EFFECT OF ACID ETCHING TIME AND TECHNIQUE ON BOND STRENGTH OF AN ETCH-AND-RINSE ADHESIVE

André L. Faria-e-Silva[1], João L. Silva[2], Thauanna G. Almeida[2], Francielle B. Veloso[2], Sandra M. Ribeiro[2], Tiago D. Andrade[2], Bruna V. Vilas-Boas[2], Marisa C. Martins[2], Murilo S. Menezes[3]

[1] Department of Dentistry, Federal University of Sergipe, Aracaju, SE, Brazil.

[2] Department of Dentistry, State University of Montes Claros, Montes Claros, MG, Brazil.

[3] Department of Restorative Dentistry and Dental Materials, Federal University of Uberlândia, Uberlândia, MG, Brazil.

ABSTRACT

The aim of this study was to evaluate the effect of acid etching time and technique on bond strength of a two-step etch-and-rinse adhesive system to dentin and enamel. Thirty human third molars were mesio-distally sectioned, parallel to the long axis of each tooth, in two halves. Buccal/lingual surfaces were abraded to obtain both flat exposed enamel and dentine. The etchant was applied with and without the use of dispensing tips provided by manufacturer. When the tip was not used, the etchant was agitated (active) over the substrate or left undisturbed (passive). The etchings were done for 15 or 30s. After rinsing the acid, the adhesive XP Bond (Dentsply Caulk, Milford, DE, USA) was applied and light-cured. Resin composite cylinders were built up on dentin and enamel substrates. A shear load was applied to the samples at a crosshead speed of 0.5 mm/min until failure. Data were statistically analyzed by three-way ANOVA and Tukey test (α=0.05). There was no difference between the etching techniques in bonding to enamel. Application with the tip or active without the tip promoted higher bond strength to dentin than passive application. Extending the etching time reduced the bond strength to dentin and did not alter the values for enamel. The passive application without tips produced the lowest bond strength when the etchant was applied for 15s. All techniques demonstrated similar values for application during 30s. The acid etching time and technique significantly influence the bond strength of etch-and-rinse adhesive to dentin.

Key-words: Adhesives, Dental bonding, Dental etching.

EFEITO DO TEMPO E DA TÉCNICA DE CONDICIONAMENTO ÁCIDONA RESISTÊNCIA DE UNIÃO DE UM ADESIVO CONVENCIONAL

RESUMO

O Objetivo deste estudo foi avaliar o efeito do tempo e da técnica de condicionamento ácido na resistência de união de um sistema adesivo convencional de dois passos à dentina e ao esmalte. Trinta terceiros molares humanos foram mésio-distalmente seccionados, paralelo ao longo eixo, em duas metades. As superfícies vestibular/lingual foram desgastadas para obter dentina exposta e esmalte planos. O condicionador foi aplicado com ou sem o uso da ponta aplicadora da seringa disponibilizada pelo fabricante. Quando a ponta não foi usada, o condicionador foi agitado (ativa) ou deixado (passiva) sobre o substrato. Os condicionamentos foram realizados por 15 ou 30s. Após a lavagem do ácido, o adesivo XP Bond (Dentsply Caulk, Milford, DE, USA) foi aplicado e fotoativado. Cilindros de resina composta foram construídos sobre a dentina e o esmalte. Carga de cisalhamento foi aplicada sobre as amostras numa velocidade de 0,5 mm/min até a falha. Os dados foram estatisticamente analisados usando ANOVA três fatores e teste de Tukey (α=0.05). Não houve diferença entre as técnicas de condicionamento no esmalte. As aplicações com a ponta ou sem a ponta de forma ativa promoveram maior resistência de união à dentina que a aplicação passiva. Aumentando o tempo de condicionamento reduziu a resistência de união à dentina e não alterou os valores do esmalte. A aplicação passiva sem a ponta produziu os menores valores quando utilizada por 15s. Todas as técnicas demonstraram valores similares na aplicação por 30s. O tempo e a técnica de condicionamento ácido influenciam significativamente a resistência de união de adesivo convencional à dentina.

Palavras chave: Adesivo, Adesão dental, Condicionamento dental.

INTRODUCTION

The fundamental principle of bonding to dental hard tissues is based on micromechanical interlocking of the adhesive resin with dentin/enamel surfaces[1]. While bonding to enamel depends on the micromechanical retention to the etched substrate,[2] bonding to dentin relies on hybridization with the exposed collagen mesh[3]. With etch-and-rinse systems, an acid conditioner selectively dissolves the hydroxyapatite crystals and creates porosities[4]. This step is followed by penetration of the adhesive resin involving the apatite crystal. This approach has shown a high success rate, primarily on enamel, because its mineral content is high[5]. However, the dentinal tissue has a higher content of organic phase and water, representing a challenge to bonding.

The acid etching of dentin removes the mineral phase from the surface layer and exposes the dentinal collagen mesh[3]. Because of the hydrophilic nature of this mesh, it is necessary to use monomers with both hydrophilic and hydrophobic groups for improved adhesion. The hydrophilic functionality permits the permeation of the monomer into the collagen mesh, whereas the hydrophobic functionality facilitates bonding to the hydrophobic resin matrix in the restoration[6]. Thus, the hybridization process is the result of infiltration of adhesive resin into collagen mesh exposed by acid etching followed by its *in situ* polymerization[1, 3].

Acid etching is an essential step for obtaining proper bonding both to dentin and enamel[7,8]. Several factors have been associated with the performance of etchants. The buffering capacity of hydroxyapatite could dramatically limit the reaction of the acid[9]. The smear layer and the presence of dentinal collapsed collagen may also act as a barrier that decreases the demineralization rate[10,11]. Furthermore, limitation could be related to etchants themselves. Most commercial etchants are available as gels at 30 to 40% phosphoric acid. However, it has been demonstrated that the concentration of phosphoric acid is not the only factor responsible for promoting proper etching. Similar concentrations of phosphoric acid etchants containing distinct thickeners result in different demineralization depths as well as different morphology of etched dentin[12]. Thus, the interaction between the etchant and the dental tissue is fundamental to proper etching.

Adequate viscosity permits the etchant to flow and allows a reaction between the phosphoric acid and the dental tissue. Wang & Spencer[13] demonstrated that the technique of etchant gel application on the dentin surface has a significant effect on both the depth and extension of demineralization. The etchant application using dispensing tips had a higher and more homogeneous pattern of etching than when the acid gel came directly from the syringe. The alteration of this physical property is called "shear thinning", where the gel becomes thin and flows like a runny liquid under shear load.[13] However, the same authors showed that the efficacy of etching can be improved even when the etchant gel is used without dispensing tips. Longer duration of etching or the agitation of etchant over the dental tissue improved the performance of the etchant gel. Thus, the aim of this study was to evaluate the effect of acid etching time and technique on the bond strength of adhesive to dentin and enamel. The null hypothesis is that neither time nor etchant application technique hinder bond strength.

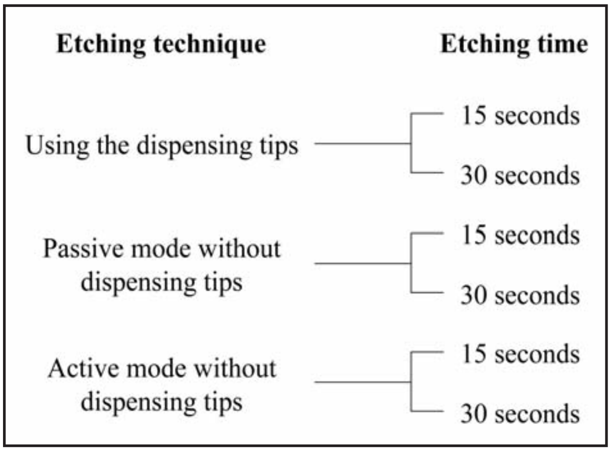

Fig. 1. Experimental design.

MATERIALS AND METHODS

Thirty non-carious human third molars stored in a 0.05% thymol saline solution for no more than 6 months were used. In order to obtain two halves, the teeth were sectioned along the mesio-distal axis, parallel to the long axis of tooth, using a slow speed diamond saw under water cooling. Each half was embedded in acrylic resin (Class-Mold, Clássico, São Paulo, SP, Brazil) to facilitate handling, keeping the buccal/lingual surfaces exposed. Cavity preparation was performed with a cylindrical diamond bur (#3097, KG Sorensen, Barueri, SP, Brazil) operated with a high-speed hand-piece using copious air-water spray. The bur was used parallel to the bucco-lingual surface until a flat surface on both the enamel and dentin was obtained, with sufficient surface area to build up two resin composite cylinders (1 mm in diameter). Afterwards, the surfaces were additionally wet-ground with 320-grit SiC paper (Norton, Lorena, SP, Brazil).

The specimens were randomly allocated according to acid etching time and technique (n=10). Fig. 1 shows the experimental design. The etchant (34% Tooth Conditioner Gel, Dentsply Caulk, Milford, DE, USA) was applied with or without dispensing tips simultaneously over the dentin and enamel. In the latter case, the etchant was placed, spread over the substrates with an applicator brush and either left to stand undisturbed (passive) or applied and gently agitated (active) during the etching time. The etchant remained on the dental tissues for 15 or 30s for all application techniques. The passive application of etchant using a dispensing tip for 15s was considered the control group, as this application technique is recommended by several manufacturers of adhesive systems.

After acid etching, the specimens were rinsed with water for 10s and blot-dried, leaving the dentin moist. The adhesive XP Bond (Dentsply Caulk, Milford, DE, USA) was applied with an applicator brush and left undisturbed for 20s. The solvent was gently evaporated for 5s with directed low-pressure air stream and the adhesive light-cured for 10s. After the adhesive procedures, polyvinyl tubes with a cylinder-shaped orifice (1 mm in inner diameter × 2 mm in height) were individually placed onto the dentin and enamel surfaces. The composite resin Spectrum TPH (Dentsply, Petrópolis, RJ, Brazil) was inserted into the tube and light-polymerized for 20 s. Light-polymerization procedures were performed using the LED unit Optilight LD Max (Gnatus, Ribeirão Preto, SP, Brazil) with approximately 500 mW/cm^2 of irradiance. Afterwards, the tubs were removed to expose the resin cylinders. Four cylinders (two per substrate) were made for each specimen.

The embedded specimens were attached to the testing device and each resin composite cylinder was tested on a mechanical testing machine (EMIC DL 2000, São José dos Pinhais, PR, Brazil). A thin steel wire (0.2 mm in diameter) was looped around each cylinder and a shear load was applied to the base of the cylinder at a crosshead speed of 0.5 mm/min until failure. Shear bond strengths were calculated and expressed in MPa. The average value of the two bonded cylinders for each substrate in the same specimens was recorded as the shear bond strength for that specimen. Data were put into the three-way ANOVA and Tukey's test, at a 95% confidence level. The factors evaluated were "etching technique," "etching time," and "substrate".

RESULTS

The statistical analysis showed a significant effect for all factors ($p < .05$) and interactions between factors ($p < .05$). Comparisons according to the Tukey's test are shown in Tables 1, 2, 3 and 4. Table 1 shows the results for the interaction between factors "etching technique" and "substrate." There was no difference between the etching techniques on bonding to enamel. However, passive application of the etchant without dispensing tips produced the lowest values of bond strength to dentin. There was no difference between substrates when the etchant was applied with agitation. Conversely, the other techniques produced lower bond strength to dentin than enamel.

Table 2 shows the results for the interaction between factors "etching time" and "substrate." The 30-second

Table 1: Bond strength means (SD) for interaction between factors "etching technique" and "substrate" in MPa.

	Etching technique		
Substrate	*With tips*	*Without tips - Passive*	*Without tips - Active*
Enamel	36.7 (10.1) Aa	32.0 (8.3) Aa	33.4 (9.5) Aa
Dentin	29.5 (9.8) Ab	19.4 (7.8) Bb	32.5 (14.1) Aa

Means followed by different capital letters in the same line, and lowercase letters in the same column, are significantly different at $p < 0.05$.

Table 2: Bond strength means (SD) for interaction between factors "etching time" and "substrate" in MPa.

	Etching time	
Substrate	*15 s*	*30 s*
Enamel	34.2 (9.7) Aa	33.8 (9.2) Aa
Dentin	30.8 (14.5) Aa	23.5 (7.8) Bb

Means followed by different capital letters in the same line, and lowercase letters in the same column, are significantly different at $p < 0.05$.

Table 3: strength means (SD) for interaction between factors "etching technique" and "etching time" at dentin in MPa.

	Etching technique		
Etching time	*With tips*	*Without tips - Passive*	*Without tips - Active*
15 s	36.6 (9.3) Aa	16.8 (4.7) Ba	37.6 (10.3) Aa
30 s	22. (6.4) Ab	23.0 (6.9) Aa	23.8 (8.2) Ab

Means followed by different capital letters in the same line, and lowercase letters in the same column, are significantly different at $p < 0.05$.

Table 4: Bond strength means (SD) for interaction between factors "etching technique" and "etching time" at enamel in MPa.

	Etching technique		
Etching time	*With tips*	*Without tips - Passive*	*Without tips - Active*
15 s	39.4 (8.6) Aa	30.4 (8.9) Aa	34.4 (8.8) Aa
30 s	34.4 (9.0) Aa	32.6 (9.4) Aa	36.0 (9.6) Aa

Means followed by different capital letters in the same line, and lowercase letters in the same column, are significantly different at $p < 0.05$.

etching time produced lower bond strength to dentin than the 15-second etching time. There was no significant difference between times for bonding to enamel. Within 30s of etching, the bond strength of the adhesive was higher to enamel, while the values were similar for both substrates when the etchant was applied for 15s.

Table 3 shows the results for interaction between factors "etching technique" and "etching time" at dentin. There was no difference between the techniques when the etchant was applied for 30s. However, the passive application of the etchant without dispensing tips for 15s produced the lowest bond strength compared to other techniques used for the same time. Etchant use for 30s did not alter the bond strength for the passive use of an etchant without dispensing tips, while there were reduced values for the other techniques. Table 4 shows the values obtained at enamel. The bond strength was similar for all experimental conditions.

DISCUSSION

Most available etchants on the market contain additional thickeners in order to facilitate handling. The advantages of the gel form are that the clinicians can easily control the placement of the acid on the required dental tissue surface; without which the etchant flows to other surfaces. The etching gels are commonly thickened with the addition of silica or polymers. However, acid gels have a lower diffusion rate than acid liquid etchant.[12] Pushing the gel against the nozzle of the dispenser tip reduces its viscosity and can improve its effectiveness.[13] The results of the present study show that using dispensing tips affected the bond strength to dentin. Thus, the null hypothesis was rejected.

The etching technique presented a significant effect only on dentin, while all techniques promoted similar values of bond strength to enamel. Enamel bonding has traditionally been dependent upon the infiltration of adhesive resin into surface porosity created by acid etchant agents. Etchants dissolve hydroxyapatite crystals in enamel, creating pits through which the adhesive resin is readily absorbed by capillary attraction[14]. The resin tags provide the bond strength to this substrate[1,2]. The efficient etching of enamel seems more related to the pH of the etchant versus other factors[15,16]. Furthermore, it has been demonstrated that longer times do not significantly improve the bond strength[17]. The same can be expected for viscosity and application mode. A possible deeper etching promoted by the lowest viscosity of etchant or by active application does not result in higher bond strength[8,17]. In contrast to enamel, dentin is a more heterogeneous substrate, consisting of hydroxyapatite and collagen fibrils. The acid conditioning of dentin leaves a wide opening in the dentinal tubules, which exposes a layer of mineral depleted collagen fibrils[18]. An effective dentin bond depends on proper adhesive resin penetration into the partially demineralized dentin to form the so-called hybrid layer[3,19]. In this approach, the exposed collagen fibrils function as a micro-retentive network for mechanical interlocking. It has been demonstrated that penetration of acid etchant occurs primarily along the tubules, while the penetration on intertubular dentin occurs at a lower rate[12]. The latter seems be essential for providing proper bond strength once it creates a space between collagen fibrils[3].

Leaving the etchant applied without a tip on the dentin undisturbed resulted in the lowest bond strength. The passive application of the etchant was found to promote only a non-uniform and partial demineralization of dentin[13]. This non-effective etching may be responsible for the low bond strength using this etching technique. However, agitating the etchant improved the bond strength to dentin. This agitation helps to replenish H^+ ions of dentin and to remove the reaction products, resulting in more effective etching[13]. However, etchant application with dispensing tips increases its flow and dispenses its agitation on the substrate.

The outcomes of the present study show that extending the etching time of dentin to 30s reduces the bond strength. Conversely, time did not hinder bonding to enamel. It has been demonstrated that increasing the etching time on enamel can lead to the formation of an insoluble reaction product and result in weaker bond strength[20]. However, an upper limit of 60s for enamel time is generally well agreed upon[17]. Thus, the etching times used in this study are safe for producing proper bonding.

Within dentine, extending the etching time promotes deeper demineralization[13,21]. In etch-and-rinse adhesives, there is often discrepancy between the depth of dentin demineralization and adhesive resin infiltration[22]. Higher discrepancy is expected with deeper demineralization[21]. Thus, the increase of etching time to dentin results in a thicker layer of an unprotected mineral-depleted collagen at the base of the hybrid layer. This mineral-depleted layer can act as weak link during shear testing and reduce the bond strength[21]. Another explanation may to relate to possible collagen damage during acid etching[23,24]. Longer etching time could damage the tertiary structure and result in poorer resin infiltration.

The outcomes also demonstrated significant interaction between etching time and technique only for bonding to dentin. Extending the etching time reduced the bond strength for application with tip as well as for application without tip but with agitation (active mode). This reduction could be related to poorer bonding achieved on dentin with longer application times. The same did not occur when the etchant was applied without the tip in the passive mode. Despite the numerical increase of bond strength, there was no difference for the latter technique when applied for 15 or 30s. Thus, the worst efficiency on dentin of etchant applied passively without the dispensing tips was not compensated by an increase in etching time.

In several clinical conditions, proper bonding to dentin substrate is essential to promote the retention of restoration or adequate seal. In contrast to enamel, the etching technique of etchant application is essential for reaching a proper bonding to dentin. The outcomes of this study showed that the use of etchants with their dispensing tips can produce proper bonding to both enamel and dentin. Furthermore, a time conditioning of 15s promoted the best results in both substrates. However, in contrast to enamel, the etching time should not exceed 15s on dentin. In the absence of dispensing tips, the agitation of the etchant on the dentin promoted similar results.

CONCLUSION

Based on the results of the present study, it can be concluded that:

The application mode of the etchant had a significant effect only on bond strength to dentin.

The use of dispensing tips and agitation of etchant (applied without the tips) promoted the highest values of bond strength to dentin.

The etching time did not influence the bond strength to enamel.

Extending the etching time to dentin reduced the bond strength, except for the passive application of etchant without the dispensing tips. For this technique, the longer time did not affect bond strength.

CORRESPONDENCE
André Luis Faria-e-Silva
Departamento de Odontologia
Centro de Ciências Biológicas e da Saúde
Universidade Federal de Sergipe
Rua Cláudio Batista, s/n – Sanatório
49060-100 Aracaju-SE, Brazil
andrelfsilva@hotmail.com

REFERENCES

1. Van Meerbeek B, De Munck J, Yoshida Y, et al. Buonocore memorial lecture. Adhesion to enamel and dentin: current status and future challenges. Oper Dent 2003;28:215-235.
2. Buonocore MG. A simple method of increasing the adhesion of acrylic filling materials to enamel surfaces. J Dent Res 1955;34:849-853.
3. Nakabayashi N, Kojima K, Masuhara E. The promotion of adhesion by the infiltration of monomers into tooth substrates J Biomed Mater Res B Appl Biomater 1982; 16:265-273.
4. Erickson RL, Barkmeier WW, Latta MA. The role of etching in bonding to enamel: a comparison of self-etching and etch-and-rinse adhesive systems. Dent Mater 2009; 25:1459-1467.
5. Peumans M, Kanumilli P, De Munck J, et al. Clinical effectiveness of contemporary adhesives: a systematic review of current clinical trials. Dent Mater 2005; 21:864-881.
6. Reis A, Pellizzaro A, Dal-Bianco K, Gones OM, Patzlaff R, Loguercio AD. Impact of adhesive application to wet and dry dentin on long-term resin-dentin bond strengths. Oper Dent 2007; 32:380-387.
7. Jacques P, Hebling J. Effect of dentin conditioners on the microtensile bond strength of a conventional and a self-etching primer adhesive system. Dent Mater 2005;21:103-109.
8. Barkmeier WW, Erickson RL, Kimmes NS, Latta MA, Wilwerding TM. Effect of enamel etching time on roughness and bond strength. Oper Dent 2009;34:217-222.
9. Camps J, Pashley DH. Buffering action of human dentin in vitro. J Adhes Dent 2000;2:39-50.
10. Wang Y. Spencer P. Analysis of acid-treated dentin smear debris and smear layers using confocal Raman microspectroscopy. J Biomed Mater Res 2002; 60:300-308.
11. Spencer P, Wang Y, Walker MP, Swafford JR. Molecular structure of acid-etched dentin smear layers – in situ study. J Dent Res 2001;80:1802-1807.
12. Perdigão J, Lambrechts P, Van Meerbeek, Tomé AR, Vanherle G, Lopes AB. Morphological field emission-SEM study of the effect of six phosphoric acid etching agents on human dentin. Dent Mater 1996;12:262-271.
13. Wang Y, Spencer P. Effect of acid etching time and technique on interfacial characteristics of the adhesive-dentin bond using differential staining. Eur J Oral Sci 2004;112: 293-299.
14. Marshall SJ, Bayne SC, Baier R, Tomsia AP, Marshall GW. A review of adhesion science. Dent Mater 2010; 26:e11-16.
15. Perdigão J, Gomes G, Lopes MM. Influence of conditioning time on enamel adhesion. Quintessence Int 2006;37:35-41.
16. Ostby AW, Bishara SE, Denehy GE, Laffoon JF, Warren JJ. Effect of self-etchant pH on the shear bond strength of orthodontic brackets. Am J Orthod Dentofacial Orthop 2008; 134:203-208.
17. Kimmes NS, Barkmeier WW, Erickson RL, Latta MA. Adhesive bond strengths to enamel and dentin using recommended and extended treatment times. Oper Dent 2010; 35:112-119.
18. Fawzy AS, Farghaly AM. Probing nano-scale adhesion force between AFM and acid demineralized intertubular dentin: Moist versus dry dentin. J Dent 2009;37:963-969.
19. Takagaki T, Nikaido T, Tsuchiya S, Ikeda M, Foxton RM, Tagami J. Effect of hybridization on bond strength and adhesive interface after acid-base challenge using 4-META/MMA-TBB resin. Dent Mater J 2009;28:185-193.
20. Sadowsky PL, Retief DH, Cox PR, Hernández-Orsini R, Rape WG, Bradley EL. Effects of etchant concentration and duration on the retention of orthodontic brackets: an in vivo study. Am J Orthod Dentofacial Orthop 1990;98:417-421.
21. Hashimoto M, Ohno H, Kaga M, et al. Over-etching effects on micro-tensile bond strength and failure patterns for two dentin bonding systems. J Dent 2002;30:99-105.
22. Sano H, Shono T, Takatsu T, Hosoda H. Microporous dentin zone beneath resin-impregnated layer. Oper Dent 1994; 19:59-64.
23. Habelitz S, Balooch M, Marshall SJ, Balooch G, Marshall GW Jr. In situ atomic force microscopy of partially demineralized human dentin collagen fibrils. J Struct Biol 2002; 138:227-236.
24. Brajdi D, Krznarić OM, Azinović Z, Macan D, Baranović M. Influence of different etching times on dentin surface morphology. Coll Antropol 2008;32:893-900.