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ORIGINAL ARTICLE

Push-out bond strength of three calcium silicate cements to root canal dentine after two different irrigation regimes

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Abstract
Objectives The aim of this study was to compare the push-out bond strength of three calcium silicate cements to dentine after two different irrigation regimes.
Materials and methods One hundred eighty 2-mm-thick root sections were prepared. The sections were divided into three main groups ($n=60$). In group A, the sections were immersed in 1 % NaOCl for 3 min. In group B, the sections were immersed in 17 % EDTA for 3 min followed by 1 % NaOCl for the same period of time. In group C (control group), no irrigation was applied to the sections. The samples from each group were divided into four subgroups ($n=15$). In subgroups A1, B1 and C1, DiaRoot BioAggregate; in subgroups A2, B2 and C2, MTA-Angelus and in subgroups A3, B3 and C3, MM-MTA were mixed with hand-mixing. In subgroups A4, B4 and C4, MM-MTA was mixed with auto-mixing. The mixed cements were then placed into cavities with a carrier. The samples were submitted to the push-out test and were loaded. All the data were analysed with one-way analysis of variance and the Tukey HSD multiple comparisons ($p<0.05$).
Results There were no statistically significant differences between the irrigation regimes regardless of the used materials. The resistance to displacement in subgroup B4 was significantly greater than that presented by subgroups A3, B3 and C3 ($p<0.05$) while not different than all other groups.
Conclusions Irrigation regimes had not affected the push-out bond strength of the calcium silicate cements. Also, strength of auto-mixed MM-MTA was similar to the other cements.
Clinical relevance The novel calcium silicate cements would be a potentially useful material in endodontic procedures with favourable properties.

Keywords Calcium silicate cement · Mineral trioxide aggregate · Push-out bond strength · Irrigation

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Introduction

An ideal repair material should be able to prevent bacterial leakage into the pulp and the periradicular tissues and remain in place under dislodging forces [1, 2]. Therefore, the marginal adaptation and bond strength of these materials are important features because after its placement functional pressures on the teeth or the application of other materials over it might dislodge the material. There are various methods for evaluating the adhesion of a dental material to dentine including tensile, shear and push-out strength tests [3]. One of these, the push-out test has been frequently used in endodontics [4–6]. It has been suggested that the push-out test provides a better evaluation results than the conventional shear test because using the push-out test, fracture occurs parallel to the dentine-bonding interface, which makes it a true shear test for parallel-sided samples [7].

Calcium silicate cements (CSC), e.g. mineral trioxide aggregate (MTA), has been shown to be biocompatible and can be used safely when placed adjacent to pulp and periradicular tissues [8, 9]. It can also provide an environment that supports cementum regrowth [10]. Sealing ability, low solubility and antibacterial characteristics are some of the other properties of CSC [2]. Thus, it appears to be the most promising material for use in a variety of clinical applications, including vital pulp treatments [11], root-end fillings [8], apexification [12],

regenerative endodontic treatments [13] and repair of root perforation and resorption [9], and as a root canal filling material [14]. Despite the numerous favourable properties that support its clinical use, there are some disadvantages when using CSC, such as its extended setting time, difficult handling and potential of discoloration [15]. Thus, several modifications in its original formulation have been suggested to improve its physicochemical and mechanical properties (e.g. flow, setting time and adhesion) [2]. MTA-Angelus (Angelus, Londrina, PR, Brazil) and DiaRoot BioAggregate (DiaDent, Burnaby, BC, Canada) are two of these products. MTA-Angelus is composed of calcium carbonate, calcium silicate, calcium aluminate and barium zinc phosphate than conventional MTA, which contribute to improve setting time and workability [2, 15]. DiaRoot BioAggregate is similar to those in conventional MTA, differing mostly by being aluminium-free for minimizing any toxic effects [16]. It is contained of tricalcium silicate, dicalcium silicate, calcium phosphate monobasic and amorphous silicone dioxide with the addition of tantalum pentoxide, instead of bismuth oxide in conventional MTA, for radiopacity [17].

Recently, a new CSC, MM-MTA (Micro-Mega, Besancon Cedex, France), has been introduced. It has the same indications for use and is composed of similar materials and exhibits comparable technological characteristics to conventional MTA. According to the manufacturer's description, MM-MTA is an insoluble, biocompatible, excellent adhesion to the dentine and radiopaque endodontic repair material. It consisted of special capsules containing MM-MTA powder and liquid; automatic mixing is achieved quickly with a vibrating mixer or an amalgamator. In addition, the resulting blend is extremely homogenous with transformation properties which are always optimal and reproducible. The addition of calcium carbonate to the formula considerably reduces the setting time (20 min) and also allows filling in the same session. It does not contain any pigments or colouring agents so it will not stain teeth.

The aim of this study was to compare the push-out bond strength of three CSCs (DiaRoot BioAggregate, MTA-Angelus and MM-MTA) to root canal dentine after two different irrigation regimes (NaOCl and NaOCl+EDTA).

Materials and methods

Test materials

Composition of the tested materials and their manufacturers were shown in Table 1.

Specimen selection and preparation

One hundred eighty single-rooted, extracted human teeth with similar sizes were used in this study. The crowns of each tooth were sectioned at the cement-enamel junction using a water-cooled diamond disc. The teeth were stored in saline solution until required.

All the middle third of the roots were sectioned perpendicular to the longitudinal axis of the root using a low-speed diamond-coated saw (Micracut 125, Metkon, Bursa, Turkey) under water cooling. Two sections were prepared at a thickness of 2±0.1 mm middle parts of each root. The thickness of each section was measured with a digital calliper. The lumens of the root slices were then enlarged with a spherical diamond bur and two complete passes of a no. 5 Gates-Glidden bur to obtain a 1.3-mm diameter standardized cavities in each section [18]. The root sections were randomly divided into three main groups (n=60). In group A, the sections were immersed in 1 % NaOCl for 3 min. In group B, the sections were immersed in 17 % EDTA for 3 min, followed by 1 % NaOCl for the same period of time [6]. In group C (control group), no irrigation was applied to the sections. All specimens were then immediately washed in distilled water and dried. After these irrigation procedures, each group was randomly divided into four subgroups (n=15). In subgroups A1, B1 and C1, DiaRoot BioAggregate (DiaDent, Burnaby, BC, Canada) was mixed following the manufacturer's instructions; in subgroups A2, B2 and C2, MTA-Angelus (Angelus, Londrina, PR, Brazil) was mixed following the manufacturer's instructions; in subgroups A3, B3 and C3, MM-MTA (Micro-Mega, Besancon Cedex, France) capsule was opened, and its liquid was added to the powder. The liquid was absorbed by the MTA powder before gentle mixing (hand-mixing), and in subgroups A4, B4 and C4, MM-MTA was mixed following the manufacturer's instructions (auto-mixing); MM-MTA capsule was placed in a vibrating mixer and mixed for 30 s at 4.300 oscillations/min frequency.

The mixed materials were then placed into cavities with an MM-MTA gun (in A4 and B4 groups) or carrier (in other groups) and compacted with a plugger. Excess material was trimmed from the surface of the specimens with a scalpel. All specimens were examined under a microscope in order to discard any cracks, defects or gaps between the material and dentine walls. Immediately after filling, a wet cotton pellet was placed over the cement for 4 h, and the samples were incubated for 7 days at 37 °C and 100 % relative humidity.

Push-out test

After the experimental period, the samples were submitted to the push-out test. The push-out bond strength values were measured by using a universal testing device (Lloyd Instruments Ltd., Fareham, UK). After the samples were placed, the compressive load was applied by exerting a downward pressure on the surface of endodontic material by using a 1-mm diameter cylindrical stainless steel plunger at a speed of 1 mm/min. The push-out bond strength at failure was calculated in megapascals

Table 1 Composition of the materials and their manufacturers

Materials	Composition	Lot number	Manufacturers
DiaRoot BioAggregate	Tricalcium silicate, dicalcium silicate, tantalum pentoxide, calcium phosphate monobasic and amorphous silicon oxide	1201BA	DiaDent, Burnaby, BC, Canada
MTA-Angelus	Tricalcium silicate, dicalcium silicate, tricalcium aluminate, tetracalcium aluminoferrite, bismuth oxide, iron oxide, calcium carbonate, magnesium oxide and crystalline silica and residues (calcium oxide, free magnesium oxide, potassium and sodium sulphate compounds)	17897	Angelus, Londrina, Brazil
MM-MTA	Tricalcium silicate, dicalcium silicate, tricalcium aluminate, bismuth oxide, calcium sulphate dehydrate and magnesium oxide	7105278	Micro-Mega, Besancon Cedex, France

(MPa) by dividing the load in newtons (N) by the area of the bonded interface (mm^2). The bonded area of each section was calculated using the following formula: $2\pi r \times h$, where π is the constant 3.14, r is the radius of the intra-radicular space, and h is the height of section in millimetre.

Scanning electron microscopy analysis

Two samples in each group were randomly chosen for scanning electronic microscopy (SEM) examination, and they were mounted on an aluminium stub and sputter-coated with gold layer. SEM photomicrographs were taken using an SEM (LEO 1430; Zeiss, London, UK) at magnifications of ×100 and ×500 for determining the nature of the bond failure. Each sample was categorized into one of three failure modes: adhesive failure at the MTA and dentin interface, cohesive failure within MTA or mixed failure.

Statistical analysis

All statistical analyses were performed with SPSS, 13.0 (SPSS for Windows; SPSS Inc, Chicago, IL, USA). One-way analysis of variance and the Tukey HSD multiple comparisons test were used for the analysis of data; the independent variables were endodontic material and third root canal ($P=0.05$).

Results

The mean values of the push-out strength (MPa) for each group and subgroup are shown in Table 2. There were no statistically significant differences between the irrigation regimes regardless of the used CSCs ($P>0.05$). The resistance to displacement in subgroup B4 (NaOCl+EDTA, auto-mixed MM-MTA) was significantly greater than that presented by subgroups A3 (NaOCl, hand-mixed MM-MTA), B3 (NaOCl+EDTA, hand-mixed MM-MTA) ($P<0.05$) and C3 (control, hand-mixed MM-MTA) while not different than all other groups.

Inspection of the samples revealed that the bond failure was of the adhesive type in the majority of the samples; however, some samples exhibited cohesive and mixed failure patterns (Fig. 1).

Discussion

To date, numerous studies have been published about CSC's physical and chemical properties, biocompatibility and clinical applications [2, 15, 19]. CSC is a very important

Table 2 Mean and standard deviation (SD) of the push-out strength values (in megapascal) for the displacement of the endodontic material from the specimen in the middle thirds of each group

Groups	Subgroups	Mean	Median	SD
Group A (NaOCl)				
A1	DiaRoot BioAggregate	5.5520^{a}	5.3100	0.86589
A2	MTA-Angelus	5.5373^{a}	5.9000	0.89674
A3	Hand-mixed MM-MTA	$5.3180^{b,a}$	5.9600	0.91334
A4	Auto-mixed MM-MTA	5.7860^{a}	5.9500	0.79813
Group B (NaOCl+EDTA)				
B1	DiaRoot BioAggregate	5.9607^{a}	5.8500	0.80964
B2	MTA-Angelus	6.0340^{a}	6.0000	1.08634
B3	Hand-mixed MM-MTA	$5.4513^{b,a}$	5.4100	0.84667
B4	Auto-mixed MM-MTA	$6.4840^{c,a}$	6.4900	0.84129
Group C (Control, no irrigation)				
C1	DiaRoot BioAggregate	5.6510^{a}	5.0100	0.06964
C2	MTA-Angelus	5.6571^{a}	5.0000	0.06634
C3	Hand-mixed MM-MTA	$5.3055^{b,a}$	5.0500	0.06667
C4	Auto-mixed MM-MTA	5.9040^{a}	5.5000	0.04129

The same superscript letters indicate statistically no significant values. Mean with same letter is not significantly different at the $P=0.05$ level.

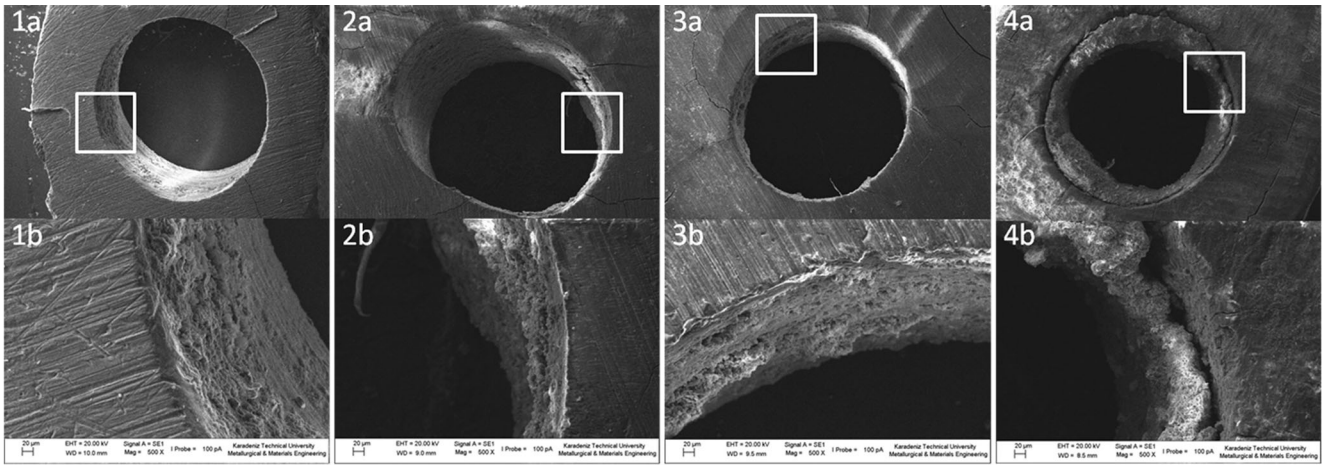


Fig. 1 SEM images of the samples. Bond failure was of the adhesive type in the majority of samples; however, some samples exhibited cohesive and mixed failure patterns. *1* Subgroup C2, *2* subgroup B4, *3* subgroup B3, *4* subgroup A1, *a* ×100, *b* ×500

endodontic material for endodontists because it is used in critical areas of the tooth such as external root surface for perforations and apical resections. Therefore, it should be biocompatible, dimensionally stable, adhere to dentine, resist dislocating forces, prevent the bacterial passage and be unaffected by the presence of different tissue fluid conditions [20]. To provide these characteristics, several modifications in its original formulation have been introduced. DiaRoot BioAggregate, MTA-Angelus and MM-MTA are modified formulations of them. This study was designed to assess the push-out bond strength of these endodontic materials to root canal dentine after two different irrigation regimes.

To research the bond strength between dentine and CSC can be revealed by the value of adhesion between them [3]. For this reason, the push-out bond strength of CSCs has been evaluated in previous studies [2, 15]. Some studies have evaluated the influence of different conditions (wet/dry and acidic/alkaline pH) on push-out bond strength of CSC [1, 3, 5, 21, 22]. According to the researchers, the push-out strength of CSC increased in wet conditions [1, 21].

Moisture environment can affect the setting time of CSC. In clinical practice, a wet cotton pellet placed over the material for a while. Gancedo-Caravia & Garcia-Barbero [1] showed significantly higher push-out strength with the application of moisture after placement of CSC mixture. In this study, a wet cotton pellet was placed over the material for 4 h, and the samples were incubated for 7 days at 37 °C and 100 % relative humidity to allow for complete setting of materials. When choosing the time to place cotton pellet, manufacturers' instructions were taken into account. Minimum setting time is 10–15 min for MTA-Angelus, 20 min for MM-MTA and 4 h for DiaRoot BioAggregate.

Mixing and compaction techniques can be affected by the chemical, physical and biological properties of CSC. On mixing, it becomes a slurry paste that is difficult to handle and compact into the root canal spaces without creating voids [23]. Several techniques have been suggested for sending and condensing the CSC into the root canal spaces such as the use of a lentulo, hand files and pluggers and ultrasonic activation [14, 23–25]. Hand condensation with indirect ultrasonic activation has been suggested to improve the compaction of CSC and its adaptation to dentine by generating compressive forces that lead to reorganizing of particles and facilitate the escape of entrapped air [25]. On the other hand, El-Ma'aita et al. [23] demonstrated on extracted teeth that manual compaction of CSC produced significantly denser root fillings than ultrasonic activation. In addition, Aminoshariae et al. [24] compared the ultrasonic and hand condensation techniques on adaptation of CSC in simulated root canals. They concluded that ultrasonic activation resulted in more voids and poorer adaptation. According to the Namazikhah et al. [26] and Nekoofar et al. [27], voids might not be a problem for the optimal setting because they might provide pathways for diffusion of water into the CSC.

To date, hand and mechanical (trituration or ultrasonic vibration) techniques are used for mixing the CSC [6, 28]. Nekoofar et al. [28] compared the effects of various mixing techniques including ultrasonic vibration, trituration of customized encapsulated CSC and hand condensation on the microhardness of CSC. They demonstrated that an ultrasonic vibration produces a significantly higher surface microhardness value than the other techniques. Whereas, Shahi et al. [6] showed that the conventional mixing technique results in a higher push-out strength compared with ultrasonic and trituration techniques, with no statistically significant differences between the three techniques. In the present study, two mixing techniques (hand-mixing and auto-mixing) were applied. Auto-mixing was used only at MM-MTA according to the manufacturer's suggestions. But, in A3 (NaOCl, hand-mixed MM-MTA) and B3 (NaOCl+EDTA, hand-mixed MM-MTA) subgroups, as with other hand-mixed groups, MM-MTA was used with hand-mixing technique in a different way from the

manufacturer's suggestion. The resistance to displacement in subgroup B4 (NaOCl+EDTA, auto-mixed MM-MTA) was significantly greater than that presented by subgroups A3, B3 and C3 while not different than all other groups. A possible explanation to this result is that auto-mixing of the MM-MTA may change material's physical properties such as viscosity, setting time and flowability. The mixing of the particles in optimum conditions in liquid with titration may provide a more homogeneous mixture according to the hand-mixing. This may also increase the push-out values of the mixture by increasing the amount of water absorbing. In addition, the difference between MM-MTA and the other CSC mixtures is the presence of calcium sulphate dehydrate in the content. This may increase the push-out value by absorbing more water. This feature of it may be with its enough water attraction in the homogeneous mixture.

The advantage or disadvantage of leaving the smear layer intact is still a controversial issue among endodontists. Some authors suggested that keeping the smear layer may block the dentinal tubules and hinder bacterial or toxin penetration [29]. In contrast, other authors believe that the smear layer must be completely removed because it can increase the risk of infection and microleakage by preventing root canal irrigants and the fillings from penetrating into the dentinal tubules [30, 31]. To date, NaOCl and EDTA are routinely used to dissolve the inorganic and organic substances in the smear layer. Several studies [4, 32–34] have investigated effects of root canal irrigants and smear layer on the chemical and physical properties of CSC. In a study, researchers found that the use of CSC under the NaOCl irrigation was effective in perforation repair without altering its hydration behaviour even in the early setting phase [4]. Whereas Lee et al. [33] showed the adverse effects of EDTA on CSC hydration, microhardness and cell adhesion. Additionally, Yan et al. [32] showed that EDTA/carbamide peroxide decreased the bond strength between dentine and CSC. Uyanik et al. [35] examined the effect of different irrigation regimens on the sealing ability of CSC and showed that using MTAD and EDTA significantly increases leakage of CSC. In another study [36], CSC showed significantly higher leakage when smear layer was removed before placing the material. This might be because of the reaction between CSC and residual EDTA inside the root canal [36]. In the present study, there was no difference between the irrigation regimes regardless of the used materials. This showed that the removal of the smear layer is not important for push-out bond strength of CSC. The push-out values were increased in all groups including EDTA. Auto-mixed MM-MTA values were found highest according to the other groups. This difference can be related with the particle size of MM-MTA that is penetrating into the dentinal tubules that are smaller than the other CSC and homogeneous mixture gaining while auto-mix. Homogeneous mixture may be changing the particle size.

Conclusions

Within the limitations of the current study, it can be concluded that none of the irrigation regimes evaluated had any significant effect on push-out booth strength of three CSCs. Also, push-out bond strength of auto-mixed MM-MTA was similar to the other tested cements.

Conflict of interest The authors deny any conflicts of interest

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