EFFECTS OF CAVITY CLEANSING AGENTS ON BOND STRENGTH OF TWO ADHESIVE SYSTEMS TO DENTIN

Efeitos de agentes de limpeza cavitária na resistência de união de dois sistemas adesivos à dentina

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SUMMARY

The aim of this study was to evaluate the effect of cleasing agents and air abrasion on microtensile bond strength of two bonding systems to dentin. Exposed flat dentin surfaces of extracted human third molars were treated with either one of the cleansing agents or air-abrasion before being bonded with either Single Bond or Clearfil Liner Bond 2V adhesive systems, according to manufacturer's instructions. Composite build-ups were incrementally constructed with Z100 resin composite and the teeth stored for 24 hours in water at 37°C. Specimens were then vertically sectioned to obtain several bonded sticks with a cross-sectional area of 1.0 mm^2 and each one stick was tested in tension at 0.6 mm/min. Two-way ANOVA showed significant influence of both factors on bond strength ($p<0.05$). For Single Bond, while none of the cleansing agent adversely affected the bond strength as compared to control values ($p>0.05$), air-abrasion significantly decreased bond strength ($p<0.05$). For Clearfil Liner Bond 2V, the application of either Tergensol or Cav-Clean did not affect bond strength, however, the other treatments caused significant reduction of bond strength ($p<0.05$). These results suggest that application of cavity cleasing agents and air-abrasion prior to bonding may adversely affect the adhesion to dentin. *Clinical Significance*: Application of cavity cleansing agents and air-abrasion prior to bonding may adversely affect the adhesion to dentin. This effect is dependent on the treatment and type of adhesive system used.

UNITERMS

Dentine, Adhesion, Bond strength.

INTRODUCTION

Recognized by their bactericidal / bacteriostatic action or detergent power, cavity cleanser has been recommended after preparation as a treatment for residual bacteria (Meiers & Kresin[16] 1996). Into this group, are classified as non-demineralizing substances germicides, detergents or alkaline solutions. However, there is no conclusive evidence for their effectiveness and little information is available about proper clinical indications for their use (Shortall[26] 1981).

Germicides are considered penetrating agents that can flow into dentinal tubules encountering bacteria and odontoblastic-cell extensions. They are generally products based on ethanol, hydrogen peroxide or chlorhexidine and may prevent bacterial growth in cavities (Chan & Nield[5] 1995). Detergents are used for degreasing prepared surfaces and have low antimicrobial potential. Alkaline agents can switch the acidic pH of the cavity, that is favorable for bacterial proliferation, to an alkaline, bactericidal pH (Forsten & Berhenholtz[7] 1977; Bergenholtz & Reit[2] 1980). Alkaline pH is desirable because of inhibitory effect of $Ca(OH)_2$ on the microbial enzyme activity, probably due to its high pH (ca. 12.0) (Forsten & Berhenholtz[7] 1977). The application of germicides such as 2% chlorhexidine digluconate in preparations has the purpose of eliminating residual bacteria, thus preventing recurrence of caries, especially if an adequate marginal seal against Microleakage is not achieved (Meiers & Shook[17] 1996). Its use as an antiseptic agent before placing adhesive restorations has been previously described (Perdigão *et al*[20] 1994; Meiers & Kresin[16] 1996; Meiers & Shook[17] 1996; Gürgan *et al*[8] 1999). Detergents and alkaline agents are generally used as cavity cleansing in non-adhesive restorations (Bachmann *et al*[1] 1997). Little to none information is available about their effect on adhesive restorations. Moreover, the need for adding disinfectants to the conditioners or even if it is necessary to treat dentin surfaces with separate antimicrobials prior to the placing of etching agents in adhesive restorations have been questioned (Settembrini *et al*[25] 1997).

Air abrasion with aluminum oxide (Al_2O_3) particles has been considered a viable and safe alternative to both prepare dental structure and remove defective restorations (Laurell *et al*[13] 1995). The size particles and air pressure or velocity dictate the using objective. Small particles (27- or 27.5-) cut smoother and faster as the cutting surface is increased, whereas the bigger particles (50-) are slower and heavier with less cutting surface, in addition to needing increased velocity to keep moving (Reyto[21] 2001). Anyway, the particle abrasion produces a rough surface on dental tissues. Because of that, some researchers (Los & Barkmeier[14] 1994; Roeder *et al*[23] 1995; Nikaido *et al*[18] 1996; Rinaudo *et al*[22] 1997; Hannig & Femerling[11] 1998; Manhart *et al*[15] 1999) see this method as a possible way to modify the surface of the dental tissues and improve the performance of adhesive

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systems.

Simplified adhesive systems are currently available in the market. These include the "one-bottle" systems, which combine priming and bonding into one step, while the acid conditioning remains as a separate step; and the "self-etching" systems, which combine conditioning and priming into one step, followed by the application of the bonding agent. In the latter systems, there is no separate acid-etching step and no rinsing is necessary after the application of the self-etching primer to a smear layer-covered dentin surface (Watanabe *et al*[28] 1994; Tanumiharja *et al*[27] 2000).

The application of cleansing agents and air particle abrasion to cavities before bonding may chemically and/or physically alter the dentin surface in a way that could interfere with proper adhesion of the bonding agents. Although such alterations may be eliminated by the etching and rinsing steps of the traditional adhesive systems, there is a risk that surface modifications could affect the adhesion of self-etching systems that are applied directly on the treated surface without a previous etching and rinsing steps.

The purpose of this study was to determine the microtensile bond strength of a conventional and a self-etching adhesive systems to dentin that had been previously treated with several cleansing agents and air particle abrasion. The null hypothesis tested here is that there is no influence of treatments on bond strength of either adhesive to dentin.

MATERIALS AND METHODS

Twelve, non-carious, extracted human third molars were stored in 0.05% thymol solution for a period no longer than 6 months before being used. The teeth were scraped to remove any residual soft tissue and immersed in distilled water for 1 week to remove residual thymol solution. A flat dentin surface was obtained by transversally sectioning the crown with a slow-speed diamond blade (EXTEC Co., Enfield, CT, USA) under water lubrication. The exposed dentin surfaces were polished with wet 320-grit SiC paper to remove remnants of enamel and create a standard smear layer.

The characteristics of the products used in this study are listed in Table 1. All cleansing agents and air abrasion were applied to intact, smear layer-covered dentin surfaces before application of the respective bonding procedures. Cav-Clean comes in pre-filled syringes containing 0.8 ml of the solution. An amount enough to cover the entire surface was dispensed on dentin, left undisturbed for 10 seconds and rinsed off with water syringe (10 sec). Tergensol was applied with a thin brush, left undisturbed for 15 seconds and rinsed off with water (10 sec). Cavidry was applied without surface saturation and left to evaporate for 5 seconds. The 0.2% calcium hydroxide solution was applied with a cotton pellet, left in contact with dentin for 15 seconds and the excess removed with an absorbent paper without rinsing. Air abrasion was perpendicularly applied to the dentin surface with 50 m aluminum oxide particles using an intra-oral sandblaster (Micro-Etcher, Danville Engineering, Danville, CA, USA) for 15 seconds at a 1 cm distance and output pressure of 120 psi, followed by water rinse. Control specimens were prepared on dentin surface pre-treated with distilled water. The dentin surfaces were gently air-dried with air syringe prior to the application of the adhesive systems. The selected bonding systems, Clearfil Liner Bond 2V (CLB), a self-etching primer, and Single Bond (SB), a total etch "one-bottle" adhesive system were applied according to manufacturers' directions. Prior Single Bond (SB) application, 37% acid phosphoric was applied on dentin surface for 15 seconds.

Following the application of the adhesive, Z100 resin composite was incrementally placed on the surface in four 1 mm thick layers that were light cured for 40 seconds each. The light source was a Curing Light XL 3000 (3M ESPE, St. Paul, MN, USA) with energy intensity higher than 450 mW/cm^2, as measured by a curing radiometer (Curing Radiometer, Model 100 P/N 10503, Demetron / Kerr, Danbury, CT, USA). The bonded teeth were stored in distilled water at 37°C for 24 h prior to preparation for testing.

Specimen preparation for microtensile bond strength testing.

The bonded teeth were laterally attached to the arm of a slicing machine and serially, vertically sectioned in both "x" and "y" directions to obtain several bonded sticks with a cross-sectional area of approximately 1.0 mm^2 (Figure 1). Each stick was fixed with cyanoacrylate glue (Zapit, DVA, Corona, CA, USA) to the grips of a Vitrodyne V-1000 (Chantillon, Bros. Greensboro, NC, USA) testing machine and tested in tension at 0.6 mm/min until failure. After fracture, the specimen was removed from the testing apparatus and the cross-sectional area at the site of fracture measured to the nearest 0.01 mm with a digital caliper (Mitutoyo Sul Americana Ltda, Suzano, SP, Brazil) The bond strength was calculated by dividing the maximum load at failure by the cross-sectional area and expressed in MPa. The fractured ends of the specimens were examined under light microscopy at 40X to determine the mode of failure. Failures were classified as being adhesive

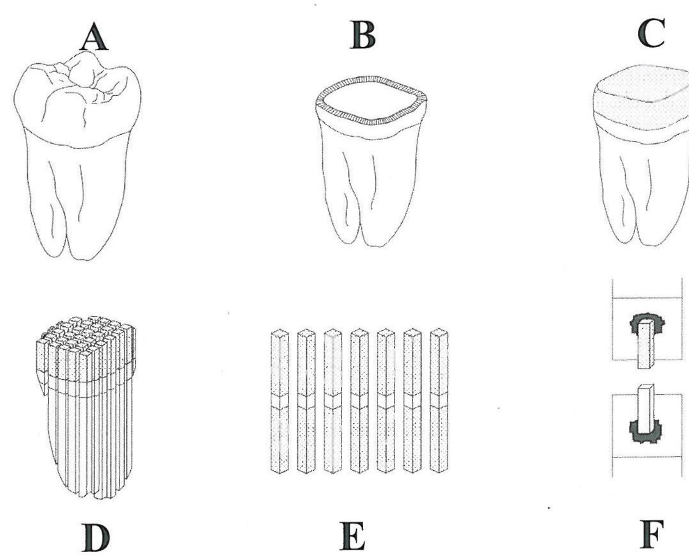

Figure 1 - Schematic on how the specimens were prepared for the microtensile testing. The teeth were transversally sectioned to expose a flat dentin surface (A, B) followed by the application of the experimental treatments, bonding agents and resin build-up crowns (C); After storage, they were longitudinally sectioned in both "x" and "y" directions (D). The bonded sticks were individually tested in tension (E,F).

Table 1 - Materials used in this study.

Product	Manufacturer	pH	Composition
Cav-Clean	HERPO Ltda., Rio de Janeiro, RJ, Brazil	6.6	2% chlorexidine digluconate
Cavidry	DFL Indústria e Comércio Ltda, Rio de Janeiro, RJ, Brazil	9.2	39.6% Trichloroethylene,39.7% isopropyl alcohol, 10.5% benzine, 1.7% amine acetate
Tergensol	INODON, Porto Alegre, RS, Brazil	6.5	0.22% Sodium Lauryl-sulfate
Calcium hydroxide solution	Laboratory made	11.9	0.2% aqueous solution of calcium hydroxide
Aluminum oxide powder	Micro-Etcher, Danville Engineering, San Ramon, CA, USA	—	50 µm aluminum oxide particles
Single Bond	3M ESPE, St. Paul, MN, USA	*3.6*⊗	HEMA, BIS-GMA, polyalkenoate copolymer, photo initiators, stabilizers, water and ethanol
Clearfil Liner Bond 2V	Kuraray Medical Co., Osaka, Japan	*Primer 3.03*•	Primer A – MDP, HEMA, water, PI, accelerators Primer B - HEMA, water, accelerators Bond Liquid A - MDP, HEMA, BIS-GMA, microfiller, PI, accelerators
Z100	3M ESPE, St. Paul, MN, USA	—	BIS-GMA, TEGDMA, photo initiator, stabilizer, inorganic filler (66 vol%): ZrO/SiO_2

Abbreviations: MDP = 10-methacryloyloxydecyldihydrogenphosphate; BIS-GMA =bisphenol A glycidylmethacrylate; HEMA = 2-hydroxyethyl methacrylate; PI = photoinitiator; TEGDMA = tetraethylenglycoldimethacrylate.
•(Sanares et al., 2001) •(Hannig & Femerling, 1998)

when the fracture occurred along the interface; cohesive, when occurred within dentin or resin; and mixed, when occurred along the interface extending to dentin or resin.

Determination of the pH of the cleansing agents

The pH of the cleansing agents was determined in 20 mL aliquots using a pH-meter (pHmetro B371,Micronal, São Paulo, SP, Brazil). Respective values are expressed in Table 1.

Statistical analysis

Bond strength data were analyzed by two-way ANOVA (materials *vs.* treatments) followed by Student-Newman-Keuls *post-hoc* tests. The level of significance was established at =5%.

RESULTS

Microtensile Bond Strength

Mean microtensile bond strength values for both adhesive systems according to the experimental treatments are expressed in Table 2. The application of cavity cleansing agents did not cause any significant change in the bond strength of Single Bond to dentin ($p>0.05$). However, when air-abrasion was applied before bonding with this adhesive, the resultant bond strength dropped significantly ($p<0.05$). For Clearfil Liner Bond 2V, the application of either Tergensol or Cav-Clean resulted in bond strength values that were not different from the control (distilled water) ($p>0.05$).

Table 2 - Bond strength of adhesives according to surface treatments. Values are in MPa±SD.

Treatment \ Adhesive	N	Single Bond		Clearfil Liner Bond 2V	N
Control (distilled water)	9	36.0 ±9.3 [A]		45.6 ±12.4 [a]	9
Tergensol	19	32.1 ±7.3 [A]	*	43.1 ±8.5 [a]	16
Cav-Clean	16	27.1 ±13.0 [A]		34.8 ±12.6 [ab]	20
Calcium hydroxide solution	15	35.8 ±12.6 [A]		27.5 ±10.7 [b]	12
Cavidry	20	27.4 ±13.2 [A]	*	13.6 ±4.2 [c]	21
Aluminum oxide (air abrasion)	18	18.8 ±6.3 [B]		15.9 ±7.1 [c]	13

Same letters indicate no significant differences among treatments ($p>0.05$)
* Indicates significant differences between materials ($p<0.05$).

When calcium hydroxide solution was used, mean bond strength dropped significantly when compared to the control or Tergensol treatments ($p<0.05$). However, it was not statistically different from the means obtained with Cav-Clean ($p>0.05$). Significantly lower mean bond strengths were obtained when dentin was treated with either Cavidry or air-abrasion and bonded with CLB ($p<0.05$). In general, mean bond strengths between materials for each treatment were similar, except for Tergensol and Cavidry groups, which had statistically significant values between bonding agents evaluated. The failure modes observed were mostly adhesive or mixed for both materials. After treatments, CLB showed a relatively higher percentage of adhesive failures than SB.

DISCUSSION

The bacteria left in the cavity floor can live for a long time and an incomplete removal of infected dentin would reduce the longevity of restoration by problems associated with recurrence decay (Boston & Graver[4] 1989; Meiers & Kresin[16] 1996). The method used to detect carious dentin using dyes does not necessarily result in removal of all bacteria (Boston & Graver[4] 1989). The use of cavity cleansing agents after tooth preparation is advocated as an additional aid to reduce the number of microorganisms on the tooth surface and postoperative sensitivity as consequence (Meiers & Shook[17] 1996). However, the procedure may switch from beneficial to detrimental if it interferes with the bonding mechanism in adhesive restorations. The ideal moment for the application of cavity cleansing or antibacterial agents during adhesive restorative procedures is still unclear. It conducted some in vitro studies that evaluate the effects of such solutions on bond strength immediately after cavity preparation, prior to any conditioning or priming of the smear layer (Meiers &[17] Shook, 1996), an also after conditioning dentin with acidic agents (Perdigão *et al*[20] 1994; Gwinnett[9] 1992a). Additionally, some evaluate if rinsing off or not the desinfectant before the bonding procedure would affect bonding process (Perdigão *et al*[20] 1994; Meiers & Shook[17] 1996; Gürgan *et al*[8] 1999). When total etch adhesive systems are used, both application procedures are possible, however, when a self-etching system is the adhesive of choice, cleansing agents and air abrasion are not to be applied after priming the surface. In order to be consistent, we chose to apply the experimental treatments immediately after smear layer preparation and before etching procedure. Moreover, we followed classical dentistry basis where cavity disinfection step should be done prior to start any restorative sequence (Brännström *et al*[3] 1980).

In this study, only Tergensol and Cav-Clean did not cause any statistically effect on the bond strengths obtained with both adhesive systems used. Cav-Clean is a 2% water solution of chlorhexidine digluconate. Since most of its composition is water, it was expected that it would not interfere with bonding for both adhesive systems. The chlorhexidine solution was rinsed off the surface before the application of the adhesive systems. Gürgan *et al*[8] (1999) claimed that rinsing is important to avoid decreasing bond strengths due to formation of an acid-resistant layer by the agent. We do not believe this is the case because other studies have demonstrated no effects of chlorhexidine solutions on bond strength, even when the solution was not rinsed off the surface (Meiers & Kresin[16] 1996; Meiers & Shook[17] 1996). Tergensol is not ordinarily used as a cavity cleansig agent for adhesive restorations. However, Bachmann *et al*[1] (1997) reported an adverse effect on bond strength of All Bond 2 adhesive system to dentin that had been previously treated with 5% sodium lauryl sulfate (cationic soap). They recommended that soaps or detergents should not be used as a clinical mean to remove remnants of provisional cements prior to adhesive cementations. This study showed that Tergensol had no influence on bond strength of the two adhesive systems to dentin. Although it is a detergent-based agent and could alter the physico-chemical interaction of the bonding agents with dentin, it was rinsed off the surface before bonding and eventual effects may have been eliminated by the water rinse. Calcium hydroxide solution as a cleansing agent is beneficial due to precipitation in contact with dentin surface and its bactericidal potential due to the very high pH (Forsten & Karjalainen[7] 1977). Since the solution was not rinsed off the surface, scattered particles may have remained on the surface. This cleansing agent decreased bond strength only when CLB was used. The calcium hydroxide solution is highly alkaline and may have markedly raised the pH of the surface that immediately neutralized the acidity of the self-etching primer, thus preventing its infiltration in the top dentin surface. The same effect was not observed with SB presumably, because the highly acidic phosphoric acid eliminated the detrimental effects of calcium hydroxide solution. Moreover, the surface was rinsed after etching and this would facilitate even more the elimination of remaining particles on the surface.

Cavidry contains isopropyl alcohol included in the formulation. It is likely that when applied to dentin, it renders the surface dehydrated. For some adhesive systems, a wet surface is required to improve bond strength (Gwinnett[9] 1992b; Kanca[12] 1996). Although Cavidry may have dehydrated the dentin surface in SB group, it was followed by etching and rinsing that remoistened the substrate and did not significantly influence the results with SB. Significant decrease in bond strength was, however, observed for CLB. Perhaps the dehydration caused by Cavidry was detrimental to the self-etching system since remoistening of the surface was not performed with this material. Additionally, Cavidry was not rinsed off the surface, hence, it is also possible that residues of the solution may have reacted with the components of the self-etching primer and diminished its potential to infiltrate the dentin. Another possible explanation could be related to pH changes of the surface caused by this solution, which would affect bond strength in a similar way as observed with calcium hydroxide solution.

Mechanical alteration of dentin surface using air abrasion with 50m aluminum oxide particle prior to chemical conditioning resulted in significant lower mean bond strengths for both bonding systems. Although increased roughness of the surface could be theoretically interpreted as beneficial for bonding purposes, some studies have reported no improvement in bond strength after application of air abrasion to dentin (Los & Barkmeier[14] 1994; Roeder *et al*[23] 1995; Nikaido *et al*[18] 1996; Rinaudo *et al*[22] 1997; Manhart *et al*[15] 1999). Despite being different in their application technique, the two adhesive systems used in this study bond to dentin by micromechanical retention within the top of the dentin surface. Increased roughness on the surface does not contribute significantly to bonding (Los & Barkmeier[14] 1994; Roeder *et al*[23] 1995; Nikaido *et al*[18] 1996; Rinaudo *et al*[22] 1997; Manhart *et al*[15] 1999) and, as shown in this study, may even cause reduction of bond strengths. Presumably, increased roughness may have caused difficulty for proper infiltration of the resin agents within dentin and permitted void formation at the interface that would act as stress raisers and cause reduction of bond strengths. Nikaido *et al*[18] (1996) evaluated the influence of air abrasion on bonding of resins to bovine enamel and dentin using tensile bond testing and SEM. They showed an increase in surface roughness of dentin after air-abrasion with aluminum oxide particles, however, that was not followed by an increase in bond strength. Although in their study, SEM examination of the interfaces indicated close apposition of resin and dentin, an irregular hybrid layer could be detected below the bonding resin, suggesting the presence of areas with improper resin infiltration.

It is clear from our findings that the effects of cleansing agents on bond strength to dentin are dependent on the type of the adhesive system used. While chemical agents did not cause any significant effect with the total etch system (SB), the results with the self-etching system (CLB) varied according to the solution used, and the detrimental effects were more evident when non-rinsing solutions were used before bonding with this system. The relatively higher percentage of adhesive failures observed with CLB suggest that the treatments tested may have indeed compromised the bonding of this system to dentin substrate.

Although the decrease in bond strength may be system specific, it seems wise to avoid using cleansing agents that are not rinsed off the surface or those which may alter the pH of the surface to an alkaline one.

The results with air abrasion are aligned with others regarding the fact that it does not improve bond strength. The reason as to why it caused significant decrease in bond strength for both adhesive systems is not fully understood. Since no benefits are confirmed from using such solutions or air abrasion for cleansing purposes, we do not recommend the application of them before bonding to dentin. There are evidences in literature (Settembrini *et al*[25] 1997; Emilson & Berhenholtz[6] 1993) that indicates the antimicrobial activity potential of the etchants. Moreover, this would represent an additional, undesirable extra step in the clinical bonding procedures (Meiers & Shook[17] 1996; Gürgan *et al*[8] 1999; Bachmann *et al*[1] 1997, Nikaido *et al*[18] 1996).

RESUMO

O objetivo deste trabalho foi avaliar o efeito dos agentes de limpeza cavitária e jato abrasivo na resistência de união à microtração de dois sistemas adesivos em dentina. Superfícies planas de dentina obtidas de terceiros molares extraídos foram tratadas com agentes de limpeza cavitária ou jato de ar abrasivo antes da hibridização com os sistemas adesivos Single Bond ou Clearfil Liner Bond 2V, de acordo com as recomendações do fabricante. Estruturas de resina composta foram incrementalmente realizadas com a resina composta Z100 e os dentes foram armazenados por 24 horas em água a 37°C. Os espécimes foram posteriormente seccionados verticalmente para obtenção de vários "palitos" de união entre resina composta e estrutura dental com dimensões de área de 1,0 mm^2 e cada palito foi submetido a teste de microtração a 0.6 mm/min. Análise de variância a dois critérios mostrou que existe influência significante em ambos os fatores na resistência de união ($p<0.05$). Para o grupo do Sing-Bond os agentes de limpeza cavitária não afetam a resistência de união, quando comparados aos valores do grupo controle ($p>0.05$). Entretanto o jato abrasivo reduziu significantemente a resistência de união. Para Clearfil Liner Bond 2V, tanto a aplicaçao do Tergensol ou Cav-Clean não influenciaram nos valores de resistência de união, entretanto, os outros tratamentos causaram redução significativa nos valores de resistência de união ($p<0.05$). Estes resultados sugerem que a aplicação de agente de limpeza cavitária e jatos abrasivos antes da hibridização podem prejudicar a adesão à dentina. Significância Clínica: a aplicação de agentes de limpeza cavitária e jatos abrasivos antes da hibridização podem causar efeitos adversos na adesão à dentina. Este efeito depende do tratamento e do tipo de sistema adesivo utilizado.

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Dentina, Adesão, Resistência de união.

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