

## Effect of adhesive system and application strategy on reduction of dentin permeability

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**Abstract:** This study evaluated the effect of adhesive systems and application strategies on dentin hydraulic conductance (HC). The buccal enamel was removed from bovine incisors to simulate laminate cavity preparations. After removing the roots and the coronal pulp, the buccal dentin was treated with EDTA solution (0.5 M) for 5 minutes, rinsed, ultrasonicated for 12 minutes and connected to a permeability device. HC of the specimens was measured at 10 psi (n = 5). Permeability was measured before and after bonding procedures using G-Bond (GB), Clearfil Tri-S Bond (CTS), Hybrid Coat (HY), Bond Force (BF), Adper Easy Bond (AEB) Silorane (SI), Clearfil SE Bond (CSE) and Adper Scotchbond Multi-Purpose (SMP) adhesives systems, which were applied following three strategies: 1) according to the manufacturers' instructions; 2) two coats of all-in-one self-etching adhesives (GB, CTS, HY, BF, AEB) or priming step plus two coats of bonding resin for the other systems (SI, CSE and SMP); and 3) a thin layer of a flowable composite applied over one coat of all-in-one self-etching adhesives or primed surface for SI, CSE and SMP adhesives. No significant difference was observed among the application modes concerning their ability to reduce HC. None of the adhesives showed complete sealing (100%) of the bovine tooth dentin. SI exhibited lower HC than SMP, however, they were not significantly different from the other systems. The results suggest that all systems tested result in an HC reduction of more than 90%. The wet bonding technique seemed to be more sensitive for dentin sealing.

**Descriptors:** Dentin-Bonding Agents; Dental Bonding; Dentin Permeability.

**Declaration of Interests:** The authors certify that they have no commercial or associative interest that represents a conflict of interest in connection with the manuscript.

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### Introduction

The hybridization of dental tissues promoted by adhesive systems was first described by Nakabayashi *et al.* in 1982.<sup>1</sup> The authors etched the dentin with a 10-3 solution (10% citric acid + 3% ferric chloride) and used hydrophobic and hydrophilic groups (4-META/MMA-TBB) to infiltrate monomers into dentinal tubules and intertubular dentin. The resin-dentin interdiffusion zone or hybrid layer is the main bonding mechanism of contemporary bonding agents.

The development of dentin bonding agents has produced adhesives that are classified according to the etching type and number of steps required.<sup>2-4</sup> The hybridized layer produced by one-step self-etching or all-

Submitted: Dec 11, 2011  
Accepted for publication: May 21, 2012  
Last revision: Jun 06, 2012

in-one adhesives has been considered a permeable membrane because they did not seal the bonded interface adequately.<sup>5-7</sup> Previous one-step self-etching adhesives had low pH (less than 1) due to the composition which was based on acidic primers with a high concentration of hydrophilic monomers. This category of adhesives seems to produce a low degree of conversion, forming regions of incomplete polymerization, which are capable of attracting water and increasing permeability through the bonded interfaces.<sup>8-10</sup>

The one-step self-etching adhesive systems have since been modified,<sup>11-15</sup> however, it is not known if these changes in composition improve the properties of the seal formed by this category of adhesives. The lack of proper marginal sealing leads to water/saliva infiltration, which can contaminate the mineralized dental tissues involved in the cavity preparation.<sup>16,17</sup> To improve the sealing ability of one-step self-etching adhesives, some authors have suggested the application of an additional layer of hydrophobic material in an attempt to reduce the concentration of hydrophilic monomers to be light cured.<sup>9,18-22</sup>

The importance of studying dentin permeability in relation to the sealing ability of adhesive systems is that, besides bonding the restorative material to the tooth, adhesive systems are responsible for sealing the resin-dentin interface against microleakage and avoiding restorative material debonding, which could compromise the restorations.<sup>2,16,17</sup> The objective of this study was to measure fluid flow through the adhesive-dentin interfaces created by eight bonding agents using a permeability device. The null hypothesis was that the mode of application of the adhesives has no effect on dentin permeability.

## Methodology

One hundred and twenty bovine incisors of similar size, stored in a saturated thymol (Labsynth, Diadema, Brazil) solution at 5°C for at least one month, were used in this study. Crown segments were obtained by removing the roots 1 mm below the cementum-enamel junction, using a water-cooled diamond saw (Isomet, Buehler Ltd., Lake Bluff, USA). The pulp tissue was removed with #80 Hedstroem files (Maillefer/Dentsply, Petrópolis,

Brazil). Teeth were divided into 24 groups (n = 5).

The buccal enamel was removed during preparation of the laminate-like cavity, to expose the superficial dentin. A #4138 diamond bur (KG Sorensen Ind. Com. Ltda., Barueri, Brazil) was used to prepare the teeth, and the bur was changed after every 5 teeth. The dentinal depth was controlled using a pincer-type caliper (Golgran, São Paulo, Brazil), which measured the remaining dentin thickness that was between 1.8 and 2.9 mm. The incisor surfaces were reduced until the dentin tissue appeared and the cavity preparation was surrounded by at least 2 mm of enamel thickness, such as in the cervical area.

Each prepared tooth was attached to a polyester platform (30 × 30 × 5 mm), perforated in the center by an 18 gauge stainless steel tube fixed using cyanoacrylate glue (Super Bond, Loctite/Henkel, Diadema, Brazil). The tooth attached to the polyester platform was connected to a hydraulic pressure device (Figure 1) that delivered 10 psi (703 cm H<sub>2</sub>O or 51.7 cm Hg) of water pressure during the measurement of dentin permeability.<sup>23</sup> The hydraulic pressure device used in this study was similar to that designed by Sauro *et al.*<sup>6</sup>

Before applying the bonding agents, the teeth were treated with 0.5 M EDTA (pH 7.4) for 5 minutes. Afterwards, they were rinsed with water for 1 minute, ultrasonicated (Maxi Clean 750, Unique, Indaiatuba, Brazil) in water for 12 minutes and each tooth was submitted to measurement of the maximum fluid filtration (MFF), which was assigned as 100% permeability. After measuring the MFF (μL/min), a new smear layer was created on the dentin surface using a fine-grit-diamond bur (3139 fine-grit, KG Sorensen Ind. Com. Ltda., Barueri, Brazil) for 30 seconds under water cooling. The adhesives were applied to the dentin while connected to the permeability device, but in the absence of pulpal pressure<sup>6</sup> in an attempt to simulate the clinical conditions in which anesthetic salts cause inhibition of pulp blood flow.<sup>24</sup>

Eight commercially available dentin adhesive systems were tested:

- Adper Scotchbond Multi-Purpose (3M ESPE, St. Paul, USA),

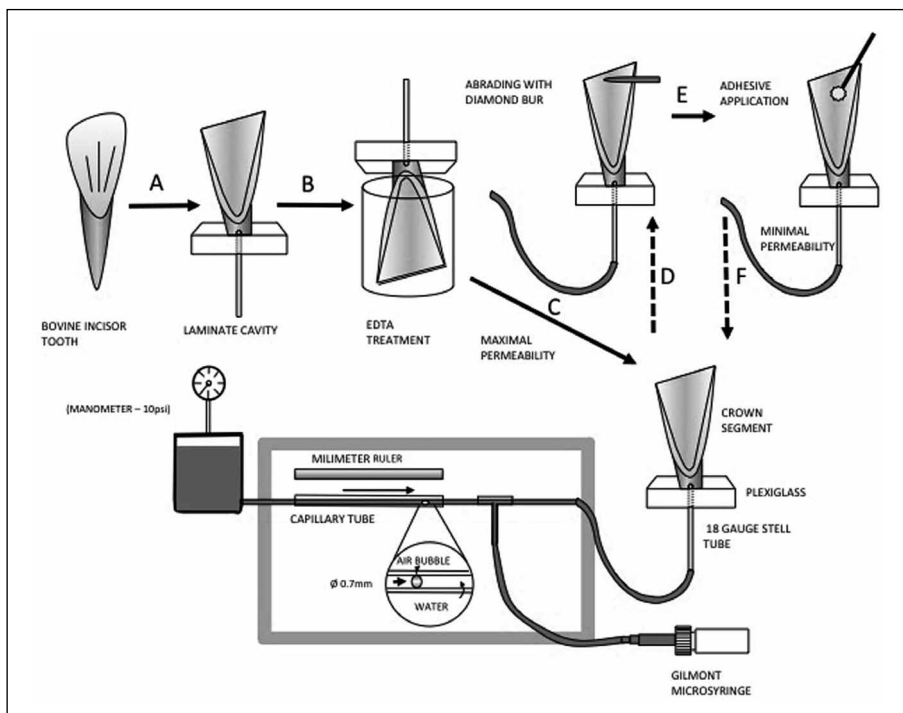
- Clearfil SE Bond (Kuraray Medical Inc, Kurashiki, Japan),
- Silorane (3M ESPE AG, Seefeld, Germany),
- Adper Easy Bond (3M ESPE, St. Paul, USA),
- Bond Force (Tokuyama Dental Corp., Taitou-ku, Tokyo, Japan),

- Clearfil Tri-S Bond (Kuraray Medical Inc.),
- G-Bond (G-Cem, GC Corp., Tokyo, Japan) and
- Hybrid Coat (Sun Medical, Moriyama, Japan).

The adhesives were applied following three different application methods (Table 1):

**Table 1** - Characteristics of adhesive systems and application modes.

Adhesive system	Characteristics	Application mode 1	Application mode 2	Application mode 3
Adper Scotchbond Multi-Purpose	3-step etch-and-rinse	according to manufacturer's instructions	two coats of adhesive resin	flowable composite layer over primed dentin
Clearfil SE Bond	2-step self-etching primer	according to manufacturer's instructions	two coats of adhesive resin	flowable composite layer over primed dentin
Silorane	2-step self-etching primer	according to manufacturer's instructions	two coats of adhesive resin	flowable composite layer over primed dentin
Adper Easy Bond	one-step self-etching adhesive	according to manufacturer's instructions	two coats of adhesive	one coat of adhesive + flowable composite layer
Bond Force	one-step self-etching adhesive	according to manufacturer's instructions	two coats of adhesive	one coat of adhesive + flowable composite layer
Clearfil Tri-S Bond	one-step self-etching adhesive	according to manufacturer's instructions	two coats of adhesive	one coat of adhesive + flowable composite layer
G-Bond	one-step self-etching adhesive	according to manufacturer's instructions	two coats of adhesive	one coat of adhesive + flowable composite layer
Hybrid Coat	one-step self-etching adhesive	according to manufacturer's instructions	two coats of adhesive	one coat of adhesive + flowable composite layer



**Figure 1** - Schematic representation of permeability measurement methodology.

1. According to the manufacturers' instructions;
2. Two coats for the one-step self-etching adhesives. For the other adhesives the priming step (Primer, Clearfil Primer, or LS System Adhesive Primer, respectively) was followed by two coats of adhesive resin (Adhesive, Clearfil Bond, or LS System Adhesive Bond, respectively). Each coat was light-cured (650 mW/cm<sup>2</sup>, Demetron LC, Kerr Corp., Orange, USA) separately for the time recommended by the manufacturer.
3. One coat for the one-step self-etching adhesives plus a thin layer of a flowable composite (Filtek-Flow, 3M ESPE, St. Paul, USA). For the Scotch-Bond Multi-Purpose, Clearfil SE Bond and Silorane adhesives, the priming step was followed by the application of a thin layer of flowable composite (Filtek-Flow) instead of the adhesive resin. The adhesive coats and the low-viscosity resin were light-cured with the same curing unit.

After the bonding procedures, the fluid flow through the hybridized dentin (FFHD - final measurement) was remeasured to calculate the percentage reduction of fluid flow through the adhesive-dentin interface or dentin seal provided by the adhesives. This represented the permeability after application of the adhesives relative to its maximum EDTA-treated pre-bonded value, or maximum fluid filtration (MFF - initial measurement), with each

tooth serving as its own control. The equation below was used to calculate the percentage dentin sealing provided by the application of adhesive:<sup>25</sup>

$$\text{Dentin sealing (\%)} = 100 \times (\text{MFF} - \text{FFHD}) / \text{MFF}$$

The data were analyzed by two-way ANOVA (adhesive type and application mode were the factors under study) and Tukey test. Statistical significance was set at  $\alpha = 0.05$ .

## Results

Table 2 shows the mean percentage of dentin sealing provided by the different bonding procedures and the standard deviations of the experimental groups. Two-way ANOVA revealed a significant difference only for the factor "adhesive type" ( $P = 0.0495$ ). Neither the "application mode" factor nor the interaction between the factors were significant ( $P = 0.1006$  and  $P = 0.6321$ , respectively).

All three application modes resulted in a similar percentage of dentin sealing for the same adhesive used ( $P > 0.05$ ). Only one significant difference was observed between the adhesives ( $P < 0.05$ ): the Silorane two-step self-etching adhesive ( $96.6 \pm 2.6\%$ ) resulted in a higher percentage of dentin sealing than the Scotchbond Multi-Purpose three-step etch-and-rinse adhesive ( $92.4 \pm 4.4\%$ ). Both adhesives resulted in a similar percentage of dentin sealing to

**Table 2** - Percentage of dentin sealing provided by bonding procedures (means  $\pm$  SD).

Adhesive system	Application mode 1	Application mode 2	Application mode 3	Means of 3 application modes	Tukey test
Adper Scotchbond Multi-Purpose	88.7 $\pm$ 5.1	93.9 $\pm$ 5.5	94.7 $\pm$ 2.7	92.4 $\pm$ 4.4	a
Clearfil SE Bond	95.1 $\pm$ 3.5	96.6 $\pm$ 2.2	95.4 $\pm$ 2.9	95.7 $\pm$ 2.9	ab
Silorane	96.4 $\pm$ 2.4	97.6 $\pm$ 1.8	95.8 $\pm$ 3.6	96.6 $\pm$ 2.6	b
Adper Easy Bond	94.6 $\pm$ 3.1	95.6 $\pm$ 2.6	96.9 $\pm$ 1.2	95.7 $\pm$ 2.3	ab
Bond Force	94.9 $\pm$ 3.1	93.5 $\pm$ 3.8	94.7 $\pm$ 4.7	94.4 $\pm$ 2.3	ab
Clearfil Tri-S Bond	94.5 $\pm$ 3.8	94.6 $\pm$ 3.5	96.3 $\pm$ 2.6	95.1 $\pm$ 3.3	ab
G-Bond	94.9 $\pm$ 1.9	94.5 $\pm$ 4.2	96.4 $\pm$ 2.4	95.3 $\pm$ 2.8	ab
Hybrid Coat	93.9 $\pm$ 3.1	95.9 $\pm$ 2.5	94.9 $\pm$ 2.6	94.9 $\pm$ 2.7	ab
ANOVA	A	A	A		

Means of the 3 application modes followed by different lower case letters (column - comparison among adhesives) are statistically significant by Tukey test ( $P < 0.05$ ). Groups having the same upper case letters (row - comparison among application modes) are not statistically significant by ANOVA ( $P < 0.05$ ).

other adhesives, and the difference between them was significant ( $P < 0.05$ ).

## Discussion

The dentin permeability measurements after EDTA-etching (MFF - maximum fluid filtration) and after bonding (FFHD - fluid flow through hybridized dentin) showed that no adhesive system completely sealed the dentin surface. However all adhesives sealed the dentin more than 90%. The maximum mean value was  $96.6 \pm 2.6\%$  for the Silorane two-step self-etching adhesive. This was significantly higher than that observed for the Scotchbond Multi-Purpose three-step etch-and-rinse adhesive ( $92.4 \pm 4.4\%$ ). The application of flowable resin or an additional layer of adhesive resin neither improved nor impaired the dentin sealing. Thus, the null hypothesis was accepted since the application mode of the adhesives had no effect on percentage dentin sealing.

Because of the highly hydrophilic nature of some bonding agents, such as one-step self-etching systems, they can act as permeable membranes, attracting water and degrading faster than more hydrophobic adhesives.<sup>5,7,8,10,16</sup> To overcome this early degradation, some investigations have revealed that the application of more hydrophobic resins over hybridized tissues provides better sealing of acid-etched dentin<sup>9,18-21</sup> and higher bond strength of composite to dentin.<sup>19,20,22</sup> In this study, the use of flowable resin or an additional layer of more hydrophobic adhesive did not improve the percentage dentin sealing of any type of adhesive.

As the results of this study indicated that the percentage dentin sealing was independent of the use of an additional layer of more hydrophobic resin, the correct application of adhesives was essential to reduce dentin permeability. This study also showed that only one adhesive layer of the one-step self-etching systems resulted in similar dentin hydraulic conductance when compared to use of an additional layer of the same adhesive or an additional layer of flowable or adhesive resin. Therefore, the quality of the adhesives is important since they are directly in contact with the dentin tissue and are responsible for sealing it against fluids that tend to pass through

hybridized dentin and bond to the restorations.<sup>6,16</sup> The seal provided by the bonding agents was measured only immediately after adhesive application. The durability of hybridization ensures that dentin sealing is maintained; however, the one-step self-etching adhesives may lose their effectiveness, increasing the hydraulic conductance faster than other types of adhesive systems.<sup>5-7</sup> The bonding durability depends on the monomeric composition of each adhesive and the loss of marginal sealing can compromise a restoration.<sup>15,16</sup> Further studies will investigate the durability of bonding and its relationship with the sealing of dentinal tubules.

Two two-step self-etching primers were evaluated: Clearfil SE Bond and Silorane. The acidic primers used hybridized smear plugs instead of removing them, sealing the dentinal tubules.<sup>3,26</sup> The Clearfil SE Bond bonding resin is mixed with primer over the dentin surface and then both components are polymerized together *in situ*. For Silorane adhesive, primer and bond were light-cured separately; however, no difference was observed between these two-step self-etching primers. Changing the bonding resin to flowable resin or application of an additional layer of bonding resin did not alter the performance of either adhesive.

Two one-step self-etching adhesives are HEMA-free: G-Bond and Hybrid Coat. The HEMA monomer is commonly added to bonding agents and improves the stability of adhesive solutions that contain hydrophobic and hydrophilic components. However, HEMA has been considered vulnerable to hydrolysis because it is highly hydrophilic and absorbs water.<sup>27</sup> While the Hybrid Coat adhesive contains 4-META, polyfunctional acrylate and monomethacrylates as functional monomers, G-Bond contains 4-MET and phosphoric ester-monomer as the acidic monomer. 4-META is a crystalline powder, which in contact with water can react and form 4-MET. Two carboxylic groups attached to the aromatic group of the 4-MET molecule provide the acidic characteristic that is important for the demineralization of the dentin and formation of the hybrid layer.<sup>28</sup>

Adper Easy Bond, Bond Force and Clearfil Tri-S Bond one-step self-etching adhesives contain

HEMA, Bis-GMA and a specific acidic monomer, methacrylated phosphoric ester, methacryloyloxyalkyl acid phosphate and 10-MDP, respectively.<sup>15</sup> The presence of HEMA did not influence the results of this study; however, the monomeric composition of these adhesives could be a determinant that requires evaluating over time with the specimens stored in water.

The sensitivity of the bonding technique to acid-etched dentin has been considered more critical than self-etching adhesives.<sup>3,6,16</sup> ScotchBond Multi-Purpose uses the total-etch and moist bonding techniques and the water is provided from rinsing and dentin tubules. Adequate moisture control before bonding of etch and rinse adhesive is difficult, which can lead to compromising of the dentin hybridization. The lower numeric reduction on the hydraulic conductance or the percentage dentin sealing for this adhesive can be related to the moisture control

technique, which can produce more permeable hybrid and adhesive layers due to the presence of water during polymerization. Even the application of two layers of bonding resin did not improve the acid-etched dentin sealing.

## Conclusions

No significant difference was observed among the application modes concerning their ability to reduce hydraulic conductance. None of the adhesives showed complete sealing (100%) of the bovine tooth dentin. The Silorane adhesive showed lower hydraulic conductance than SMP; however, they were not different from the other systems.

## Acknowledgments

This study was supported by grants from CAPES and CNPq (303587/2007-5), Brazil.

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