented greater loss of luminosity, which represents greater darkening of the teeth. Considering clinical parameters, loss of luminosity (darkening) may be interpreted as dental structures staining, a fact confirmed in the present study, because samples of groups GMTA and WMTA presented signs of staining in the cervical region, area close to the site of application of the cements (Fig. 1), an alteration not presented in the groups EB and EBBO.

In spite of the lower concentration of iron oxide, WMTA presented the highest loss of luminosity (darkening) at 360 days than the other groups, demonstrating that the difference in the concentrations of this compound between the two commercial forms of MTA is not the determinant factor in the staining ability of the material (16,17,24). WMTA was introduced into the market as low-iron concentration cement; nevertheless, according to Camilleri and Pitt Ford (9), the cement also promotes tooth darkening due to iron content oxidation, resulting in tetracalcium aluminoferrite, which indicates that not only the composition of the cements, but also the form of obtaining and sintering them is related to their ability to stain dental structures.

Calcium aluminate-based cements, such as EndoBinder, are composed of three main phases, which in turn are responsible for its hydraulic setting process: the anhydrous phase CA ($CaO.Al_2O_3$), ranging from 40% to 70% of the material; the CA_2 ($CaO.2Al_2O_3$) phase (>25%), and the $C_{12}A_7$ ($12CaO.7Al_2O_3$) phase (10%) (18). The sinterization could be described by the following chemical reaction: $CaCO_3 + Al_2O_3 = Ca(AlO_2)_2 + CO_2$. In this reaction, the crystalline phases of aluminate with high Ca content are formed. As the furnace temperature increases, CaO and Al_2O_3 react with the compounds formed in the beginning of the sinterization process, producing phases with a low Ca content (18). Under controlled conditions, it is possible to obtain materials with high levels of calcium aluminate purity, removing traces of free MgO and CaO, responsible for the expansion of the cement in contact with water (1) and Fe_2O_3, responsible for tooth staining, as observed in both commercial forms of MTA (16).

Moreover, Namazikhah et al. (35) reported that the microstructure of Portland-based cements, such as MTA, present pH-dependent porosities, which may uptake staining substances and be responsible for dental structure staining.

Within the limitations of this *in vitro* study, the null hypothesis must be rejected, because GMTA, WMTA, and EndoBinder with and without radiopacifying agent displayed color alteration (ΔE) after 360 days aging. However, the luminosity change (ΔL) was lower in EndoBinder group in comparison with GMTA and WMTA groups.

Acknowledgement

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

References

1. Ørstavik D, Nordahl I, Tibballs JE. Dimensional change following setting of root canal sealer materials. Dent Mater 2001;17:512–9.
2. Torabinejad M, Chivian N. Clinical applications of mineral trioxide aggregate. J Endod 1999;25:197–205.
3. Menezes R, Bramante CM, Letra A, Carvalho VG, Garcia RB. Histologic evaluation of pulpotomies in dog using two types of mineral trioxide aggregate and regular and white Portland cements as wound dressings. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2004;98:376–9.
4. Simon S, Rilliard F, Berdal A, Machtou P. The use of mineral trioxide aggregate in one-visit apexification treatment: a prospective study. Int Endod J 2007;40:186–97.
5. Hsien HC, Cheng YA, Lee YL, Lan WH, Lin CP. Repair of perforating internal resorption with mineral trioxide aggregate: a case report. J Endod 2003;29:538–9.
6. Accorinte MdeL, Holland R, Reis A, Bortoluzzi MC, Murata SS, Dezan E Jr et al. Evaluation of mineral trioxide aggregate and calcium hydroxide cement as pulp-capping agents in human teeth. J Endod 2008;34:1–6.
7. Parirokh M, Torabinejad M. Mineral trioxide aggregate: a comprehensive literature review-Part I: chemical, physical, and antibacterial properties. J Endod 2010;36:16–27.
8. Orosco FA, Bramante CM, Garcia RB, Bernardineli N, de Moraes IG. Sealing ability, marginal adaptation and their correlation using three root-end filling materials as apical plugs. J Appl Oral Sci 2010;18:127–34.
9. Camilleri J, Pitt Ford TR. Mineral trioxide aggregate: a review of the constituents and biological properties of the material. Int Endod J 2006;39:747–54.
10. Asgary S, Parirokh M, Eghbal MF, Brink F. Chemical differences between white and gray mineral trioxide aggregate. J Endod 2005;31:101–3.
11. Chng HK, Islam I, Yap AU, Tong YW, Koh ET. Properties of a new root-end filling material. J Endod 2005;31:665–8.
12. Torabinejad M, Hong CU, McDonald F, Pitt Ford TR. Physical and chemical properties of a new root-end filling material. J Endod 1995;21:349–53.
13. Ber BS, Hatton JF, Stewart GP. Chemical modification of ProRoot MTA to improve handling characteristics and decrease setting time. J Endod 2007;33:1231–4.
14. Porter ML, Bertó A, Primus CM, Watanabe I. Physical and chemical properties of new-generation endodontic materials. J Endod 2010;36:524–8.
15. Schembri M, Peplow G, Camilleri J. Analyses of heavy metals in mineral trioxide aggregate and Portland cement. J Endod 2010;36:1210–5.
16. Jacobovitz M, de Lima RKP. The use of calcium hydroxide and mineral trioxide aggregate on apexification of a replanted tooth: a case report. Dent Traumatol 2009;25:e32–6.
17. Krastl G, Allgayer N, Lenherr P, Filippi A, Taneja P, Weiger R. Tooth discoloration induced by endodontic materials: a literature review. Dent Traumatol 2012; doi: 10.1111/j.1600-9657.2012.01141.x
18. Pandolfelli VC, Oliveira IR, Rosseto HL, Jacobovitz M. A composition based on aluminate cement for application in endodontics and the obtained cement product. Patent registration INPI 0704502-6. (Patent registration, Portuguese); 2007.
19. Aguilar FG, Garcia LdaF, Rossetto HL, Pardini LC, Pires-de-Souza FdeC. Radiopacity evaluation of calcium aluminate cement containing different radiopacifying agents. J Endod 2011;37:67–71.
20. International Standards Organization. Dental Root Canal Sealing Materials. BS EN ISO 6876-7.8; 2001.
21. Bakhtiar H, Heidari N, Mehrvarzfar P, Ghazvini K, Habibi M, Jafarzadeh H et al. In vitro comparative study of the microbial leakage of one-step, thermafil and lateral condensation techniques. J Contemp Dent Pract 2012;13:27–30.
22. Pires-de-Souza FdeC, Garcia LdaF, Roselino LdeM, Naves LZ. Color stability of silorane-based composites submitted to accelerated artificial ageing–an in situ study. J Dent 2011;39: e18–24.
23. Pires-de-Souza FdeC, Casemiro LA, Garcia LdaF, Cruvinel DR. Color stability of dental ceramics submitted to artificial accelerated aging after repeated firings. J Prosthet Dent 2009;101:13–8.
24. Jacobovitz M, de Lima RKP. Treatment of inflammatory internal root resorption with mineral trioxide aggregate: a case report. Int Endod J 2008;41:905–12.
25. Schmalz G, Hiller KA, Nunez LJ, Stoll J, Weis K. Permeability characteristics of bovine and human dentin under different pretreatment conditions. J Endod 2001;27:23–30.

26. Húngaro Duarte MA, de Oliveira El Kadre GD, Vivan RR, Guerreiro Tanomaru JM, Tanomaru Filho M, de Moraes IG. Radiopacity of Portland cement associated with different radiopacifying agents. J Endod 2009;35:737–40.
27. Dammaschke T, Gerth HUV, Zuchner H, Schafer E. Chemical and physical surface and bulk material characterization of white ProRoot MTA and two Portland cements. Dent Mater 2005;21:731–8.
28. Camilleri J. Characterization of hydration products of mineral trioxide aggregate. Int Endod J 2008;41:408–17.
29. Kim EC, Lee BC, Chang HS, Lee W, Hong CU, Min KS. Evaluation of the radiopacity and cytotoxicity of Portland cements containing bismuth oxide. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2008;105:e54–7.
30. Ribeiro DA. Do endodontic compounds induce genetic damage? A comprehensive review. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2008;105:251–6.
31. Camilleri J. Evaluation of the physical properties of an endodontic Portland cement incorporating alternative radiopacifiers used as root-end filling material. Int Endod J 2010;43:231–40.
32. Saliba E, Abassi-Ghadi S, Vowles R, Camilleri J, Hooper S, Camilleri J. Evaluation of the strength and radiopacity of Portland cement with varying additions of bismuth oxide. Int Endod J 2009;42:322–8.
33. Ruyter IE, Nilner K, Moller B. Color stability of dental composite resin materials for crown and bridge veneers. Dent Mater 1987;3:246–51.
34. 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;45:942–9.
35. Namazikhah MS, Nekoofar MH, Sheykhrezae MS, Salariyeh S, Hayes SJ, Bryant ST et al. The effect of pH on surface hardness and microstructure of mineral trioxide aggregate. Int Endod J 2008;41:108–16.