

Effect of endodontic irrigating solutions on the adhesive bond strength to dentin

Efeito de soluções irrigadoras endodônticas na resistência adesiva à dentina

Abstract

Purpose: To evaluate the bond strength of a total-etching adhesive system to dentin irrigated with 1% sodium hypochlorite (NaOCl) and ethylenediaminetetraacetic acid 17% (EDTA).

Methods: Thirty human molars were sectioned 3mm below the occlusal surface and were randomly divided into 3 groups (n=10): G1, no treatment (control); G2, 1% NaOCl; G3, NaOCl followed by EDTA. The specimens received the total-etching adhesive system, restored with microhybrid composite resin, sectioned and trimmed to create 4 hourglass-shaped slabs of each tooth. The slabs were tested in microtensile bond strength in a universal testing machine (Emic DL 2000) at a crosshead speed of 0.5 mm/min until fracture. The results were analyzed statistically by ANOVA and Newman-Keuls test. G2 samples were not submitted to the test because they fractured just before testing, and its values were considered zero to the statistical analysis.

Results: The means and standard deviations (MPa) were: G1, 8.41 (± 3.51); G2, 0.0 (± 0.00); G3, 8.47 (± 3.53).

Conclusion: It was concluded that the application of the NaOCl irrigating solution significantly decreased the bond strength values. The use of NaOCl followed by EDTA resulted in bond strength values not statistically different from control group.

Key words: Adhesives; dentin; root canal irrigants

Resumo

Objetivo: Avaliar a resistência de união de um sistema adesivo condicione-e-lave à dentina irrigada com hipoclorito de sódio 1% (NaOCl) e ácido etilendiaminotetracético 17% (EDTA).

Metodologia: Trinta molares humanos foram seccionados 3mm abaixo da superfície oclusal e aleatoriamente divididos em 3 grupos (n=10): G1 (controle), nenhum tratamento; G2, NaOCl; G3, NaOCl seguido por EDTA. Os espécimes foram hibridizados com um sistema adesivo condicione-e-lave, foram restaurados com resina composta microhíbrida e seccionados de forma a originar 4 amplulhetas de cada dente. As fatias foram submetidas ao teste de resistência de união à microtração em máquina universal de ensaios (EMIC DL 2000) à uma velocidade de 0,5 mm/min até o momento da fratura. Os resultados foram analisados estatisticamente por ANOVA seguido por Newman-Keuls. As amostras do G2 não foram submetidas aos testes de resistência de união pois fraturaram antes dos testes, e seus valores foram considerados zero para análise estatística.

Resultados: A média e desvios-padrão foram (MPa): G1 – 8,41 ($\pm 3,51$); G2 – 0,0 ($\pm 0,00$); e G3 – 8,47 ($\pm 3,53$).

Conclusão: Pode-se concluir que a irrigação com NaOCl diminuiu significativamente os valores de resistência de união. O uso de NaOCl seguido por EDTA resultaram em valores de resistência de união sem diferença estatística com o grupo controle.

Palavras-chave: Adesivos; dentina; irrigantes endodônticos

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Introduction

The dentine substrate is a hydrated biologic complex, with differential regional patterns that can be modified by physiological processes, age and disease (1). Bonding to dentin is a very complicated process, mainly due to its predominantly organic content, tubular structure, variety of substrate mineralization degrees and to the external fluids flux (2).

The actual adhesive systems contain substances that prepare the dentinal substrate for bonding. The acid etching removes the mineral portion and exposes the collagen fibers that must be kept moist. The fourth generation adhesive systems bond to the demineralized and moist dentine tissue by means of acetone or alcohol-based primers that remove the water and maintain the collagen network, permitting resinous monomers penetration (3). This is a critical technique because of the subjectivity inherent to the ideal humidity and because of the possibility of maintaining exposed collagen fibers where the microleakage could be initiated (4). Therefore, one of the main reasons of the hybridization success is the achievement of dentine ideal conditions (2).

In the teeth where endodontic treatment is indicated, the combined use of endodontic irrigating solutions that clean, disinfect and shape the root canal system for posterior filling are necessary and essential for the therapy success (5,6).

Among the endodontic irrigating solutions, indicated as auxiliaries for endodontic therapy, sodium hypochlorite is the most used all over the world due to its anti-bacterial and organic tissue dissolving properties, which are directly proportional to its concentration (7).

EDTA is also a solution commonly used in Endodontics. It acts on the dentin mineral content and promotes smear layer removal (8). The use of sodium hypochlorite followed by the final irrigation with EDTA, for smear layer removal, could enhance the contact between intra-canal irrigating solutions and filling materials with dentine walls (9).

However, the use of irrigating solutions during endodontic treatment could modify the dentine structure (10) and interfere on dentin/resin bonding (11-13). Therefore, the purpose of this study was to evaluate the bond strength of a total-etching adhesive system to dentin irrigated with 1% sodium hypochlorite (NaOCl) and ethylenediaminetetraacetic acid 17% (EDTA). The null hypothesis that endodontic irrigants (NaOCl and EDTA) do not affect the bond strength of the self-etching adhesive system to dentin.

Material and Methods

Thirty sound freshly-extracted human third molars were used in this study. Teeth were stored 0.5% chloramine solution at 4°C for 48 h for disinfection. Next, the teeth were cleaned with pumice/water slurry in Robinson brushes (Microdont; Socorro, SP, Brazil) and analyzed under x10 magnifying glass (Carl Zeiss; Jena, Germany) to exclude those with fracture lines or fissures. The teeth were stored in distilled water at 4°C.

Each tooth was vertically embedded in a PVC cylinder (25 mm diameter and 20 mm high) using an auto-polymerizing acrylic resin (Jet Clássico Ind.; São Paulo, Brazil). The occlusal third (3 mm) of the molar crowns was removed by means of a slow speed saw (400 rpm) with a diamond disk (Arotec; São Paulo, Brazil) in a cutting machine (Struers Miniton; Paraná, Brazil) under constant water-cooling. The dentin was flattened with 600 and 1200-grit silicon carbide abrasive papers.

The specimens were randomly distributed into the following groups (n=10): in group 1 (control), no irrigating solution was applied; in group 2, 1% NaOCl (5 mL) was applied to the dentin surface every 5 min for 1 h, simulating the time that NaOCl (based irrigants are usually left in the root canals during endodontic treatment under clinical conditions); in Group 3, 1% NaOCl was applied as described for Group 2, followed by a 5-min final rinse with 17% EDTA (5 mL), simulating the duration of the final flush with this chelating agent during endodontic treatment under clinical conditions. After dentin treatments, all specimens were washed with distilled water for 60 s.

Two coats of the adhesive system (Single Bond, 3M/ESPE; St. Paul, MN, USA) were applied, with a break of 20s after each adhesive coat, and photo-polymerized (Radii, SDI; São Paulo, SP, Brazil) for 20s after the second coat. After hybridization, 3 increments (2 mm each) of composite resin (Z250, 3M/ESPE; St. Paul, MN, USA) were applied over the dentine surface, using a spatula n. ½, completing 6mm high. Each increment was photo-polymerized for 20s. After bonding procedures specimens were stored in a sterilization oven in water for 24h, at 37°C.

After storage in distilled water for 24 h, the specimens were placed in a metallographic sectioning machine (Struers Miniton; Copenhagen, Denmark) and a water-cooled double-faced diamond disk was used to cut sequential longitudinal 1.0-mm-thick sections in a mesiodistal direction. Care was taken not to separate the slices. The specimens were then removed from the acrylic resin base through a transversal section, to obtain resin/dentin slabs measuring approximately 10-mm high, 5-mm wide and 1-mm thick. The slabs were trimmed on both sides of resin-dentin interface with a #1093 FF bur (KG Sorensen; Barueri, SP, Brazil) at a high-speed handpiece (Kavo; Joinville, SC, Brazil) to obtain a 1-mm thick central area and produce standard hourglass-shaped specimens.

The specimens were individually fixed in a metallic device with a cyanocrylate adhesive (Loctite Super Bonder; São Paulo, SP, Brazil) so that the resin/dentin interface remained without any contact for the microtensile test. The metallic device coupled to a universal testing machine (Emic DL 2000; São José of Pinhais, PR, Brazil) and the specimens were subjected to a microtensile strength at a crosshead speed of 0.5 mm/min until fracture. At the moment of fracture, the resistance values were recorded in Newtons (N) by computer software. Before the test, the area was measured with a digital caliper (Vonder Digital Electronic Paquímetro; Curitiba, PR, Brazil) and the bond strength was

calculated in MPa using the following equation: $Rt=F/A$, where Rt is the μ TBS value, F is the force applied and A is the bond area between the dentin and restorative system. The data obtained were subjected to ANOVA and Newman Keuls tests ($\alpha=0.01$).

For scanning electronic microscopy (SEM) analysis, specimens were immersed in 2.5% glutaraldehyde for 12 hours at 4°C for fixation, washed with 20mL buffer solution of sodium cacodylate 0.2M with pH 7.4 for 1 hour, and washed in distilled water three times for 1 minute. For dehydration, the specimens were sequentially immersed in ethyl alcohol (25% for 20 minutes, 50% for 20 minutes, 75% for 20 minutes, and 95% for 20 minutes), and dried at 37°C for 48 hours with silica gel drying pearls. The prepared specimens were gold-sputtered at 10mA for 1 minute and observed by SEM.

Results

Table 1 shows the mean values and standard deviations (MPa) of groups tested in this study. There was no statistical difference between G1, where no treatment was performed and G3, where dentine surface was treated with NaOCl followed by EDTA. However, between G2, where dentinal surface was treated only with NaOCl, and the other groups,

there was statistical difference at a significance level of 1%. An important fact to report is that specimens of G2 were useless just before the test, since they fractured at a minimum touch, not allowing the microtensile testing. It indicates that NaOCl had a strong negative influence on the adhesive/dentin bond. The G2's mean value was considered as zero in the statistical analysis.

Table 1. Microtensile bond strength mean values (MPa) and the respective standard deviations.

Groups	Mean	Standard deviation	P
G1=Coontrol	8.41 ^a	±3.51	
G2=NaOCl	0.00 ^b	±0.00	<0.01
G3=NaOCl+EDTA	8.47 ^a	±3.53	

* Mean followed by the same letters do not show statistical difference

Figure 1 shows by SEM a modified smear layer (1A) that could difficult phosphoric acid etching. It can be observed opened dentine tubules (Fig. 1B) with few pulp tissue remnants and no smear layer. Less resin tags formation are observed in Figure 1C because of the difficulty of the adhesive system to penetrate throughout the smear layer. In the other hand, effective resin tags formations are seen in Figure 1D.

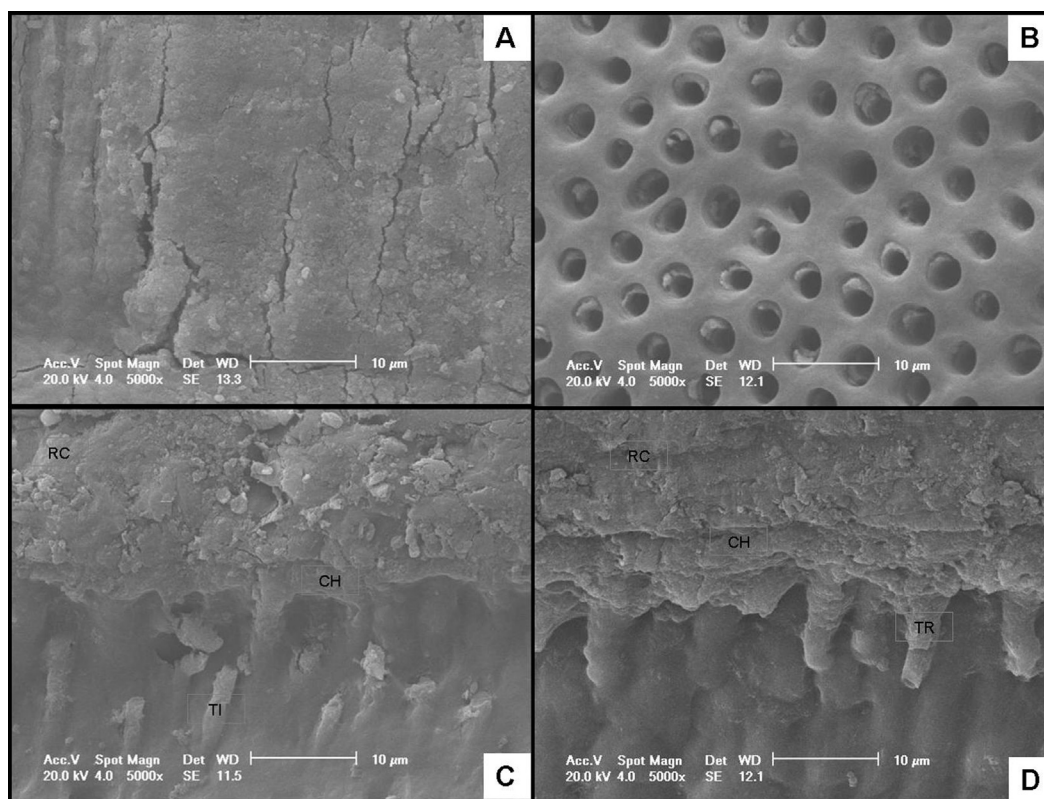


Fig. 1. SEM photomicrographs of dentin after: (A) NaOCl treatment for 1 h (5000x); (B) NaOCl treatment for 1 h followed by EDTA treatment for 5 min (5000x); (C) NaOCl treatment for 1 h (RC – composite resin, CH – hybrid layer, TI – incomplete resin tag (5000x); and (D) NaOCl treatment for 1 h followed by EDTA treatment for 5 min (CR – composite resin, CH – hybrid layer, TR – resin tag (5000x).

Discussion

The results showed that G2, treated only with 1% NaOCl, had its bond strength strongly compromised when compared to the control group G1, and G3 that was treated with EDTA after NaOCl, rejecting the hypothesis under study. This could be caused by organic matrix damage, mainly collagen, after NaOCl application and could also be attributed to the presence of a NaOCl modified smear layer after treatment (Figure 1A). This smear layer could difficult phosphoric acid action and monomers infiltration throughout the demineralized dentin because the sodium chlorite already infiltrated in the smear layer could react and partially neutralize the phosphoric acid action (12). Moreover, NaOCl releases oxygen, and this oxygen could cause a huge inhibition of the adhesive system penetration and polymerization, consequently decreasing the bond strength values (14-16).

The use of EDTA after NaOCl (G3) in dentinal surface resulted in an increase in bond strength, when compared to the groups that received NaOCl treatment only (G2) and these results are in accordance to the study of Lay et al. (15). These authors suggest that the EDTA increases bond strength due to its anti-oxidant power via redox reaction, allowing for free radicals polymerization without premature chains or failures. Moreover, it has the ability to remove the smear layer (Fig. 1B) and the residual chlorine ions, what could benefit phosphoric acid etching and enhance resinous monomer penetration through dentinal tubules (17).

Our results are in agreement with the study of Morris et al. (18) and Vongphan et al. (19) who evaluated the chemical effect of irrigating solutions on microtensile bond strength of adhesive system after acid etching (Single Bond) to dentine walls. They noticed that the use of NaOCl significantly reduced microtensile bond strength. However, the use of 10% sodium ascorbate significantly increased dentin bond

strength after NaOCl treatment. In this case, the 10% sodium ascorbate had a similar effect of EDTA in our study.

Nikaido et al. (12) reported that the NaOCl used during endodontic treatment produces adverse effects on etch-and-rinse adhesive systems. Although the use of EDTA had recovered the adhesive resistance to the reference levels, the use of NaOCl alone had significantly reduced bond strength.

Until recently, endodontically treated teeth were usually treated with a crown, core or dowel (20). However, when they have a sufficient amount of complete coronal structure they can be restored with an adhesive system and resin composite (21,22). It is known that a better bond of restorative materials to dentine walls increases the possibility of marginal sealing, mechanical resistance to mastication stress and durability of restorations (23). The clinical significance of the present study is that the use of EDTA as final irrigating solution during endodontic treatment with NaOCl is fundamental for tooth restoration without compromising the bond strength when using etch-and-rinse adhesive systems.

However, several aspects of the bond strength of a resin composite in dentin need further research, including the effect of various chemicals used during root canal treatment on canal walls. For example, Moreira et al. (24) observed that NaOCl, whether or not in association with 17% EDTA, caused alterations to the dentin collagen, whereas chlorhexidine did not. Furthermore, Cecchin et al. (25) showed that the chlorhexidine pretreatment dentin were capable of preserving the bond strength to root dentin for 12 months. Therefore, different protocols of hybridization of root and coronary dentin must be evaluated to increase the bond strength and long-term adhesion of the resin composite.

In conclusion, the application of the NaOCl irrigating solution significantly decreased the bond strength values. The use of NaOCl followed by EDTA resulted in bond strength values not statistically different from control group.

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