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ORIGINAL ARTICLE

Effect of 1% chlorhexidine gel on the bonding strength to dentin

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Background/purpose: The purpose of this *in vitro* study was to evaluate the effect of 1% chlorhexidine (CHX) gel on dentin bond strengths of posterior composite resin applied with two different adhesive systems.
Material and methods: In total, 75 extracted, caries-free human molars were used. The occlusal surface of each tooth was ground to create a flat dentin surface. Then, each tooth was mounted in acrylic. The dentin specimens were randomly assigned to five groups of 15 specimens each. In Group 1, Prime & Bond NT (PBNT) was applied; in Group 2, a 1% CHX gel+etching for 15 s+PBNT were applied; in Group 3, etching+1% CHX gel+PBNT were applied; in Group 4, Clearfil S^3 Bond was applied; and in Group 5, 1% CHX gel+Clearfil S^3 Bond were applied. A dentine bonding system was applied to dentin surfaces, and composite cylinders were built up using a special device and then light-polymerized. Specimens were mounted and sheared using an Instron universal testing machine at a cross-head speed of 0.5 mm/min.
Results: The results were recorded in megapascals. The sheared specimens were examined under a light microscope, and the type of failure (adhesive, cohesive or mixed) was recorded. Data were compared by one-way analysis of variance and Tukey's honestly significant difference tests. Means were 16.4±4.1 MPa in Group 1, 16.2±3.9 MPa in Group 2, 13.0±4.5 MPa in Group 3, 11.9±2.7 MPa in Group 4, and 11.5±2.7 MPa in Group 5. The use of 1% CHX gel before acid etching was significantly higher than after etching on the shear bond strength of PBNT ($P<0.05$), but did not differ significantly from PBNT alone ($P>0.05$).
Conclusion: Within the limitations of the present *in vitro* study, it was concluded that 1% CHX gel application did not adversely affect the shear bond strengths of dentin-bonding agents.

Introduction

In response to increasing esthetic demands of patients, the use of composite resin materials for posterior tooth restorations is increasing.[1] This increase is due primarily to demands for improved esthetics. However, it is generally accepted that resin composites are not yet able to guarantee excellent results when used for posterior tooth restorations. This is due to postoperative sensitivity and penetration of

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microorganisms and/or their toxic products that, in turn, can cause pulpal lesions and secondary caries.[2]

Bacterially contaminated cavity walls associated with caries are a potential problem in restorative dentistry.[3] Bacteria can remain in the smear layer or in dentinal tubules, and can potentially multiply.[4] Studies indicate that residual bacteria might proliferate from the smear layer beneath restorations, allowing toxins to diffuse to the pulp, resulting in irritation and inflammation.[3,4] It was argued that microorganisms that are present in the cavity walls cannot be removed by the use of water spray or by the effect of restorative materials containing disinfecting agents.[5] Therefore, the adjunctive use of antibacterial solutions after cavity preparation may be considered a method to reduce the incidence of postoperative sensitivity by eliminating viable bacteria and their toxins from the restorative interface.[6] The use of a cavity disinfectant before applying a dentin adhesive agent can reduce or eliminate postoperative sensitivity in composite restorations.[3]

Researchers have applied various alternative approaches to eliminate residual bacteria left in cavity preparations. Treatments with a disinfectant wash and different antibacterial agents have been tested.[4,5] Commercially available disinfectants containing compounds such as chlorhexidine (CHX) digluconate, disodium ethylenediaminetetraacetic acid (EDTA) dihydrate, sodium hypochlorite, hydrogen peroxide, and iodine are used to remove bacterial contaminants.[7,8] Of these, CHX is commonly used to remove bacterial contaminants. CHX is an effective agent for disinfecting dentin. Silva et al.[9] reported a significant decrease in the number of bacteria in dentinal tubules after application of 0.2% CHX for 5 minutes. It is also effective in reducing the levels of *Streptococcus mutans* found on exposed carious root surfaces.[10]

CHX has a strong suppressive effect on *S. mutans* and *S. sobrinus*.[3,11] These microorganisms are of major importance in the development of initial caries.[4] The use of a CHX cavity cleanser after tooth preparation can reduce residual caries and postoperative sensitivity.[3,12,13]

CHX is widely used as an antimicrobial agent and for disinfection before placement of restorations.[7] A CHX solution is active against a wide range of microorganisms, because it is bacteriostatic at low concentrations and bactericidal at higher concentrations.[13] CHX may also inhibit bacterial adherence to surfaces and to each other by competing with calcium for retention sites and, thus, may prevent the formation of calcium bridges between bacteria and oral surfaces or between bacteria.[12] Therefore, the use of solutions such as CHX, which have an antibacterial or bactericidal effect, provides an adjunct treatment that contributes to the suppression of residual infection, thereby increasing survival of restored teeth.[11] Thus, this study evaluated the effects of CHX gel (before and after acid etching) on the shear bond strength of composites with two bonding systems.

Materials and methods

Seventy-five extracted, intact human molars were chosen for this study. All teeth were hand-scaled; all soft tissue was removed, and the teeth were stored in room-temperature tap water for 1 week prior to bonding. Teeth were sectioned with a low-speed diamond disk saw (IsoMet; Buehler, Lake Bluff, IL, USA) under water coolant to expose the mid-coronal dentin. Sections of the teeth, including the roots, were mounted inside a cylindrical-shaped plastic material, 2.5 cm in diameter and with a height of 5 cm, using autopolymerizing acrylic resin. Dentin surfaces were flattened using 600-, 800-, and 1200-grit waterproof polishing papers to create a standardized dentin surface, and the teeth were randomly divided into five groups of 15 teeth each.

In Group 1, the dentin surface was etched with 34% phosphoric acid gel (Dentsply Caulk, Milford, DE, USA) for 15 seconds, rinsed with water for 20 seconds, and dried with absorbent paper. Then, Prime & Bond NT (PBNT) (Dentsply Caulk, Milford, DE, USA) was applied and left undisturbed for 30 seconds, lightly air-dried for 2 seconds, and light-cured for 20 seconds with a light-emitting diode (LED) (Elipar FreeLight; 3M ESPE AG, Seefeld, Germany). After application of an adhesive, specimens were clamped in an Ultradent bonding jig (Ultradent Products, Inc., South Jordan, UT, USA). A posterior composite (Quixfil; Dentsply DeTrey, Konstanz, Germany) was carefully inserted into the surface by packing the material into cylindrical-shaped plastic matrices with an internal diameter of 2.34 mm and a height of 3 mm. Excess composite was carefully removed from the periphery of the matrix with an explorer. The composite was cured with an LED for 20 seconds.

In Group 2, CHX gel (at 1%; Drogsan Pharmaceuticals, Ankara, Turkey) was applied using a disposable brush tip and left undisturbed for 20 seconds. Next, the dentin surface was etched with 37% phosphoric acid for 15 seconds, rinsed with water for 20 seconds, and dried with absorbent paper. The bonding procedure was the same as that in Group 1.

In Group 3, the dentin surface was etched with 37% phosphoric acid for 15 seconds, rinsed with water for 20 seconds, and dried with absorbent paper. Next, 1% CHX gel was applied using a disposable brush tip, and left undisturbed for 20 seconds. The

application of PBNT and the posterior composite resin was the same as in Group 1.

In Group 4, Clearfil S^3 Bond (Kuraray Medical, Okayama, Japan) was applied and left in place for 20 seconds, dried by blowing high-pressure air over it for 5 seconds, and light-cured for 10 seconds with an LED. After applying the adhesive, specimens were clamped in the Ultradent bonding jig. Posterior composite resin (Clearfil Majesty Posterior Shade A3; Kuraray Medical) was carefully inserted and cured with an LED for 20 s.

In Group 5, 1% CHX gel was applied using a disposable brush tip, and left undisturbed for 20 seconds. Then, Clearfil S^3 Bond was applied and left in place for 20 seconds, dried by blowing high-pressure air over it for 5 seconds, and light-cured for 10 seconds with an LED. Clearfil Majesty Posterior was applied in the same way as in Group 4.

Specimens were stored in distilled water at 37ºC for 24 hours. They were then mounted with the treated surfaces parallel to the shearing rod of an Instron universal testing machine (Instron Corp., Canton, MA, USA) and sheared to failure at a crosshead speed of 0.5 mm/min. The results were recorded in megapascals (MPa). The testing was carried out at room temperature (23ºC) and a relative humidity of 50%.

A one-way analysis of variance was used to detect any significant differences ($P \leq 0.05$) in bond strengths among the groups. *Post hoc* comparisons were made using Tukey's honestly significant difference test. After the test procedure, fractured surfaces were observed with a dissecting microscope (SZ-TP; Olympus, Tokyo, Japan) at a magnification of 20 to determine the failure modes, classified as adhesive failure, cohesive failure within the composite, and cohesive failure within the tooth.

One specimen from each group was randomly selected and sputter-coated with gold after fracture and prepared for scanning electron microscopy (SEM). Coated specimens were then observed under an SEM (JSM-5600; JEOL, Tokyo, Japan) at different magnifications.

Results

Mean shear bond strengths to dentin for the five groups are shown in Table 1. Mean shear bond strength values ranged 11.5–16.4 MPa. In particular, Group 1 (PBNT) showed the highest mean shear bond strength value at 16.4 MPa. No significant differences were found between Groups 1 and 2. The statistical analysis showed that the bond strengths of Group 1 (no CHX gel treatment) were significantly higher than those of Group 3 (CHX gel treatment after etching). In contrast, Group 3 (PBNT) demonstrated a significant decrease in bond strength when 1% CHX gel was applied after acid etching (the statistical differences are given in Table 1).

Groups 1 and 2 demonstrated statistically significant differences with Groups 4 and 5 ($P < 0.01$). When the 1% CHX gel was applied to the dentin surface before Clearfil S^3 Bond (Group 5), the shear bond strength was not affected in this study. No significant differences were found between Groups 4 and 5.

The examination of the debonded samples with a stereomicroscope at 20× magnification showed that the fractures were predominantly adhesive for the agents, as shown in Table 2.

SEM analysis revealed that the dentin surface was covered by a hybrid layer. In all SEM samples, composite resin remnants were found. Fig. 1 shows the dentinal surfaces after applying the shear bond strength test.

Discussion

Recent advances in resin-based adhesives and restorative materials, as well as increased patient demands for esthetic restorations, have increased the use of resin-based composites in posterior teeth. However, secondary caries were found to be the

Table 1. Mean bond strengths (in megapascals) and standard deviations (SDs) of the test groups

Group	*n*	Mean (MPa)	SD
Group 1	15	16.4*	4.1
Group 2	15	16.2*	3.9
Group 3	15	13.0†	4.5
Group 4	15	11.9†‡	2.7
Group 5	15	11.5†‡	2.7

Groups identified with different symbols significantly differ ($P < 0.05$).

Table 2. Failure modes of the test groups*

Group	Failure mode		
	Adhesive	Cohesive	Mixed
Group 1	8	5	2
Group 2	7	5	3
Group 3	7	4	3
Group 4	11	2	2
Group 5	9	5	3

*Although the results of the χ^2 analysis were highly significant ($P < 0.01$), inferences could not be made because of the small sample size and the fact that 75% of cells had an expected count of fewer than 5.

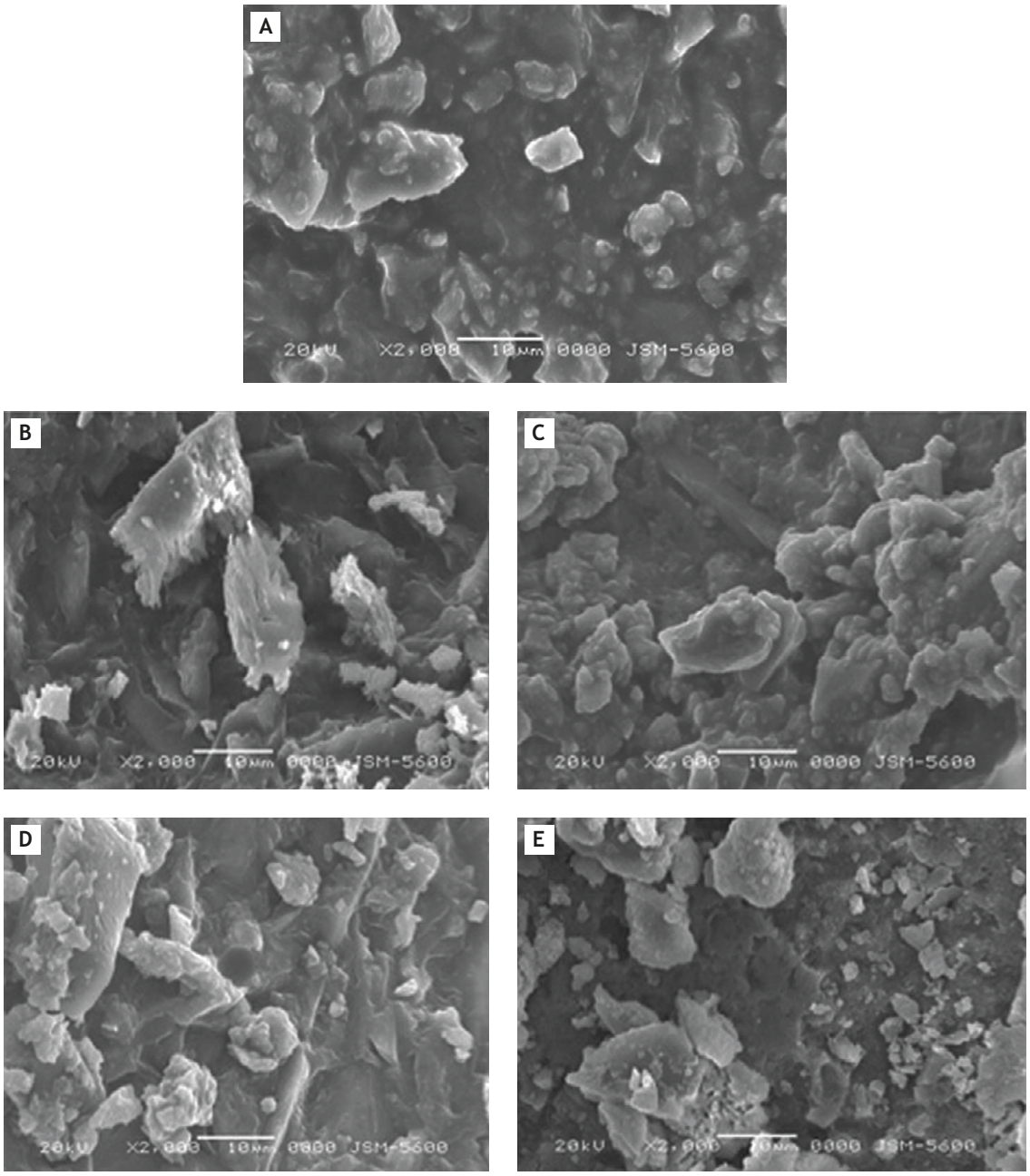

Fig. 1 Scanning electron microscopy photographs of dentin surfaces. (A) Prime & Bond NT was applied; (B) 1% chlorhexidine (CHX) gel was applied before acid etching; (C) 1% CHX gel was applied after acid etching; (D) Clearfil S^3 Bond was applied; (E) Clearfil S^3 Bond was applied after the 1% CHX gel. All images are at the same magnification.

most common reason for replacing resin composite restorations.[14] This may be a result of polymerization shrinkage, which causes a gap between the material and the tooth structure, allowing bacterial penetration.[15] Another source of secondary caries is the presence of bacteria in the smear layer after cavity preparation, which can remain viable for long periods of time.[15] A disinfectant solution, which eliminates these residual bacteria, could be useful after cavity finishing.

One study reported the efficacy of disinfectant solutions.[3] Meiers and Kresin[3] showed that use of CHX products as a cavity cleaner after tooth preparation could reduce the potential for residual caries and postoperative sensitivity. The application of CHX did not negatively affect shear bonding.[16]

The present *in vitro* study showed that CHX gel did not affect the shear bond strength of PBNT before etching the dentin. This result is consistent with recent studies which found that CHX application before and after acid etching did not significantly affect the dentin bonding system.[8,17] In addition, similar studies demonstrated that CHX application prior to acid etching had no adverse

effects on immediate composite adhesive bonds in dentin.[17,18] However, CHX gel adversely affects the shear bond strength of PBNT after etching the dentin. These results correspond to those of Vieira Rde and da Silva,[19] who showed that a cavity disinfectant containing 2% CHX had an adverse effect and produced significantly lower shear bond strengths. In contrast, Gürgan, et al.[20] indicated that application of the CHX before and after acid etching significantly decreased the shear bond strength to dentin. In contrast, those results are inconsistent with the results of Meiers and Shook[4] who found that CHX had no influence on the shear bond strength to dentin.

In vivo and *in vitro* studies by Carrilho et al.[7] and Hebling et al.[21] showed that CHX preserved the hybrid layers with CHX treatment after acid etching.[7,21] The hybrid layer might have decreased the shear bond strength to dentin. The bonding mechanism of the material to the dental structure is another relevant factor. When the smear layer is removed before the restorative material is put in place, the surface's wetting ability is enhanced, leading to the formation of a material tag. Phosphoric acid removes the smear layer, exposes collagen, and reveals the open tubules. The absence of a smear layer after acid treatment produces relatively smooth intertubular dentin without peritubular dentin. Open tubules facilitate the formation of a hybrid or resin-infiltrated layer, creating large surface areas for bonding and allowing the development of resin tags.[22]

Breschi et al.[23] found that the use of CHX as a primer on acid-etched dentin could prevent collagen degradation even after 12 months at a very low concentration (0.2%). In addition, the concentration of CHX did not affect the bond strength. CHX was shown to inhibit the activity of matrix metalloproteinases-2, -8 and -9 through a chelating mechanism, and it also had antibacterial activity.[24] However, it may show a higher antibacterial effect when applied at higher concentrations.[25] In the present study, we applied a 1% gel form of CHX. This form contains a higher concentration of CHX, and it may show longer-lasting adhesion to dentin surfaces than the liquid form of CHX. Further research is needed on adhesion of different CHX forms, between gel and liquid, to dentin surfaces when used to prevent degradation and cavity disinfection.

In the present study, we found that self-etching bonding systems showed lower bond strengths than did etch-and-rinse systems. However, CHX gel application did not affect the shear bond strength. This finding is in agreement with a previous study by Ercan et al.[26] who showed that CHX solution application as a cavity disinfectant decreased the bond strength in self-etching bonding systems. However, the shear bond strength was not adversely affected by CHX solution application when the etch-and-rinse system was used. In addition, CHX gel application had no adverse effect on the shear bond strength of the composite resin. This may have been due to a limited penetration depth of the material in the dentin structure.[26]

In our study, Clearfil S^3 bond did not present any statistically significant difference after CHX gel treatment. This result is in accordance with the study by Soares et al.[17] in which application of a concentration of 0.12% and 2% CHX produced similar behaviors, with no adverse effects on the bond strength. Our results for the shear test showed that bonding with a composite on the dentin was better for teeth treated with PBNT than with Clearfil S^3 Bond. Clearfil S^3 Bond showed the lowest bonding strength on dentin. The differences were statistically significant ($P<0.05$).

Modes of use vary before etching, after etching, rinsing off, or not rinsing. Use of a CHX cleanser before etching was shown not to affect bonding to enamel or dentin.[27] Another study, however, reported reduced dentin bond strengths when a CHX cleanser was used before or after etching, but rinsing the cleanser off before bonding produced bond strengths that were similar to no-cleanser controls.[28] Rinsing away cleansers prior to bonding will most likely prevent undesired material interactions.

The simultaneous etching of enamel and dentin, or total etching techniques, and developments made in chemical adhesives have improved bond strengths.[29] Current developments are focused on simplifying the application of bonding agents by decreasing the time and steps required for placement. As a result, manufacturers have combined the primer and adhesive into a single component but still maintain separate etching and rinsing steps. This method is called two-step bonding.[30]

All dentin surfaces were coated with a hybrid layer; high ratios of composite resin remnants were found on the SEM examination. This may have been due to cohesive or mixed failures of the samples that were selected for SEM examination.

The results from this study indicated that: (1) the shear bond strength was not significantly affected when CHX was applied before etching the dentin surface; (2) there was a significant decrease in bond strength when the 1% CHX gel was applied after acid etching; and (3) when the 1% CHX gel was applied to the dentin surface before Clearfil S^3 Bond, the shear bond strength was not affected.

Within the limitations of this *in vitro* study, the following conclusions were drawn: (1) CHX gel application before phosphoric acid did not influence the shear bond strength of PBNT on dentin, but

CHX gel applied after acid etching had an adverse affect on the shear bonding of PBNT; and (2) the 1% CHX gel had no adverse effect on the bond strength of Clearfil S[3].

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