

Bond Strength of Composite Resin to Enamel: Assessment of Two Ethanol Wet-Bonding Techniques

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Abstract

Objective: Ethanol wet-bonding (EWB) technique has been stated to decrease degradation of resin-dentin bond. This study evaluated the effect of two EWB techniques on composite resin-to-enamel bond strength.

Materials and Methods: Silicon carbide papers were used to produce flat enamel surfaces on the buccal faces of forty-five molars. OptiBond FL (OFL) adhesive was applied on enamel surfaces in three groups of 15 namely:

1. Enamel surface and OFL (control);
2. Protocol 1 of the EWB technique: absolute ethanol was applied to water-saturated acid-etched enamel surfaces for 1 minute before the application of ethanol-solvated hydrophobic adhesive resin of OFL 3 times;
3. Protocol 2: progressive ethanol replacement; water was gradually removed from the enamel matrix using ascending ethanol concentrations before OFL application.

Composite build-ups were made and the specimens were stored for 24 hours at 37°C and 100% relative humidity. Shear bond strength test was performed using a universal testing machine at 1 mm/min crosshead speed. Fracture patterns were evaluated microscopically. Data were analyzed with one-way ANOVA and Fisher's exact test ($\alpha=0.05$).

Results: There were no significant differences in bond strength between the groups ($P=0.73$). However, regarding failure patterns, the highest cohesive enamel fractures were recorded in groups 2 and 3.

Conclusion: In this study, although both methods of EWB did not influence immediate bond strength of composite resin to enamel, the majority of failure patterns occurred cohesively in enamel.

Key words: Bond strength; Composite resin; Ethanol wet-bonding; Enamel

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Received: 11 July 2013

Accepted: 6 December 2013

Journal of Dentistry, Tehran University of Medical Sciences, Tehran, Iran (2014; Vol. 11, No. 2)

INTRODUCTION

The past two decades have witnessed a worldwide popularity of esthetic tooth-colored restorations along with great advances in den-

tin-bonding technology of resin-based composite restorations [1]. However, resin-dentin bonds have less durability compared to resin-enamel bonds because the former is dependent

on organic components [2]. Although moisture is required for successful bonding, it has a detrimental effect on bond longevity. The immediate bond strength values of currently available adhesive agents have been reported to be quite high; however, aging results in a significant decrease in resin-dentin bond strength, [2] leading to continuous failure of bonded restorations with time in both etch-and-rinse and self-etch adhesives [3].

Bond strength and durability are critically important for the long-term success of restorations because bond degradation compromises adhesion, leading to formation of gaps between the tooth structure and restorative materials [4]. Thorough infiltration of the adhesive agent into tooth structures is the aim of bonding procedures; it is recommended that the collagen fibrils be encapsulated by the bonding resin so that they would be protected against degradation [2, 3]. It is well established that the collagen fibril encapsulation degree varies based on the type of the bonding agent used, i.e. an etch-and-rinse or a self-etch technique.

With etch-and-rinse adhesives, creation of a decreasing gradient of resin monomer diffusion in the acid-etched dentin structure gives rise to incompletely infiltrated zones at the base of the hybrid layer, consisting of denuded collagen fibrils in the demineralized zone of dentin, manifested by the inadequate penetration of the adhesive resin into the acid-etched tooth structure [5, 6]; this phenomenon may be attributed to insolubility of BisGMA in water-saturated dentin [2].

Previous studies have shown a correlation between the instability of resin-dentin bonds and an increase in the content of hydrophilic resin monomer in dentin adhesives; which accelerates water sorption and compromises the mechanical properties of the adhesive [7, 8]. Fluid sorption can deteriorate resin-dentin bonds produced by currently available hydrophilic adhesives [9, 10].

Relatively hydrophobic monomers can be applied to acid-etched dentin in an EWB procedure [9,10-15] because dehydration by ethanol results in less hydrophilicity of acid-etched dentin, making it possible to apply comparatively hydrophobic monomers in order to penetrate the demineralized collagen network which has shrunk but has not collapsed after application of ethanol [9,14]. From a theoretical point of view, this would improve resin-dentin bond longevity by decreasing water sorption through polymerized hydrophobic adhesive [9]. It has been demonstrated that substituting ethanol for water in Bis-GMA/TEGDMA mixtures contributes to a better infiltration of dentin, resulting in higher bond strength values. Therefore, EWB protocol makes it possible to use hydrophobic resins that absorb little water to affect dentin bonding [1, 9, 10]. Recent studies have shown that EWB increases dentin-adhesive bond strength and longevity [9, 16]. Sadek et al. showed in an in vitro study that this novel bonding protocol is superior to currently available hydrophilic etch-and-rinse adhesives. They showed that a decrease in water sorption and better encapsulation of resin in the demineralized collagen network may improve the longevity of hydrophobic resin-dentin bonds produced by EWB [9]. Li et al. [17] and Sadek et al. [9] reported that EWB protocol results in bond strength values higher than or equal to those produced by water wet-bonding protocol. In addition, Hosaka et al. reported increased bond strength values and longevity with EWB protocol compared to the water wet-bonding procedure [18]. Clinically, the use of ethanol on dentin without contact with enamel margins is impossible. Ethanol has a low surface tension and spreads easily on the surface [19]. Therefore, the aim of this study was to evaluate and compare the influence of two protocols of EWB technique on the bond strength of composite resin to enamel.

The null hypothesis was: There are no differences in bond strength, ultrastructural integrity or fracture patterns of enamel bonded with two different protocols of EWB with a hydrophobic adhesive and the conventional acid-etching technique.

MATERIALS AND METHODS

Shear bond strength evaluation

A total of 45 extracted sound third molars were used in this study. The samples were stored in 0.2% thymol solution at 4°C and used within 2 months after extraction. The crowns were cut from the roots and placed in flat cylindrical acrylic resin molds, with buccal surfaces placed horizontally. Subsequently, the buccal surfaces were ground using wet silicon carbide papers up to 600 grit to produce flat and smooth surfaces. The samples were randomly divided into three groups of 15. In group 1 (control), phosphoric acid was used to etch the flat and smooth enamel surfaces; then, OptiBond FL (OFL) was applied according to the manufacturer's instructions (Table 1).

In group 2, protocol 1 of the ethanol wet-bonding technique, or the simplified technique, was used, i.e. absolute ethanol was applied to water-saturated acid-etched enamel for 1 minute and then hydrophobic adhesive resin of OFL was applied [6, 9, 20].

In group 3, protocol 2 of EWBT, or the progressive ethanol replacement technique, was used as follows: The surfaces were chemically dehydrated before OFL application. Acid-etched enamel surfaces received increasing concentrations of ethanol (50%, 70%, 80%, 95% and 100%, 30 seconds each; i.e. 3 minutes and 30 seconds on the whole), while the enamel samples were kept in the liquid phase before a more concentrated ethanol solution was used. Two coats of the hydrophobic OFL primer were applied to ethanol-treated enamel surfaces. Excess ethanol was evaporated and removed using a gentle current of air for 10 seconds.

A thin layer of the OFL adhesive was applied and light-cured for 20 seconds using a halogen light-curing unit (Coltolux 2.5, Coltene AG, Feldwiesenstrasse Altstätten/ Switzerland) with a light intensity of 480 mW/cm² [6, 9, 20]. Cylindrical plastic molds measuring 2 mm in the internal diameter and 1 mm in height (Orthorings, Ortho Organizers Inc, CA, USA) were placed and fixed on the enamel surfaces at an ambient temperature of 22±1°C. Composite resin build-ups were constructed in all groups using a light-cured composite resin (Point 4, Kerr, Orange, USA) in one increment (Table 1) according to the manufacturer's instructions.

Table1. The materials used in the study, their compositions and modes of applications.

Material	Manufacturers' instructions	Composition
OptiBond FL (Three step, etch & rinse adhesive) (Kerr, Orange, CA, USA)	1. Etch with 37.5% phosphoric acid (15 seconds) 2. Rinse (15 seconds) and dry (5 seconds) 3. Apply primer and rub for 15 seconds. Dry for 5 seconds 4. Apply adhesive in a uniform thin layer 5. Light cure for 30 seconds.	Etchant: 37.5% H ₃ PO ₄ FL Primer: HEMA, GPDM, MMEP, water, ethanol, CQ, BHT FL Adhesive: Bis-GMA, HEMA, GDMA, CQ, ODMAB, filler (fumed SiO ₂ , barium aluminoborosilicate, Na ₂ SiF ₆) coupling factor A174(48 wt% filled)
Point 4 Microhybrid composite resin Kerr Corporation, Orange, CA, USA	Apply in 2 mm layers, light cure for 20 seconds.	Ba Si glass TEGDMA, EBPADMA, silane treated barium glass, silica(spherical), photo curing system

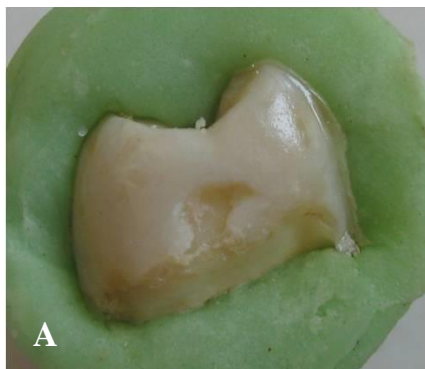


Fig1. A and B, Two specimens with cohesive enamel fracture related to group 3.

Composite resin was light-cured using a halogen light-curing unit (Coltolux 2.5, Coltene AG, Feldwiesenstrasse Altstätten/Switzerland) with a light intensity of 480 mW/cm².

The samples were stored at 37°C for 24 hours and then underwent a shear bond strength (SBS) test at a crosshead speed of 0.5 mm/min using a universal testing machine (Dartec, HC10, Dartec Ltd, Stourbridge, UK). SBS values were calculated by dividing the force at fracture by the initial bonded area.

One-way ANOVA was applied in order to analyze the effect of bonding protocol on enamel SBS values using SPSS Ver. 11.5 software. Statistical significance was set at P<0.05.

Furthermore, the fracture patterns of composite resin cylinders on enamel surfaces were evaluated under a light microscope at ×20 magnification and classified as follows using Fisher's exact test:

- Cohesive fracture: fracture in the composite resin or tooth structure
- Adhesive fracture: fracture at the adhesive interface
- Mixed fracture: adhesive/cohesive fracture (Table 3)

Interface evaluation by scanning electron microscopy (SEM)

Two extra samples were prepared for SEM evaluation in each group.

Table 2. Bond strength of the specimens in the study groups in MPa (P=0.730)

Groups	Group definitions	Mean± SD	CI 95%		Min.	Max.
			LB	UB		
1	Control	29.70 ± 11.71	23.21	36.19	14.96	53.18
2	Ethanol 100%	32.16 ± 10.82	26.17	38.16	15.60	51.91
3	Ascending ethanol	29.27 ± 9.50	24.00	34.54	11.78	46.81

Table 3. Distribution of modes of fractures in study groups, N(%)

Groups	Group definitions	Cohesive in enamel	Cohesive in composite	Mixed
1	Control	5 (33.3%)	5 (33.3%)	5 (33.4%)
2	Ethanol 100%	8 (53.34%)	0 (0%)	7 (46.66)
3	Ascending ethanol	9 (60.00%)	0 (0%)	6 (40.00%)



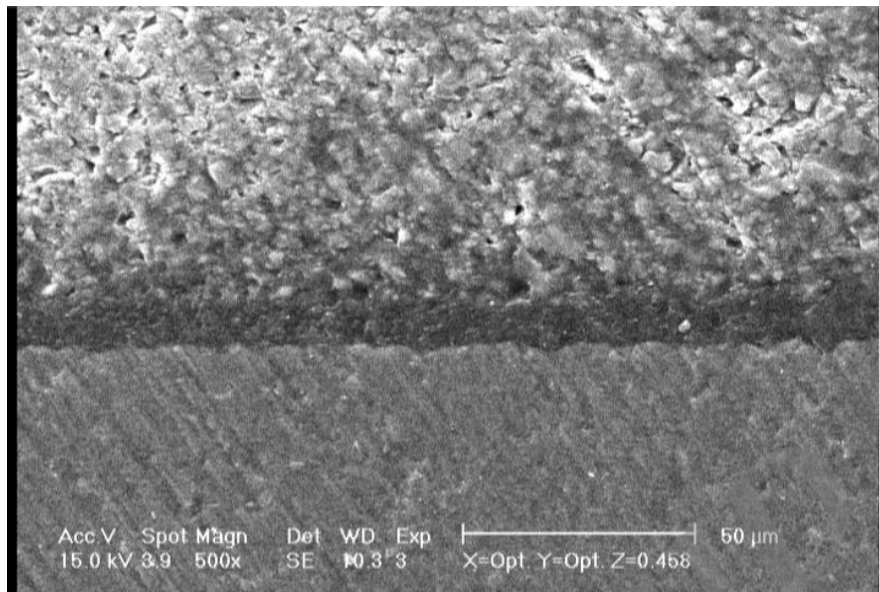


Fig 2. Enamel/composite interface in group 1 (×500)

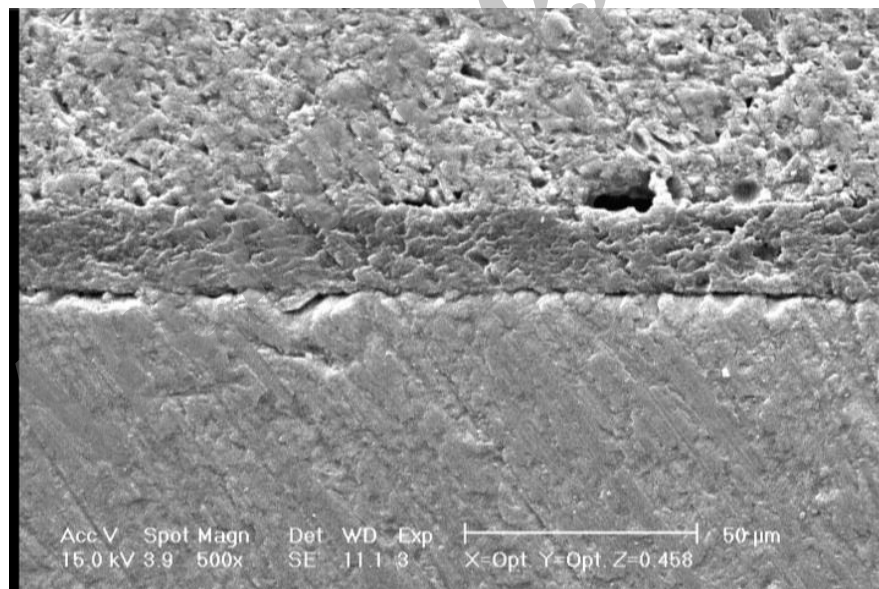


Fig 3. Enamel/composite interface in group 2 (×500)

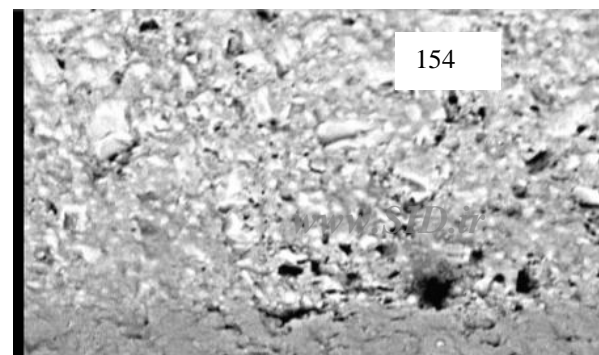


Fig 4. Enamel/composite interface in group 3 ($\times 500$)

After each sample was prepared according to the method described above, the samples were prepared by sectioning.

The samples were dehydrated using ascending concentrations of ethanol (50%, 70%, 95%, and 100%) for 1 hour, placed in acrylic resin and polished using descending grits of abrasive papers (400, 600, 800, 1200, and 1500) and 0.5 μ diamond paste with a polishing cloth. The samples were placed in an ultrasonic bath for 10 minutes between polishing steps. The exposed interfaces were treated with 6N hydrochloric acid for 30 seconds followed by a 10-minute immersion in 2.5% NaOCl.

After a 20-minute ultrasonication procedure, the samples were dehydrated for 24 hours, affixed to an aluminum mounting stub, and sputter-coated with platinum-gold to a thickness of 10 nm for analysis under a scanning electron microscope (SEM). SEM images were provided under different magnifications at a distance of 20 mm. An accelerating voltage of 15.0 kVp was used for the analysis.

RESULTS

Descriptive statistics of SBS values in MPa for the three groups under study are presented in Table 2. Bond strength values of all the study groups to enamel were not significantly different ($P=0.730$). One-way ANOVA did not reveal any significant differences in bond strength values of composite resin to enamel surfaces between groups 2 and 3 using two different protocols of EWB ($P>0.05$).

The fracture patterns are summarized in Table 3. There were significant differences in fracture patterns of composite resin/enamel surfaces between the study groups. According to the results, the majority of enamel cohesive fractures were recorded in enamel in group 3 ($P<0.04$, Figure 1). Furthermore, the SEM photomicrographs are presented in Figures 2-4 for the study groups, respectively. As it appears in the figures, the integrities of adhesive interfaces are similar in the three groups. The thickness of the adhesive interface seems to be less for group 1, which might be attributed to

lower ability of the adhesive to penetrate into the acid-etched wet enamel.

Figures 1-3 show good adaptation between the restorative material and enamel in all under study groups.

DISCUSSION

Failure at dentin-adhesive interface and reduction in bond strength values with time have been reported, although great technical advances have improved resin-dentin bond performance [2, 20] and this might explain the decreased longevity of adhesive restorations in the clinic [21]. Thus, researchers and clinicians continuously seek new strategies, like EWB, to improve resin-dentin bond durability. In the present study in group 1 (the control), the conventional gold standard etch-and-rinse technique with the application of OFL was used to bond composite resin to enamel; the bond strength was 29.70 ± 11.71 MPa, which is consistent with the results of previous studies [21].

In this study, two different protocols of EWBT were used to evaluate enamel bond strength in groups 2 and 3. According to the literature, when the bonding substrate is dentin, in the simplified technique absolute ethanol is applied to water-saturated acid-etched dentin surfaces for 1 minute before application of ethanol-solvated hydrophobic resin comonomer blends [22, 23]. This simplified technique was initially advocated in an attempt to apply hydrophobic resin comonomers to acid-etched dentin in as short a time as possible [3]. The same protocol was performed for the samples in group 2 in the present study. The mean SBS value for this group was 32.16 ± 10.82 MPa. Based on some concerns, when the bonding substrate was dentin, this protocol was highly technique-sensitive and did not completely reduce dentin permeability or replace the water by outward fluid flow without the assistance of tubular occluding agents, [3, 24] even with three consecutive applications of absolute ethanol [9].

In the alternative version of the technique, water was removed step-by-step from the collagen matrix by the application of consecutive ascending concentrations of ethanol [9, 25]. This protocol was carried out in group 3 of this study for the enamel substrate; the mean SBS value for this group was 29.27 ± 9.50 MPa. Based on the literature, this version of the technique is time-consuming and impractical in the clinic [26]. Osorio et al. reported that with both techniques when ethanol replacement technique was not carried out with sufficient care and precision to protect water-saturated collagen matrix from air, the surface tension at the air-collagen interface can easily collapse the collagen matrix, preventing sufficient infiltration of adhesive monomers [26].

Chuang et al. reported that wetting the enamel did not exert a significantly adverse effect on the marginal integrity of restorations with the use of either acetone- or ethanol-based adhesives. However, use of self-etching adhesives might give rise to a higher incidence of margin integrity loss [27]. Recently, Oyama et al. compared the surface free energies and enamel bond strengths of one-step self-etching adhesives with and without an oxygen-inhibited layer. The oxygen-inhibited layer in that study had been removed with ethanol [28]. In contrast with this study, they reported that presence of ethanol as an oxygen-inhibited layer in one-step self-etching adhesives gave rise to a higher enamel bond strength value. They concluded that the oxygen-inhibited layer might act as an electron donor, accelerating the polymerization reaction at the adhesive-composite resin interface [28].

In the present study, OFL was used as a criterion to designate the commercial adhesive as hydrophilic because of the presence of 2-hydroxyethyl methacrylate (HEMA) in both the primer and the bonding resin. The current study showed that ethanol wet-bonding protocols do not significantly decrease enamel SBS values. When bonded with OFL, ethanol treatment did not affect either the SBS or the

interfacial morphology under SEM. However, the number of adhesive fractures decreased in the study groups, except in group 1. Therefore, the null hypothesis that ethanol wet-bonding protocol has no effect on the SBS values of enamel bonded with OFL was confirmed, and the other that EWB has no effect on the fracture pattern of enamel-composite resin bond was rejected. In addition, the chemical composition of this adhesive is presented in Table 1. As it is seen in the table, the primer is composed of HEMA, polyalkenoic acid copolymer and water, and the bonding resin contains BisGMA and HEMA. The presence of HEMA in both the primer and bonding resin was used as a criterion to designate the adhesive as hydrophilic. However, in the present study there was no clear-cut hydrophobic resin as an experimental system contrary to the commercial adhesive, which might be a limitation of the present study. In a similar manner, a hydrophobic resin blend consisting of BisGMA and TEGDMA becomes more hydrophilic by incorporating these comonomers into ethanol. Regarding dentin, these steps pave the way for improved miscibility of the solvated adhesive and the collagen matrix, [5] making it possible for the ethanol-solvated hydrophobic resin blend to penetrate into ethanol-saturated collagen matrices. Although penetration of resin monomers usually gives rise to a diffusion gradient of resin infiltration within collagen matrices, recent studies with two-photon laser confocal microscopy [29] and micro-Raman spectral analysis [30] have shown that a rather homogeneous distribution of hydrophobic resins within the hybrid layer is achieved with EWB technique. It should be pointed out that ethanol is a bipolar solvent with less hydrogen bond capacity compared to water, which results in the chemical dehydration of the demineralized collagen network [10]. A study by Sadek et al. revealed that the resin-dentin bond formed with EWB technique and hydrophobic adhesives was still in place after 18

months of storage in water without the presence of MMP inhibitors [9]. With both ethanol wet-bonding protocols, evaporation of water from the water-saturated dentin collapsed the collagen matrix. However, Osorio et al. showed that the collapsed collagen matrix could be rehydrated and re-expanded by 50% water present in the 50% ethanol using the progressive technique [26].

Another noteworthy point is that since the introduction of the concept of “ethanol wet-bonding”, various ethanol-wet protocols have been used to optimize this technique and the majority of these techniques have been studied on coronal dentin; however, although some promising results have been reported with coronal dentin, when EWB technique is used on coronal substrates, in addition to being time consuming, rehydration by dentinal fluid from the pulp decreases the efficacy of ethanol dehydration in vital pulp tooth dentin [2,3,10]. Therefore, recently some researchers have postulated that a dry non-vital substrate, such as root dentin after endodontic treatment, might be a more suitable substrate for EWB technique [10,31]. Recently, Pei et al. reported that EWB technique with a stepwise application of ethanol might be potentially beneficial in the bonding of hydrophobic adhesives to root dentin [10]. However, to the best of our knowledge, no study to date has evaluated the effect of the technique on enamel bond strength. Further research is necessary to make a judgment about the efficacy of these bonding techniques with vital and non-vital teeth with different tooth substrates, including enamel. However, in the case of enamel, while there were no significant differences in bond strength between the two protocols and the control group, EWB technique influenced the enamel fracture pattern. As most fractures were in the enamel structure, it seems that the EWB protocol results in dehydration of enamel crystallites, rendering enamel rods more susceptible to fracture.

It should be noted that one of the techniques used to simulate clinical situations in vitro is thermocycling; which was not used in the present study and might be considered as one of the limitations of the present study. Given the fracture pattern results, it is possible that thermocycling or application of other ageing techniques, such as storage in water, might affect the bond strength; however, further studies are recommended in this regard.

One of the interesting findings of the present study, which attracted the attention of the authors, was a minor discoloration of the samples at enamel-composite resin interface in groups 2 and 3. Further clinical studies are recommended with EWBT to evaluate this discoloration.

CONCLUSION

Within the limitations of this study it can be concluded that:

1. There were no significant differences in enamel bond strengths between the two protocols of EWB and conventional acid-etch technique.
2. Both protocols of EWB technique might influence the enamel fracture patterns, i.e. the majority of fractures were in the enamel structure.

ACKNOWLEDGMENTS

This study (#29167) was financially supported and approved by Isfahan University of Medical Sciences, Isfahan, Iran.

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