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Research Article

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Performance of a Universal Adhesive on Etched and Non-Etched Surfaces: Changes in Dentin Permeability

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Introduction

Hybridization process for dentin is well known, but remains a difficult challenge as it is dependent on many factors related to dentin substrate, adhesive nature including its solvent. The development of universal adhesives followed the all-in-one concept of existing one-step self-etch adhesives, thus requiring water in their formulation to ionize hydrophilic acid monomers. Most often, carboxylate and/or phosphate groups are their primary functional monomers [1]. Jee, et al. published in 2016 that ethanol molecules penetrate via increasing solvent accessible surfaces induced by the rearrangement of collagen monomers in the gap region [2]. The aim of this study is to evaluate the discrepancy in dentin permeability after application of a universal adhesive used in self-etching (SE) mode or etch and rinse (E&R) mode, by means of hydraulic conductance measurement.

Material and Methods

Hydraulic Conductance

Hydraulic conductance is the fluid volume passing through a given surface per unit time under a constant pressure gradient. A fluid pressure applied on a given surface gives a certain value of dentinal flow. It is measured in this study using a Flodec apparatus (De Marco Engineering, Geneva, Switzerland) connected to a computer. A 0.2 bar pressure is applied inside a pressure container that transmits it through a small tubing connected to a sample holder where is placed a thin slice of dentine glued to a perforated polycarbonate disc. This simulates pulpal pressure on a 0.28 cm2 dentin surface, and the liquid flux is measured by a calibrated capillary tubing by following a small air bubble injected at the start of the experiment. The apparatus measures the displacement of the bubble every 30 sec. and registers the data continuously. This is then transformed in hydraulic conductance using the equation: Volume (µl) = distance (mm) × 1000 (µl) / 310 (mm).

Thus, a distance of 1 mm covered by the bubble in the capillary corresponds to a volume of 3.2 µl passing through the dentin disc. The linear movement of the bubble is converted into hydraulic conductance by the formula : $Lp= Jv / (A \bullet \Delta P \bullet t)$ [where Lp is hydraulic conductance in $\mu l \bullet cm^{-2} \bullet min^{-1} \bullet cm\ H_2O^{-1}$, Jv is fluid flow in µl, A is dentin surface area in cm2, ΔP is hydraulic pressure gradient in cm H2O and t is time in minutes].

Specimen Preparation

Twenty non-carious human molars extracted for orthodontic reasons were used in this study. From each tooth, a slice was cut off in the middle of the coronal part parallel to the occlusal surface and above pulpal chamber level, using a slow speed saw (Isomet, Buehler Ltd, Evanston, USA). These dentin slices represent deep dentin, region of dentin where the number and diameter of tubuli are more important than in superficial dentin. Similar zones were sampled to minimize variations in dentinal structure. The 20 specimens prepared as indicated above were randomly divided into two groups of 10 teeth: Group 1 received adhesive in E&R mode, while group 2 received it in SE mode. The technique compares the hydraulic conductance before and after the introduction of the adhesive system. The application of the adhesive system will close off a portion of the dentinal tubuli, resulting in a decrease of the initial permeability of dentin, which will be more or less dependent on the level of sealing due to the bonding adhesive: the higher the permeability, the lower is the sealing efficiency. The components of the adhesive tested are listed in Table 1.

Table 1 : Chemical composition of tested adhesive

Adhesive	Manufacturer	Composition	Batch
OptiBond Universal	Kerr Corporation Orange,CA USA	2-hydroxyethyl methacrylate (HEMA) Glycerol dimethacrylate Glycerol phosphate dimethacrylates (GPDM) Acetone Ethanol Water	LK02187

Measurement of Hydraulic Permeability

The initial hydraulic flow though dentin slices was measured using physiological saline. An experimental smear layer was made on the occlusal surface by means of 50 consecutive abrasions on each specimen, by manually moving fine grain sand paper (P4000, Buehler) horizontally under water irrigation without vertical force. For the two groups the hydraulic conductance was measured at time T0 after this abrasion. For Group 1 (system using separate etching and rinsing steps) the hydraulic conductance was measured again at time T1. For Group 1 and Group 2, the dentin slides received the adhesive according to the manufacturer's instructions. Hydraulic conductance was measured for all specimens at time T2, right after application. Statistical analysis comprised an analysis of the mean and standard deviation for each test group, and an analysis of variance considering the T2 measurements as repeated measurements on a given specimen, and a multiple comparison test (Duncan's test) where the significance level was fixed at 0.05.

Results

The reduction in fluid flow through the dentin slices is reported for the etch-and-rinse and self-etch protocols in Table 2. There is an important reduction in dentin permeability following adhesive application in both cases: 59.0 % for etch & rinse, and 56.6 % for self-etch. However, these differences are not statistically significant (o=0.719, greater than 0.05).

Table 2 : Means and standard deviations of penetration/permeability of test specimens by surface treatment and bonding agent.

Bonding protocols	T0	T2 post-adhesive
Etch-and-rinse	−37.1% ±7.8	−59.0% ±12.0 a
Self-etch	−32.7% ±11.8	−56.6% ±10.4 a

Values expressed in percentage decrease (−) or increase (+) with respect to baseline value (n = 10). Mean ± standard deviation, Duncan's test (the same letter "a" indicates the same group.

Discussion

Universal adhesives, like all adhesive systems, are a complex blend of different functional monomers and other major components like solvents [3]. The adhesive studied here is classified as ultra-mild (pH between 2.0 and 3.0) and contains a monomer with phosphoric acid functional group, glycerophosphate dimethacrylate (GPDM) [4,5]. Being structurally similar to 10-MDP, GPDM is known to have a large diffusing potential due to its acidic groups and its hydrophilicity, and this favours adhesion to dentine [6]. Yoshihara, et al. showed that while GPDM is capable to bond chemically to hydroxyapatite, the bond is relatively weak in stability, therefore GPDM may be more efficient in E&R mode than in SE mode [7].

The adhesive tested contains HEMA, a hydrophilic molecule that can infiltrate partially hydrated demineralized dentin. It is soluble in water and acetone which improves the miscibility and solubility of components, whether polar or non-polar. In presence of water, it can be hydrolysed to ethyleneglycol and methacrylic acid [8]. It stabilizes the collagen network and increases dentinal permeability and diffusion of monomers when applied in aqueous solution [9]. Yoshihara, et al. considers that GPDM may act similarly as HEMA and holds a high potential to penetrate deeply into exposed collagen [7]. GPDM can more efficiently polymerize with other less hydrophilic co-monomers thanks to its two methacrylate groups. These arguments may explain the good performance of OptiBond Universal during the permeability tests in this study.

The adhesive tested contains a blend of three solvents: acetone, ethanol and water. Solvents are responsible for water displacement from collagen network and infiltration of resin monomers into spaces previously occupied by water [10]. Water is an essential component to keep monomers dissolved and to ionize the acidic groups upon contact with hydroxyapatite. Water has high potential to bind via hydrogen bonds [11]. However free unbound water should be removed during penetration and diffusion of resin components inside demineralized dentin in order to create an optimal and stable hybrid layer. Ethanol easily penetrates by capillarity action demineralized collagen and convey accompanying monomers during the process. It dehydrates free unbound water while leaving enough bound water to keep the collagen fibrils expanded by hydration [12]. It also has capacity to create hydrogen bonds in the absence of water inside the demineralized tissus [11]. According to Brackett, et al. the key to creation of ideal resin bonds is to remove all unbound water with water-free but water-miscible solvents like ethanol [13]. Acetone as solvent is another important component that promotes efficient dentin hybridization. Many studies have shown that acetone thanks to its favorable physical properties (viscosity and surface tension, vapor pressure) of the adsorbed constituents increase wettability and hydrophilicity of the adsorbed collagen surface while chasing water from the inner layers of the collagen network. [14-16]. Acetone has been shown to act as vector for diffusion of soluble moieties inside demineralized dentin thereby increasing the mechanical properties of the hybrid layer [17,18]. The association of ethanol and HEMA helps to consolidate the demineralised and dehydrated dentin collagen network by maintaining interfibrillar spaces wide, thereby enhancing better penetration of more hydrophilic monomers [19]. The 3 solvents of this adhesive seem to act in a synchronized manner in order to enable intimate contact between resin monomers and collagen fibrils in an environment where unbound water content is at its lowest. Active application by rubbing the adhesive on the suface of dentin increases the potential for immediate binding and also leads to long term stability [4].

Optibond Universal used in E&R mode gives slightly better results than when used in SE mode (-59.0% reduction in permeability when acid-etched compared to 56.6% when self-etched), even though the difference is not statistically significant. Most reports do not show a significant difference when a universal adhesive is used in whichever mode [17,20-23]. The decrease in permeability values obtained with the tested adhesive is among the best values obtained compared to all the systems tested we reported previously [24-26]. This statement is true for self-etching mode and prior etching before adhesive application, where results are often less than optimum due to opening of tubuli due to the action of phosphoric acid.

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