



The effect of different solutions on bond strength of two root canal sealers

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Abstract

Objective: The aim of this study was to evaluate the effect of different solutions on bond strength of two root canal sealers.

Methods: Sixty bovine incisor roots were randomly divided into five groups: group 1, physiologic solution (control); group 2, 2% chlorhexidine digluconate (CHX) followed by EDTA; group 3, CHX followed by EDTA and ethanol (EtOH); group 4, 5.25% sodium hypochlorite (NaOCl) followed by EDTA; and group 5, NaOCl followed by EDTA and EtOH. Each group was subdivided into two subgroups: AH Plus and MTA Fillapex sealers. Bond strength was measured by push-out test and were analyzed statistically by two-way ANOVA and Tukey's ($\alpha=0.05$).

Results: The different irrigating protocols did not influence the bond strength of either sealer ($p>0.05$). AH Plus sealer showed higher values of bond strength to root dentin than MTA Fillapex ($p<0.05$).

Conclusion: The bond strength of AH Plus and MTA Fillapex were not influenced by different solutions.

Key words: Bond strength; Chlorhexidine; EDTA; Root canal sealer; Sodium hypochlorite

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Efeito de diferentes soluções na resistência de união de dois cimentos obturadores endodônticos

Resumo

Objetivo: O objetivo do estudo foi avaliar o efeito de diferentes soluções na resistência de união de dois cimentos obturadores endodônticos.

Métodos: Sessenta incisivos bovinos foram aleatoriamente divididos em cinco grupos: grupo 1, solução fisiológica (controle); grupo 2, digluconato de cloredidine 2% (CHX) seguido de EDTA 17%; grupo 3, CHX seguido de EDTA 17% e etanol (EtOH); grupo 4, hipoclorito de sódio 5,25% (NaOCl) seguido de EDTA 17%; grupo 5, NaOCl seguido de EDTA 17% e EtOH. Cada grupo foi subdividido em dois subgrupos: obturados com AH Plus e MTA Fillapex. A resistência de união foi mensurada pelo teste *push-out* e analisada estatisticamente por ANOVA com dois fatores de variação e Tukey's ($\alpha=0,05$).

Resultados: Os diferentes protocolos de irrigação não influenciaram a resistência de união dos cimentos ($p>0,05$). AH Plus mostrou maiores valores de resistência de união à dentina do que MTA Fillapex ($p<0,05$).

Conclusão: A resistência de união do AH Plus e MTA Fillapex não foi influenciado pelas diferentes soluções.

Palavras-chave: Resistência de união; Clorexidine; EDTA; Cimento obturador endodôntico; Hipoclorito de sódio

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Introduction

The principal objectives of endodontic therapy are to eliminate intracanal bacteria and to seal the root canal system and cavity access with materials that prevent microleakage [1]. Gutta-percha or Resilon cones and root canal sealers have become the most commonly used and accepted materials for the filling of endodontically treated teeth [2,3]. The sealer should present adequate flow for filling the spaces between the cones and the canal walls. Furthermore, the bond strength of root canal sealers to dentin is important for maintaining the integrity of the seal in the root canal [4] because gutta-percha does not directly bond to the dentin surface [5].

Epoxy resin-based sealer cements such as AH Plus sealer (Dentsply, Detrey, GmbH, Germany) have been widely used because of their acceptable physical properties, reduced solubility, apical sealing, adequate bond strength to root dentin, and adequate biological performance [6-8]. Some studies have shown that this sealer has higher bond strength to root dentin than other sealers [6-8]. MTA Fillapex (Angelus Indústria de Produtos Odontológicos S/A, Londrina, Brazil) is another recently introduced MTA-based sealer. According to the manufacturer, its composition after mixing is basically mineral trioxide aggregate (MTA), salicylate resin, natural resin, bismuth, and silica with claims of excellent radiopacity, easy handling, and good working time [9]. MTA Fillapex showed antibacterial activity against *Enterococcus faecalis* before setting [10]. Furthermore, Salles et al. [11] showed that after setting, the cytotoxicity of MTA Fillapex decreases and the sealer presents suitable bioactivity to stimulate hydroxyapatite crystal nucleation. However, to date, scant knowledge is available with regard to its adhesive properties.

Auxiliary chemical substances are essential for debridement of root canals during shaping and cleaning procedures. Sodium hypochlorite (NaOCl), in a concentration range from 0.5% to 5.25%, has traditionally been used for irrigation during root canal treatment because of its antimicrobial activity and ability to dissolve organic matter [12]. Chlorhexidine digluconate (CHX) has been suggested as an auxiliary substance in endodontic treatment because of its antimicrobial activity and substantivity [13,14]. EDTA is indicated as a final irrigating agent for the purpose of demineralizing the dentin and promoting appropriate cleaning of the root canal walls, thus improving the penetration of chemical substances and promoting better contact between the walls of the dentin and the filling material [12].

During the filling procedure, the intimate contact of the root canal sealer with treated root canal dentin should ideally result in excellent mechanical and chemical bonding to ensure adequate sealing [15]. Interestingly, there appears to be an impact of irrigating protocols on the adhesion of sealers to root dentin [15-17]. Moreover, the quality of adaptation between root canal dentin and sealers may also be affected by the moisture condition of the root [16]. In adhesive procedures it was recently proposed to replace residual water with ethanol (EtOH) prior to the application of bonding agents to improve adhesion [18]. However, there is little data on the bond strength of root canal sealer to EtOH-saturated dentin [16].

Therefore, the aim of this study was to evaluate the effect of dentin drying following use of different chemicals on bond strength of two root canal sealers. The hypotheses were that: 1) there was no significant difference in push-out bond strength between sealers, 2) EtOH would improve the bond strength of root canal sealers to root dentin.

Methods

Specimen Preparation

Seventy freshly extracted bovine incisors with anatomically similar root segments and fully developed apices were selected. Teeth were stored in 0.02% thymol solution and prepared within 1 month of extraction. Each tooth was decoronated below the cemento-enamel junction perpendicular to the longitudinal axis using a slow-speed, water-cooled diamond disc (Isomet 2000; Buehler Ltd., Lake Bluff, IL). The roots were cut to a uniform length of 14 mm from the apical end. The inclusion criteria for the roots were as follows: canals up to 2 mm in cervical diameter and at least 15 mm in root length. Sixty roots were used for bond strength tests. The ten remaining roots were used for analysis of interfacial morphology.

All root canals were prepared by a single trained operator. All teeth were instrumented with K-files (Maillefer, Ballaigues, Switzerland) using the crown-down preparation technique. The apical stop was established using files up to size 55 followed by a step-back instrumentation, which ended after the use of 3 files larger than the last file used for apical preparation.

The roots were then randomly divided into 5 groups of 12 roots each according to the different auxiliary chemical substances. The regimen used were in Table 1.

Table 1. Experimental Groups

Group	Chemical auxiliary	Irrigating solution	Final irrigation	Root drying
1	NaCl	NaCl	EDTA	Paper points
2	CHX	NaCl	EDTA	Paper points
3	CHX	NaCl	EDTA	EtOH + paper points
4	NaOCl	NaOCl	EDTA	Paper points
5	NaOCl	NaOCl	EDTA	EtOH + Paper points

NaCl, sodium chloride; EDTA, 17% ethylenediaminetetraacetic acid; CHX, 2% chlorhexidine digluconate; NaOCl, 5.25% sodium hypochlorite; EtOH, ethanol.

In group 1, NaCl was used as an auxiliary chemical substance as well as the irrigating solution. CHX gel in groups 2 and 3 was the auxiliary chemical used with the endodontic instrument for root canal preparation; NaCl was the irrigating solution used to remove CHX and the material originating from the instrumentation of the root canal. In groups 4 and 5, NaOCl was used as the auxiliary chemical substance as well as the irrigating solution.

Each group was further subdivided into two subgroups according to the root canal sealer used. Obturation procedures were performed by using the gutta-percha lateral condensation technique with medium cone calibrated in #55. In subgroup a, the root canals were filled using gutta-percha and AH plus root canal sealer (Dentsply, Detrey, GmbH, Germany). The root canal sealer was mixed and injected into the prepared root canal with a Lentulo spiral. A gutta-percha master cone was lightly coated with sealer and inserted to the working length. In subgroup b, the same protocol was used as in subgroup a; however, the root canal sealer used was MTA Fillapex. Thereafter, the specimens were stored at 37°C and 100% humidity for 7 days to ensure complete setting of the materials.

Evaluation of Bond Strength

Each root was cut horizontally with a slow-speed, water-cooled diamond saw (Isomet 2000) to produce five slices approximately 1 mm thick from each root. Six slices were obtained from each root canal. The first slice was excluded. Thus, five slices were considered from each root canal (n=30 sections/group).

The push-out test was performed by applying a load at 0.5 mm/min to the apex in the direction of the crown until the filling system segment was dislodged from the root slide. Care was also taken to ensure that the contact between the punch tip and filling system occurred over the greatest extended area possible to avoid any notching effect of the punch tip into the filling system's surface.

To express the bond strength in megapascals (MPa), the load at failure recorded in newtons (N) was divided by the area (mm²) of the root canal sealer-dentin interface. To calculate the bonding area, we used the formula $\pi(R+r)[(h^2+(R-r)^2)]^{0.5}$, where R represents the coronal root canal radius, r the apical root canal radius and h the thickness of the slice. The thickness of each slice was measured using a digital caliper (Vonder, Curitiba, PR, Brazil), and the total bonding area for each root canal segment was measured under $\times 20$ magnification with a stereoscope (Lambda Let 2, ATTO Instruments Co. – Hong Kong, China) and evaluated with ImageLab 2.3 software (University of São Paulo – São Paulo, SP, Brazil).

Analysis of Failure Modes

After each measurement of bond strength, both sides of each slice were examined under a light microscope at $\times 40$ magnification to determine the nature of the bond failure. Each sample was evaluated and placed into one of three failure modes: 1) adhesive failure at the filling material/

dentin interface, 2) cohesive failure within the filling material, and 3) mixed failure in both the filling material and dentin [19].

Confocal Laser Scanning Microscopy (CLSM) Analysis

One root from each group was prepared for evaluation of the interfacial morphology using a confocal laser-scanning microscope (CLSM). Prior to application, the root canal sealer had previously been stained with a fluorescent dye (Rhodamine B), which allowed for fractographic examination due to the spectral excitation. The fluorescent dye was added to the root canal sealer only for the CLSM analysis, not for the bond strength measurements. Therefore, the dye did not have any influence on the bond strength test.

Statistical Analysis

All bond strength data were analyzed with the BioEstat 2.0 program (CNPq, 2000 – Brasília, DF, Brazil). Two-way analysis of variance (ANOVA) was used to determine whether a statistically significant two-factor interaction existed between sealers and the auxiliary chemical substances ($p < 0.05$). Statistical comparisons within and between the test groups were made using Tukey's test ($\alpha = 0.05$).

Results

The means and standard deviations are presented in Table 2. The irrigating protocols did not influence the push-out bond strength of either sealer. AH Plus sealer showed higher values of bond strength to root dentin than MTA Fillapex ($p > 0.05$).

Table 2. Bond Strength Means (MPa) and Standard Deviations (\pm) According to Treatments

Groups	AH Plus	MTA Fillapex
NaCl (control)	2.07 (0.40) ^a	0.18 (0.08) ^b
CHX + EDTA	1.97 (0.48) ^a	0.21 (0.06) ^b
CHX + EDTA + EtOH	1.81 (0.44) ^a	0.13 (0.04) ^b
NaOCl + EDTA	1.91 (0.44) ^a	0.20 (0.10) ^b
NaOCl + EDTA + EtOH	1.88 (0.48) ^a	0.16 (0.07) ^b

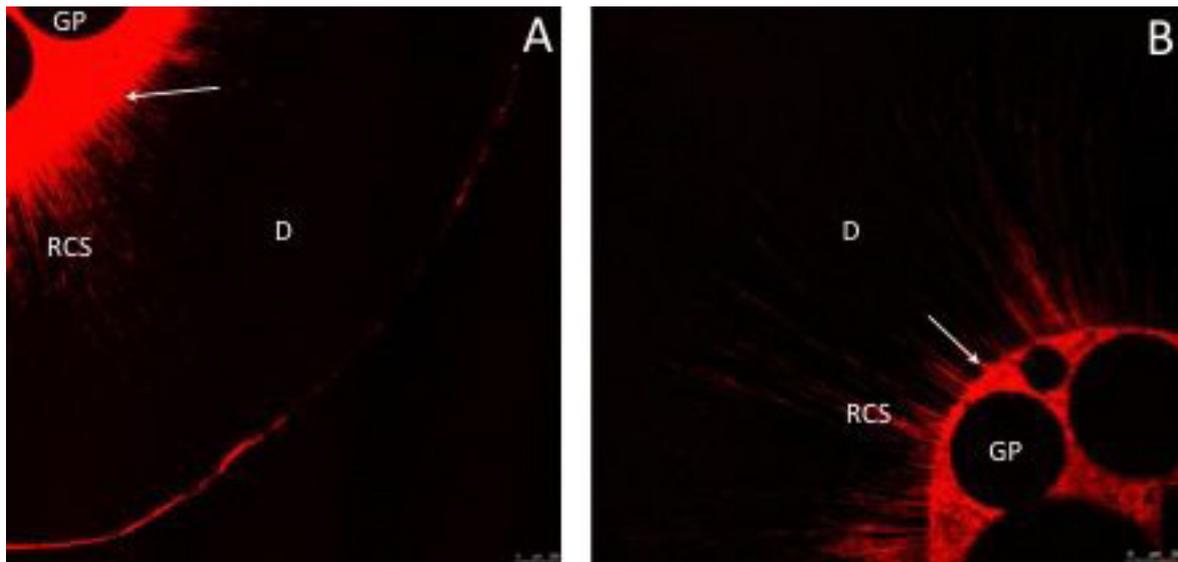
Different letters indicate a statistically significant difference at the 5% level.

Table 3 shows the failure modes observed in each group. The predominant failures were mixed in all groups. The use of CHX followed by EDTA triggered a large number of cohesive failures with both root canal sealers.

On the basis of the CLSM analysis, the interfacial micromorphology between the root canal sealer and root canal dentin is shown in Figure 1. AH Plus exhibited a more homogeneous layer of root canal sealer on dentin when compared to MTA Fillapex. Furthermore, AH Plus showed better sealer penetration into dentinal tubules than MTA Fillapex. The different auxiliary chemical substances revealed similar patterns among the control and experimental groups.

Table 3. Failure Mode Distribution in the Experimental Groups (n=30)

Root Canal Sealers	Auxiliary Chemical Substances	Failure Mode		
		Type 1: Adhesive	Type 2: Cohesive	Type 3: Mixed
AH Plus	NaCl	8	3	19
	CHX + EDTA	0	20	10
	CHX + EDTA + EtOH	0	1	29
	NaOCl + EDTA	5	1	24
	NaOCl + EDTA + EtOH	6	3	21
MTA Fillapex	NaCl	2	6	22
	CHX + EDTA	0	12	18
	CHX + EDTA + EtOH	1	0	29
	NaOCl + EDTA	6	0	24
	NaOCl + EDTA + EtOH	5	0	25

**Figure 1.** Confocal laser scanning microscopic image showing dentin-sealer interface. (A) AH Plus. (B) MTA Fillapex. Arrows indicate sealer. (D) dentin. (RCS) root canal sealer inside the dentin.

Discussion

The bond between root canal sealers and root canal walls is important in both static and dynamic situations. In a static situation, it can eliminate spaces that allow the movement of fluids and microorganisms between the dentin and the filling material. In a dynamic situation, is necessary to maintain the integrity of the sealer-dentin interface during mechanical stresses caused by tooth flexure, operative procedures, or subsequent preparation of a post space [2]. Furthermore, Hammad et al. [20] and Topçuoğlu et al. [21] showed that the filling of roots with resin-based materials increased the resistance of root-canal-filled teeth to vertical root fracture.

In light of the results, the 1) hypothesis that there was no difference between the sealers was rejected. The push-out bond strength of AH Plus was significantly superior to that of MTA Fillapex. Several studies have reported that the push-out bond strength of AH Plus is superior to those of other root canal sealers [6,7]. AH Plus is well known

for its dimensional stability and expansive properties and is considered the 'gold standard' root canal sealer [6,8]. The optimal filling shown by AH Plus may be related to its expansion [22] and ability to bond to dentin [6,8]. Balguerie et al. [23] showed that the tubular penetration and adaptation varies with the different physical and chemical properties of the sealers used. These authors showed that AH Plus exhibited the most optimal tubular penetration and adaptation to the root canal wall among the sealers tested. This is consistent with our study, because AH Plus showed better penetration into dentinal tubules compared with MTA Fillapex sealer (Figure 1). Epoxy resin-based sealers penetrate deeper into dentinal tubules owing to their flowability and long polymerization time, which contribute to enhancing the mechanical interlocking between sealer and dentin. In addition, the cohesion amongst the sealer molecules increases the bond strength value of the material on dentin surfaces, which translates into greater adhesion [8].

In the present study, MTA Fillapex had the lower bond strength to root dentin. The chemical composition of the MTA-based sealer could also influence its bonding behavior. Sarkar et al. [24] suggested that release of calcium and hydroxyl ions from the set sealer results in the formation of apatites as the material comes into contact with phosphate-containing fluids. Reyes-Carmona et al. [25] reported that the apatite formed by MTA and phosphate-buffered saline was deposited within collagen fibrils, promoting controlled mineral nucleation on dentin, seen as the formation of an interfacial layer with tag-like structures. The reason for the low bond strength of MTA Fillapex was claimed to be the low adhesion capacity of tag-like structures because of apatite formation by MTA [26]. In our study, penetration of the MTA Fillapex sealer in the dentinal tubules was poorer than with AH Plus (Figure 1), which can also help explain the low bond strength of the sealer to root dentin. Furthermore, Borges et al. [7] studied the physicochemical properties of MTA Fillapex. Exposing MTA Fillapex specimens to a solubility test in which it was suspended in deionized water for 7 days revealed porosities and cracks in the resin matrix under scanning electron microscopy. Energy-dispersive spectroscopy confirmed this observation by a decrease in the carbon content that was suggested to be caused by polymer degradation.

In the present study, the bond strengths of the tested obturation systems were found to be similar with the different irrigation regimens applied. These results are partly consistent with Nassar et al. [27] who observed that CHX had neither a negative nor positive influence on bond strength. However, this author also showed that NaOCl decreased the bond strength of the sealer to dentin. NaOCl causes problems when used with adhesive resins. Because it is a strong oxidizing agent, it leaves behind an oxygen rich layer on the dentin surface that results in reduced bond strengths [27]. In our study, the different irrigators did not affect the bond strength. This result might be attributed to the altered characteristics of dentin treated with EDTA. It has been demonstrated that EDTA decreases the wetting ability of dentin [28]. Therefore, a suitable dentin surface could be provided for the adhesion of sealers. However, the use of CHX and EDTA resulted in a large amount of cohesive failures, suggesting a better interaction between cement and dentin. Larger numbers of mixed and adhesive failure modes were observed with the NaOCl and EDTA groups, implying that the weak link was the bond between the sealers and the root canal dentin (Table 2).

The second hypothesis that EtOH could be used to improve the bond strength of root canal sealers to root dentin was rejected. In the present study, the use of EtOH had neither negative nor positive influences on bond strength. The use of ethanol aims to remove excess water from root dentin to allow better penetration of the sealer, and render the adhesive interface more stable over time [18]. However, in this study, the use of 100% EtOH for 1 minute on root dentin did not increase the values of the bond strength sealers. Sadek et al. [29] recommended the use of five ascending

EtOH concentrations for the EtOH wet bonding technique, with absolute EtOH reapplied three times, analogous to the technique used in electron microscopy for tissue embedding [30]. Ethanol has a vapor pressure of 52.50 mmHg at 23.8 °C, whereas water has a value of 21.05 mmHg. This means that EtOH evaporates much more quickly than water [31]. Sixty seconds might not have been enough time for complete replacement of water within the intertubular dentin and the dentinal tubules by EtOH; therefore, residual water was present within the root dentin.

Conclusions

Within the experimental conditions of this study, it was observed that the different auxiliary chemical substances and use of ethanol on drying dentin had neither negative nor positive influences on the bond strengths of the AH Plus and MTA Fillapex sealers. Moreover, MTA Fillapex had lower push-out bond values to root dentin compared with AH Plus. However, further studies are required to analyze the ideal irrigant for root canal preparation and clarify the physicochemical and biological properties of the MTA Fillapex sealer.

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