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Do intracanal medications used in regenerative endodontics affect the bond strength of powder-to-liquid and ready-to-use cervical sealing materials?

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Abstract

Aim:

This study evaluated the effect of four intracanal medications commonly used in regenerative endodontic procedures on the bond strength of four calcium silicate-based materials, in which two are powder-to-liquid products (MTA and MTA-HP) and the other are ready-to-use materials (EndoSequence Root Repair Material Fast Set Putty [ERRM] and Bioceramic Repair [BIO-C Repair]).

Methods:

Ten bovine central incisors were selected and 4 slices (1.0 ± 0.1 mm) were prepared from each root. Next, four 0.8-mm wide holes were drilled in each slice and specimens were filled with one of the following intracanal medications: triple antibiotic paste, double antibiotic paste, calcium hydroxide with distilled water, and calcium hydroxide with 2% chlorhexidine gel. After 21 days, holes were filled with one of the materials: MTA, MTA-HP, ERRM, or BIO-C Repair. After storage, push-out test and failure analysis were performed. Data were submitted to analysis of variance in a 4 × 4 factorial scheme. Tukey's test was used for multiple comparisons.

Results:

The use of different interappointment dressings did not influence the results ($P > 0.05$). ERRM and BIO-C Repair presented significantly higher values than MTA and MTA-HP ($P < 0.0001$). Specimens showed a 100% occurrence of adhesive failures.

Conclusion:

The use of different intracanal medications presented similar impact on bond strength of calcium silicate-based materials. Ready-to-use ERRM and BIO-C Repair materials presented the best push-out values to dentine, whereas powder-to-liquid MTA and MTA-HP cements showed the lowest results.

Keywords: Bioceramic Repair, compressive strength, nonvital tooth, regenerative endodontics, root canal therapy

INTRODUCTION

Regenerative endodontic procedures (REPs) are currently considered a viable and favorable alternative for the treatment of necrotic immature permanent teeth.[1] Its steps involve passive decontamination with irrigating solutions, use of an interappointment dressing, induction of blood clot, cervical sealing/barrier, and finally crown restoration.[1,2] The root disinfection is thus obtained by irrigating solutions and intracanal medications such as triple antibiotic paste (TAP) or calcium hydroxide-based medicaments.[2,3] Regardless of the medicament selected, it needs to present effective antimicrobial activity and low cytotoxicity.[3] Furthermore, interappointment dressings should be efficiently removed from root canal walls since its remaining can interfere with the dentine bonding of cervical sealing materials.[4,5]

The long-term success of REPs results not only from effective root disinfection but also from its maintenance.[2,3] Therefore, cervical and coronal sealing are important steps that prevent root canal reinfection. In REPs, cervical sealing stage should be accomplished with a hydraulic silicate cement.[6] Most active members of the American Association of Endodontists – about 78.5%– use mineral trioxide aggregate (MTA) for this purpose.[2] However, MTA presents adverse handling aspects and induction of crown discoloration due to the presence of bismuth oxide on its composition.[7] Following MTA, the second most used material in REPs among practicing endodontists in the United States is the EndoSequence Root Repair Material (14.1%) (ERRM; Brasseler, Savannah, GA, USA).[2] This material is ready-for-use and mainly composed of calcium silicates derivate (40%–50%) and zirconium oxide (15%–18%).[8]

New MTA formulations have been proposed to overcome MTA drawbacks. MTA Repair HP (MTA-HP; Angelus, Londrina, PR, Brazil) is based on the conventional MTA formulation, but it contains calcium tungstate as radiopacifier to prevent tooth staining and a mixing liquid as a plasticizer, which is claimed to improve handling features.[9] Moreover, Bioceramic Repair (BIO-C Repair; Angelus, Londrina, PR, Brazil) is a recently developed premixed cement that presents the non-staining radiopacifier zirconium oxide. BIO-C Repair previously demonstrated interesting properties,[10] but further results are clearly needed. BIO-C Repair resistance to dislodgment, which is a relevant physical aspect, has not been evaluated yet.

At present, there is scarce and conflicting information in literature of the effects of interappointment dressings used in REPs in the bond strength of cervical sealing materials. [4,11,12,13,14,15,16] Therefore, this study aimed to evaluate the effect of four intracanal medications commonly used in REPs – TAP, double antibiotic paste (DAP), calcium hydroxide associated with distilled water (CH), and calcium hydroxide associated with 2% chlorhexidine gel (CHP) – on push-out results of powder-to-liquid and ready-to-use hydraulic silicate cements (MTA, MTA-HP, ERRM, and BIO-C Repair). In this sense, the research question was “Which intracanal medications and materials allow adequate protocols in REPs regarding bond strength results of the cervical sealing?” The null hypotheses tested were: (1) there is no difference in bond strength results with the previous use of different intracanal medications and (2) there is no difference in the bond strength results of different materials used as cervical sealing of REPs.

METHODS

Sample size calculation

Based on a previous study,[17] an effect size of one was added to a power β = 80% and α = 5% inputs for analysis of variance (ANOVA) analysis (G*power 3.1 for Macintosh). Total size of 10 specimens per group was recommended for this study (StatPlus: Mac Software® - Version v7; AnalystSoft Inc., Alexandria, VA, USA).

Selection and preparation of specimens

The local ethics committee approved this research (1.154.233). This methodology was based on the previous studies with modifications.[17,18] Ten bovine central incisors with one circular canal, straight roots, and complete root formation were selected. Teeth presenting dentinal cracks, root resorptions, or root canal calcifications were excluded from the study. Teeth were cleaned by periodontal curettes and washed in 5.25% sodium hypochlorite solution before use.

Figure 1 details all materials used and steps of preparation and analysis of specimens. From the cementoenamel junction, teeth were sectioned to four horizontal slices with 1.0-mm thickness each (±0.1 mm) using a low-speed saw (ISOMET; Buhler Ltd. Lake Buff, NY, USA) with a diamond disc of Ø 125 mm × 0.35 mm × 12.7 mm (Buhler Ltd.), under continuous irrigation. A total of 40 slices, four per tooth, were produced following this protocol [Figure 2a]. Next, four holes were drilled on each slice using a 0.8-mm cylindrical carbide bur [Figure 2b]. These holes were prepared to simulate anatomic standardized root canals. The canal-like holes were drilled parallel to the root canal, under continuous water irrigation and by the aid of the vertical support drill stand Dremel Workstation 220®(Dremel, Mount Prospect, IL, USA). A minimum distance (≥1.0 mm) was established between the holes, the external cementum, and the main root canal.[17,18]

Disinfection protocol and filling of canal-like holes

All slices were immersed in a sequence of irrigating solutions described in Figure 1. Then, the four consecutive slices from each tooth were filled with one of the following intracanal medications[19,20,21,22]: TAP, DAP, CH, and CHP [Figure 2c].

Specimens were stored for 21 days at 37°C in contact with gauzes moistened in phosphate-buffered solution (PBS) that was renewed every 3 days. Then, intracanal medications were removed and the humidity control of specimens was performed with paper points. Subsequently, each hole in the same slice was filled with one of the tested calcium silicate-based materials: MTA, MTA-HP, ERRM, and BIO-C Repair [Table 1]. A glass plate and three drops glue (DVA Zapit Base, Dental Ventures of America, Corona, CA, USA) were used to fix and stabilize the specimens during the insertion of materials. A drop of glue was also carefully applied on the coronal surface of each slice for identification of the materials [Figure 2d]. Holes were filled until exhibiting a slight overflow of materials. After 24 h, materials' excesses were removed with 600 sandpaper and specimens were newly stored as previously mentioned for 7 days.

Push-out bond strength test

The push-out of the tested hydraulic silicate cements was assessed in a universal testing machine (Instron, Canton, MA, USA). Loading was performed with a plunger tip of 0.6 mm diameter that was positioned over only one of the tested materials for each analysis. The load was applied in a coronal-to-apical direction at a speed of 0.5 mm/min, until failure occurred [Figure 2e]. During load application, loading time (N) × displacement (mm) was recorded by a real-time software program. The bond strength was calculated in mega Pascal (MPa), dividing the load by the area of the bonded interface. The area of materials was obtained using the following formula[18]: $A = 2\pi r \times h$, whereas π = constant value of 3.14; *r* = canal-like holes' radius (0.4 mm); and h = height of the material (1.0 mm).

Failure bond analysis

After 48 h of the push-out test, failure bond analysis was performed on the hydrated samples under a stereomicroscope (Optz; Opticam Microscopy Technology, Doral, Florida, USA) at ×40. Failures were categorized in: (1) adhesive; (2) cohesive, either in dentine or material; or (3) mixed fracture. Next, a scanning electron microscope (SEM) (JSM 5600 Lv; JEOL, Tokyo, Japan), at 15 kV and 10 mm of work distance, was used after sputtering the samples with AuPd (Denton Vacuum Desk II, Denton Vacuum, Moorestown, NJ, USA) for representative images.

Statistical analysis

Data were submitted to exploratory and descriptive analysis. Afterward, the push-out bond strength values were analyzed, using ANOVA in a 4 × 4 factorial scheme (SAS, 9.4). Tukey's test was used for multiple comparisons. The significance level was set at 5% ($\alpha = 0.05$).

RESULTS

Table 2 shows the results obtained in MPa. The intracanal medications did not influence the push-out values of calcium silicate-based materials ($P > 0.05$). ERRM and BIO-C Repair presented similar results and significantly higher values of push-out compared to MTA and MTA-HP, regardless of the intracanal medicament used ($P < 0.0001$). MTA and MTA-HP demonstrated the lowest bond strength values, with no significant difference among each other ($P > 0.05$). In SEM analysis, specimens showed a 100% occurrence of adhesive failures between the material/dentine interface [Figure 2f-g].

DISCUSSION

The use of several intracanal medications, presenting different components, had similar impact on bond strength performance of four cervical sealing materials. Therefore, the first null hypothesis was accepted. This suggests that the bond strength outcomes of hydraulic cements are not dependent of previous insertion of interappointment dressings. Accordingly, the selection of intracanal medications should be based mainly based on biocompatibility and antimicrobial activity. Furthermore, the undesirable esthetic outcome caused by minocycline should be avoided. The use of calcium hydroxide-based pastes, DAP without minocycline paste, or the exchange of minocycline for other antibiotic (e.g., clindamycin, amoxicillin, and cefaclor) are possible alternatives.[2]

It should be pointed that this finding contrasts with the previous studies that used conventional push-out methods and observed a diverse interdepend association of push-out between sealing materials and interappointment dressings used.[13,14,16] Several methodology aspects can explain the conflicting results in literate: sample selection, irrigation protocol, anatomy variability, concentration of intracanal medications, environmental conditions, storage period, technique of dressings removal, and handling method of materials. However, it has been shown that the current intratooth method presents an increased internal validity and a bias reduction of push-out conditions.[17,18] This is explained by the standardized production of canal-like holes to allow equal internal anatomy for all groups. Therefore, the same dentin source was used to all materials and a reliable baseline was available for comparisons due to the elimination of biological bias associated with dentin heterogeneity.[18] The push-out test used in this study reduces the area loaded and produces a nondivergent angle in the shear surface of root canals. This is achieved using in parallel a cylindrical bur that homogenizes the dentine surface and thus the configuration of the area loaded.[23] As a result, the variability of the area analyzed is reduced and the outcomes observed in failure analysis demonstrated a homogeneous shear tensile concentration around the materials that leaded to a more homogeneous fracture pattern (100% adhesive). This homogeneity has not appeared in other bond strength tests as previously observed.[24] In addition to that, the reduction of contact area decreases the chance of uneven loads and bias of the load direction.

The limitation of this current push-out test continues to be the variability of the shear strength experiment conditions that result from the heterogeneity of the materials tested and the dentinal tubules dissimilarity trough different regions. Therefore, it should be emphasized that the complexity of compositions and differences among the materials evaluated makes them considerably heterogeneous during push-out tests. However, this method can be efficient when there is no interference of the tensile strength test of resin plugs/tags formation, as it applies for this study, where the retention relied on the setting expansion and mineralization of materials.

Another methodology aspect that should be highlighted is the selection of animal specimens. Bovine teeth demonstrated to reliably substitute human dentine in push-out analysis of endodontic materials.[18] Therefore, bovine specimens were selected for this study to obtain specimens in similar conditions, such as tooth age and diet. The used intratooth model also represents an effort to provide the same baseline substrate for all groups since all materials were applied in the same anatomy and dentine conditions.[17,18]

Calcium silicate-based materials are ruled by a pozzolanic reaction in which the amount of free Ca release could be observed as a difference factor among different products within this group.[25] MTA-based materials promote a mineral nucleation after the hydration phase of its silicates, with a considerable expansion (×3.5) during this process.[19] This process yields the mechanical retention that is their mechanism of bonding.[20] In this study, hydraulic cements ERRM and BIO-C Repair displayed the best resistance to a shear stress load, whereas MTA and MTA-HP demonstrated the lowest values. Therefore, the second null hypothesis was rejected. The main difference among the materials studied here is the presence of zirconium oxide only in the composition of ERRM and BIO-C Repair groups, in similar rates (~15%).[21] This could be related with a better biomineralization property of these groups, since they have the silicate hydration with the addition of zirconium oxide that has a nucleation activity inside the complex, leading to the formation of $CaZrPO_4$.[22] Therefore, it is more appealing to create stronger 3D networks. The increase of Ca free in the reaction also could triggers the reaction accelerating and enhancing the hydration state.[25] A significantly higher push-out value of ERRM in comparison to MTA after the application of several interappointment dressings, in different concentration, has also been recently observed.[16] Furthermore, the materials evaluated differ regarding the preparation mode. ERRM and BIO-C Repair are presented as premixed syringes ready for use, what represents an easy and predictable handling. Conversely, MTA and MTA-HP powder needs to be manually homogenized with their respective vehicle. It seems that this feature interfered in bond strength outcome of repair materials.

CONCLUSION

The use of different intracanal medications presented similar impact on bond strength results calcium silicate-based materials. Ready-to-use ERRM and BIO-C Repair materials presented the best bond strength values to root dentin, whereas powder-to-liquid MTA and MTA-HP cements showed the lowest results.

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Conflicts of interest

There are no conflicts of interest.

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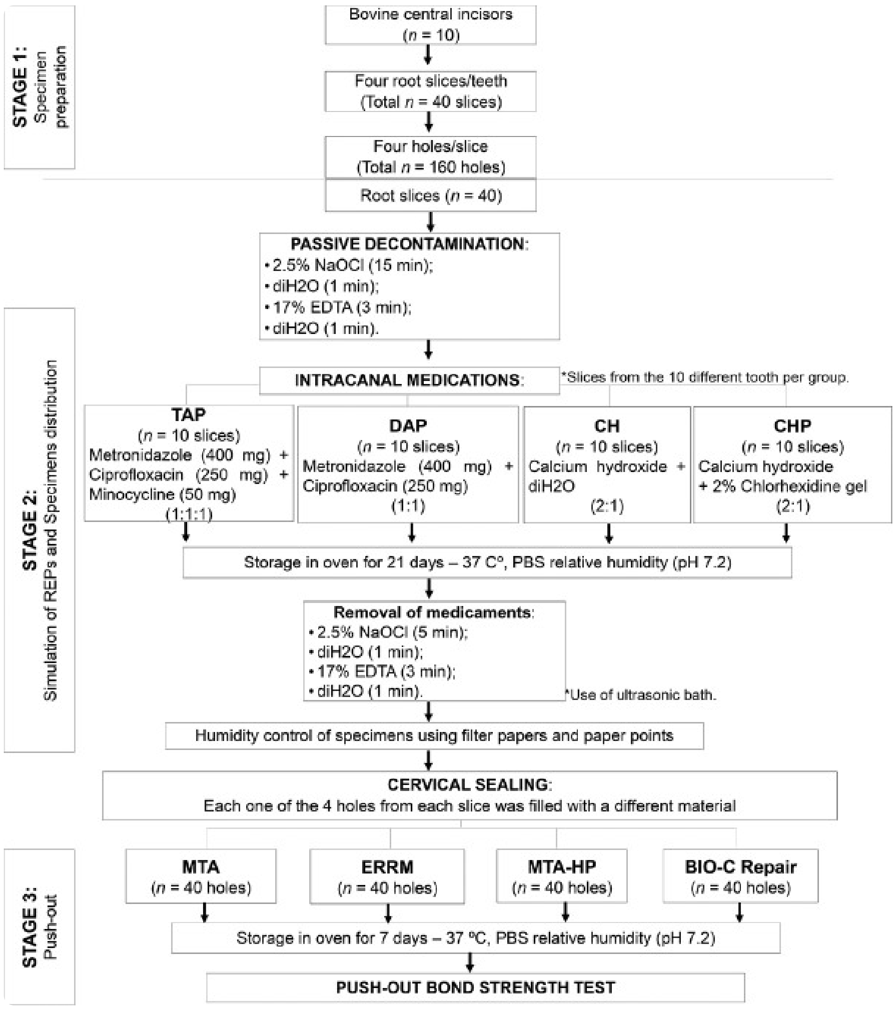
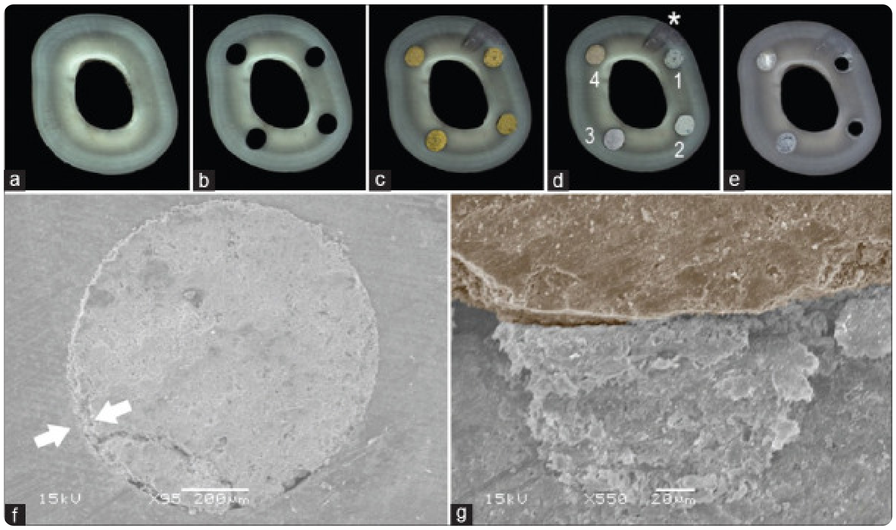
Figures and Tables

Figure 1



Flowchart of study design. Canal-like holes were prepared (Stage 1). Regenerative endodontic procedures were performed and specimens were distributed in groups according to intracanal medications and calcium silicate-based materials used (Stage 2). Push-out test was accomplished (Stage 3). NaOCl: Sodium hypochlorite solution, diH2O: Distilled water, TAP: Triple antibiotic paste, DAP: Double antibiotic paste, CH: Calcium hydroxide, CHX: Chlorhexidine

Figure 2



Stereoscopic micrograph of a specimen during experiment (a-e; ×5) and representative image of fracture in SEM (f-g). (a) Initial tooth slice. (b) Canal-like holes prepared. (c) Filling of holes in the same slice with only one intracanal medication (ex: TAP). (d) Materials inserted in a clockwise direction from the mark (*): MTA (1), MTA-HP (2), ERRM (3), and BIO-C Repair (4). (e) Adhesive bond failure. (f) Canal-like hole observed in low magnification showing complete debond area around the interface. (g) In a higher magnification (inputs), the debond area is highlighted within the margins of the material (colored)

Table 1

Description of intracanal medications and cervical sealing materials used in this study

Material	Composition	Preparation
TAP[19]	Metronidazole (400 mg), ciprofloxacin (250 mg), and minocycline (50 mg) (Drogal, Piracicaba, SP, Brazil) Liquid: Distilled water	The three components were homogenized in equal proportions (1:1:1) with distilled water
DAP[20]	Metronidazole (400 mg) and ciprofloxacin (250 mg) (Drogal, Piracicaba, SP, Brazil) Liquid: Distilled water	The two components were homogenized in equal proportions (1:1) with distilled water
CH[21]	Calcium hydroxide (biodinâmica, Ibiporã, PR, Brazil) Liquid: Distilled water	The powder was homogenized with distilled water in equal proportions (1:1)
CHP[22]	Calcium hydroxide (Biodinâmica, Ibiporã, PR, Brazil) Liquid: 2% chlorhexidine gel (Endogel; Essencial Farma Ltda., Itapetininga, SP, Brazil)	The powder was homogenized with 2% chlorhexidine gel in equal proportions (1:1)
White MTA-2001 (MTA; Angelus, Londrina, PR, Brazil)	Powder: Tricalcium silicate (40%-60%), bismuth oxide (15%-30%), dicalcium silicate (5%-12%), tricalcium aluminate (5%-12%), tetracalcium aluminoferrite (1%-7%), calcium oxide (0%-7%)* Liquid: Distilled water	One powder measure (1 sachet or 1 spoon) was homogenized with 1 drop of distilled water for 30 s
MTA repair HP-2016 (MTA-HP; Angelus, Londrina, PR, Brazil)	Powder: Tricalcium silicate (45%-55%), calcium tungstate (20%-30%), bismuth oxide (15%-30%), dicalcium silicate (10%-15%), tricalcium aluminate (5%-12%), calcium oxide (1%-5%)* Liquid: Water and plasticizer	One powder package was homogenized with two drops of the liquid for 40 s
ERRM-2008 (ERRM; Brasseler, Savannah, GA, USA)	Tricalcium silicate (30%-36%), zirconium oxide (15%-18%), tantalum pentoxide (12%-15%), dicalcium silicate (9%-13%), calcium sulfate (3%-8%)*	Premixed syringe ready-to-use
BIO-C repair-2019 (BIO-C repair; Angelus, Londrina, PR, Brazil)	Calcium silicates, calcium aluminate, calcium oxide, zirconium oxide, iron oxide, silicon dioxide, and dispersing	Premixed syringe ready-to-use

*Information obtained in the SDS sections of materials. SDS: Safety data sheet, TAP: Triple antibiotic paste, DAP: Double antibiotic paste, CH: Calcium hydroxide, CHP: Calcium hydroxide associated with chlorhexidine, MTA: Mineral trioxide aggregate, HP: High plasticity, ERRM: EndoSequence Root Repair Material, BIO-C Repair: Bioceramic Repair

Table 2

Push-out bond strength values (mean [standard deviation]), expressed in MPa, of calcium silicate-based materials according to intracanal medications previously applied

Intracanal medications	Total	Cervical sealing materials			
		MTA	MTA-HP	ERRM	BIO-C repair
Triple antibiotic paste	3.40 (3.19)[a]	1.56 (1.50)[a,B]	1.66 (1.47)[a,B]	5.72 (3.74)[a,A]	4.31 (3.25)[a,A]
Double antibiotic paste	2.94 (3.00)[a]	1.33 (1.22)[a,B]	1.88 (2.85)[a,B]	4.29 (3.85)[a,A]	3.88 (2.62)[a,A]
Calcium hydroxide + diH_2O	3.64 (3.19)[a]	1.77 (1.24)[a,B]	0.50 (0.32)[a,B]	7.70 (2.43)[a,A]	4.27 (1.73)[a,A]
Calcium hydroxide + CHX	2.75 (3.30)[a]	1.17 (0.81)[a,B]	0.40 (0.48)[a,B]	4.95 (4.70)[a,A]	4.27 (2.59)[a,A]
Total	-	1.45 (1.17)[B]	1.10 (1.65)[B]	5.66 (3.84)[A]	4.18 (2.51)[A]

Values (MPa) followed by different letters (lower letters vertically [comparison between intracanal medications] and uppercase letters horizontally [comparison between cervical sealing materials]) are significantly different ($P \leq 0.05$). diH_2O: Distilled water. CHX: Chlorhexidine, MTA: Mineral trioxide aggregate, HP: High plasticity, ERRM: EndoSequence root repair material, BIO-C Repair: Bioceramic Repair