Published in final edited form as:

J Dent. 2013 March ; 41(3): 231–240. doi:10.1016/j.jdent.2012.11.006.

Effect of application mode on interfacial morphology and chemistry between dentin and self-etch adhesives

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Abstract

Objective—To investigate the influence of application mode on the interfacial morphology and chemistry between dentin and self-etch adhesives with different aggressiveness.

Methods—The occlusal one-third of the crown was removed from un-erupted human third molars, followed by abrading with 600 grit SiC under water. Rectangular dentin slabs were prepared by sectioning the tooth specimens perpendicular to the abraded surfaces. The obtained dentin slabs were treated with one of the two one-step self-etch adhesives: Adper Easy Bond (AEB, PH~2.5) and Adper Prompt L-Pop (APLP, PH~0.8) with (15s, active application) or without (15s, inactive application) agitation. The dentin slabs were fractured and the exposed adhesive/dentin (A/D) interfaces were examined with micro-Raman spectroscopy and scanning electron microscopy (SEM).

Results—The interfacial morphology, degree of dentin demineralization (DD) and degree of conversion (DC) of the strong self-etch adhesive APLP showed more significant dependence on the application mode than the mild AEB. APLP exhibited inferior bonding at the A/D interface if applied without agitation, evidenced by debonding from the dentin substrate. The DDs and DCs of the APLP with agitation were higher than those of without agitation in the interface, in contrast to the comparable DD and DC values of two AEB specimen groups with different application modes. Raman spectral analysis revealed the important role of chemical interaction between acid monomers of self-etch adhesives and dentin in the above observations.

Conclusion—The chemical interaction with dentin is especially important for improving the DC of the strong self-etching adhesive at the A/D interface. Agitation could benefit polymerization efficacy of the strong self-etch adhesive through enhancing the chemical interaction with tooth substrate.

Keywords

self-etch adhesives; degree of conversion; micro-Raman; agitation; adhesive/dentin interface

1. INTRODUCTION

Adhesive dentistry has evolved dramatically towards a trend eliminating as many steps as possible in the binding protocol. Self-etch adhesives have attracted considerable interest due

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to their great advantages in simplifying the bonding procedure and reducing the potential for tooth sensitivity.¹⁻³ However, simplification does not guarantee equal or improved effectiveness. Self-etch adhesives, in particular the all-in-one (or one-step) adhesives have often been questioned regarding their clinical reliability. They showed reduced immediate bond strengths as compared to those measured from multi-step adhesives.^{4,5} The long-term bonding effectiveness produced by all-in-one self-etch adhesives is also a matter of concern as a result of expedited aging along the hybrid layer region.^{6,7} Numerous studies have reported the significant bond strength reduction and extensive interfacial nanoleakage within the adhesive/dentin (A/D) interface for the simplified adhesive systems.^{8,9}

The fundamental principle of adhesion to tooth substrate is based on an exchange process, in which mineral is removed from the dental surface and replaced by resin monomers that subsequently polymerize.^{2,3} This process, also known as micro-mechanical interlocking or hybridization, is believed to induce the formation of the hybrid layer. Ideally, the hybrid layer is characterized as a 3-dimensional polymer/collagen network that provides both a continuous and a stable link between the bulk adhesive and dentin substrate.¹⁰ However, considerable evidences have suggested that the hybrid layer at the A/D interface is a weak link in resin composite restorations.¹¹⁻¹⁵ It was disclosed that the A/D interface produced by self-etch adhesives performs as a water-permeable membrane after polymerization.¹⁶⁻¹⁸ This permeability feature of self-etch adhesives was shown to correlate with the presence of unreacted monomers within the hybrid layer.¹⁹⁻²¹ Especially the all-in-one self-etch adhesives contain the highest percentages of hydrophilic monomers among the currently available adhesive systems, which inevitably leads to the increase of the water uptake and plasticization.^{2,22} The presence of water and high concentration of hydrophilic domains would affect the polymerization efficacy, resulting in suboptimal degree of conversion (DC) as well as porous structure as a result of the elution of unreacted monomers.^{19,23} Consequently, the long-term stability of the A/D interface would be undermined.

Due to the importance of an optimal DC in achieving reliable bonding properties,²⁴⁻²⁶ our previous study has investigated the photopolymerization efficacy of self-etch adhesives on dentin.²⁷ By employing two all-in-one self-etch adhesives (APLP and AEB) with distinct aggressiveness, the study was focused on the influence of potentially different chemical interaction with dentin mineral (mainly hydroxyapatite, HAp) on the DCs of the two systems within the adhesive layers. The influence of application mode on the DC of the adhesive layer was also studied as recent findings had evidenced the significant role of active agitation in the improved mechanical properties.²⁸⁻³⁰ Since the chemical interaction between the acidic monomers of self-etch adhesives and dentin mineral primarily occurs at the A/D interface, it is supposed that agitation and chemical interaction would more directly affect the interface region rather than the bulk adhesives. Therefore, the objective of this study was to investigate the influence of application mode on the interfacial morphology and chemistry such as degree of dentin demineralization (DD) and DC between dentin and self-etch adhesives with different aggressiveness, and to understand the role of the chemical interaction in these processes. The hypothesis tested was that agitation would not lead to differences in DD and DC at the A/D interface for the self-etch adhesives used in this study.

2. MATERIALS & METHODS

2.1. Adhesive/dentin specimen preparation

The teeth used in this study were eight extracted non-carious, un-erupted human third molars stored at 4°C in phosphate buffered saline (PBS) containing 0.002% sodium azide. The teeth were collected after obtaining the patients' informed consent under a protocol approved by the University of Missouri Kansas City adult health sciences IRB. The occlusal one-third of the crown was removed by means of a water-cooled low-speed diamond saw

(Buehler Ltd, Lake Bluff, IL, USA). Each prepared dentin surface was examined under a light microscope (Nikon Instruments Inc., Eclipse ME600P, Japan) to ensure it was free of enamel. A uniform smear layer was created on the dentin surface by using wet 600-grit silicon carbide sandpaper for 30 s. The tooth specimens were then sectioned perpendicular to the abraded surfaces to obtain 24 equal sized rectangular slabs ($10 \times 2 \times 1.5$ mm). Each slab was notched from the middle position of the bottom side opposite to the abraded surface for later fracturing. The prepared dentin slabs were then treated with one of the two self-etch adhesives APLP (3M ESPE, Seefeld, Germany, pH~0.8) and AEB (3M ESPE, Seefeld, Germany, pH~2.5) ³¹ (see Table 1 for adhesive compositions). Each adhesive was applied onto the abraded dentin surface with (15s, active application) or without (15s, inactive application) agitation, gently air-dried for 5s, and light-cured for 10s (600 mW/cm^2 , Spectrum 800 halogen light, Dentsply, Milford, DE, USA) with glass cover slips on the top. The specimens obtained were abbreviated as APLP-WA, APLP-WOA, AEB-WA, and AEB-WOA for APLP with and without agitation, and AEB with and without agitation, respectively. To avoid water contamination and potential release of un-reacted monomers during cutting, the slabs were not cut by the water-cooled saw, instead, they were fractured from the notches. The exposed A/D interfaces and adhesive layers were ready for the following analyses.

2.2 Micro-Raman spectroscopy

A LabRam HR 800 Raman spectrometer (Horiba Jobin Yvon, Edison, NJ, USA) using monochromatic radiation emitted by a He-Ne laser (a wavelength of 632.8 nm and excitation power of 20 mW) was employed. It was equipped with a confocal microscope (Olympus BX41), a piezoelectric (PI) XY stage with a minimum step width of 50 nm, and an air-cooled CCD detector of 1024×256 . The spectra were Raman-shift-frequency calibrated with known lines of silicon. The laser was focused through a $100\times$ Olympus objective to obtain a beam diameter of $\sim 1 \mu\text{m}$. Raman spectra were acquired starting from the dentin towards the adhesive layer at $1\text{-}\mu\text{m}$ intervals. All the spectra were obtained over the spectral region of $200\text{--}2000 \text{ cm}^{-1}$ and with an acquisition time of 60 s. The degree of demineralization (DD) as a function of location was determined from the Raman band ratios of 959 cm^{-1} ($\nu_1 \text{ PO}_4$ of the mineral component in dentin) relative to 1458 cm^{-1} (δ of CH_2). ³²

$$DD = \left(1 - \frac{\text{Intensity}_{959 \text{ cm}^{-1}}^{\text{interface}} / \text{Intensity}_{1458 \text{ cm}^{-1}}^{\text{interface}}}{\text{Intensity}_{959 \text{ cm}^{-1}}^{\text{dentin}} / \text{Intensity}_{1458 \text{ cm}^{-1}}^{\text{dentin}}}\right) \times 100\% \quad (1)$$

The degrees of conversion (DCs) of the adhesives were calculated based on the band area ratios of 1640 cm^{-1} (ν of $\text{C}=\text{C}$) to 1458 cm^{-1} (δ of CH_2) ($R_{1640/1458}$), according to the following equation:

$$DC = \left(1 - \frac{\text{Intensity}_{1640 \text{ cm}^{-1}}^{\text{sample}} / \text{Intensity}_{1458 \text{ cm}^{-1}}^{\text{sample}}}{\text{Intensity}_{1640 \text{ cm}^{-1}}^{\text{monomer}} / \text{Intensity}_{1458 \text{ cm}^{-1}}^{\text{monomer}}}\right) \times 100\% \quad (2)$$

Two-point baseline and maximum band area ratio protocol were used to measure the band intensities. Each DC value was determined and averaged based on at least three Raman spectra. A one-way analysis of variance (ANOVA, $\alpha = 0.05$) was used (GraphPad InStat version 3.06, GraphPad Software, Inc.) to analyze the data on the DD and DC.

2.3 Scanning electron microscopy (SEM)

The specimens characterized by micro-Raman spectrometer were subject to SEM examination subsequently. In order to evaluate the interfacial morphology, the specimens were treated by soaking in 5N HCl for 30 sec, washed with water, followed by soaking in 5% NaOCl for 30 min and another water-wash and air-drying. The prepared specimens were mounted on aluminum stubs and sputter-coated with 20 nm layer of gold-palladium. The adhesive/dentin interfaces were then examined at a variety of magnifications and tilt angles in the SEM (Philips XL 30, Eindhoven, Netherlands) at 5kV.

3. RESULTS

Representative SEM micrographs of the A/D interfaces of the four specimen groups are shown in Figures 1 and 2. Resin tags were clearly observed in all of the images. However, APLP system (Figure 2) showed larger number of tags than AEB (Figure 1). APLP and AEB also showed difference in the formed hybrid layer. With the current SEM technique and magnification, no significantly microscopic presence of an acid resistant hybrid layer was detected in both of the AEB specimens. In contrast, APLP-WOA and APLP-WA displayed distinct hybrid zones with approximate thicknesses of 2–3 μm . Furthermore, the results of Figures 1 and 2 indicated that APLP showed more significant dependence on the application mode than AEB. As was observed in Figure 2 (B), the APLP-WA tags appeared in a form of resin tag “bunch” that consisted of multiple highly combined tags. This feature of the APLP-WA tags also led to a triangular shape at the entrance of the dentinal tubules. The formed tags of APLP-WOA specimen, however, were more separated, and displayed an evident feature of discontinuity or loss of integrity (Figure 2 (A)). Figure 2 (C) clearly showed the overall feature of the de-bonded APLP-WOA adhesive layer and tags. Same as the other three groups (APLP-WA, AEB-WOA, and AEB-WA), APLP-WOA was subject to the treatment of 5N HCl (30s) followed by 5% NaOCl (30 min), which as a common SEM preparatory procedure, was used to expose the hybrid and resin tags at the A/D interface. The present result indicated that APLP-WOA specimens could not withstand the etching and bleaching treatments. Such a debonding phenomenon was not observed with the APLP-WA, AEB-WOA, and AEB-WA specimen groups.

Representative Raman spectra of dentin and two adhesives AEB and APLP (10s-light-cured on glass) with their major bands and associated assignments are shown in Figure 3. Among them, the characteristic bands for dentin at 1667 cm^{-1} , 1245 cm^{-1} , 1069 cm^{-1} and 959 cm^{-1} , and the characteristic bands for both adhesives at 1718 cm^{-1} , 1609 cm^{-1} , and 1113 cm^{-1} do not overlap with each other, thus were used to locate the A/D interface from the micro-Raman mapping spectra. Raman bands at 1640 cm^{-1} and 1458 cm^{-1} were used to determine the DC in the interface. Since the 1640 cm^{-1} band partially overlaps with the 1667 cm^{-1} band, spectral subtraction is necessary prior to the DC calculation.

Micro-Raman mapping spectra across the A/D interface for the four groups are shown in Figures 4 and 5. The spectra representing dentin, interface, and adhesives were identified with different colors. The spectrum (or spectra) of the interface was (or were) determined in case that significant Raman bands associated with both dentin and adhesives were noted. The result from Figure 4 indicated that at our current scanning interval of 1- μm , only one spectrum of interface could be found for both the AEB groups regardless of application mode. The intensities of the major bands of AEB-WOA and AEB-WA at the interfaces were also comparable. In contrast, APLP-WOA and APLP-WA (Figure 5) displayed apparent spectral difference at the interfaces. APLP-WOA exhibited two spectra of the interface while the number of the interface spectra for APLP-WA was three. Furthermore, the Raman band at 959 cm^{-1} appeared different intensities for APLP-WOA and APLP-WA. This band is assigned to be ν_1 of PO_4 for the mineral component in dentin.³⁴ Demineralization from

acidic monomers of the self-etch adhesives can reduce the level of this band. Therefore, changes of this band in intensities can serve as the measure of dentin demineralization. In the present study, the Raman band ratio of 959 cm^{-1} relative to 1458 cm^{-1} (as an internal standard band) was employed to calculate the degree of the demineralization (DD) (equation 1) as a function of location, as shown in Figure 6. The result suggested that AEB showed no significant difference with DD for WOA and WA application modes: both exhibited DDs of about 64% at the 1st micron (interface), and about 10% or lower at 2nd and 3rd microns. For APLP, application mode led to significant difference in DD. APLP-WA showed universally higher DDs (95%, 93.5%, and 59.7% at 1st, 2nd, and 3rd microns) than APLP-WOA (84.8%, 80.4%, and 16.8% at 1st, 2nd, and 3rd microns).

The DCs at A/D interfaces of four groups were also determined based on equation (2). In order to eliminate the interference from the organic component of dentin at 1667 cm^{-1} , spectral subtraction was performed, as shown in Figure 7. The subtracted spectrum (Figure 7 C) was then used to calculate the DC. Figure 8 showed the obtained DCs at the A/D interfaces. The DCs of the adhesive layers at 1st and 15th microns from interface were also given for comparison. The result indicated that within A/D interfaces, the DCs of AEB-WOA and AEB-WA showed no significant difference (~96%), while the DCs of APLP-WA (88.2% and 89% at 1st and 2nd microns in the interface) were apparently greater than those of APLP-WOA (79.9% and 76.3% at 1st and 2nd microns in the interface). In addition, APLP-WA and APLP-WOA exhibited a slightly increasing and a decreasing trend of DC from 1st to 3rd (or 2nd) micron, respectively. But the differences among each position were not statistically significant. As the DCs at interfaces were compared with those of adhesive layers, no significant difference was found between the DCs at interfaces and those at the 1st micron in adhesive for all of the four groups. Nevertheless, the DCs at the 15th micron in adhesive layers appeared significantly lower than those at interfaces for all of the groups.

To obtain better understanding on the mechanism of the observed DCs, particularly for the APLP specimens, the spectra of APLP-WA and APLP-WOA at the 1st micron of interface, and in the adhesive layer at the 1st and 15th μm from the interface were compared. The spectrum of the APLP cured on glass was also given for reference. The spectral contribution of collagen had been subtracted from the spectra in the interface, so that the influence from the organic component of dentin could be avoided. Spectral comparison of Figure 9 indicated that the band 959 cm^{-1} for $\nu_1\text{ PO}_4$ in the interface showed greater intensities than those in adhesive layers. In addition, this band appeared at the shifted wavenumbers of $960\text{--}962\text{ cm}^{-1}$ in adhesive layers. Moreover, it was observed that as compared with the band position of $\nu_3\text{ PO}_4$ (at 1037 cm^{-1}) for APLP specimens in the interface and the 1st micron in the adhesive layer, the same band for APLP-WOA at the position of the 15th micron in the adhesive layer exhibited a higher wavenumber at 1042 cm^{-1} , which was closer to that of APLP-on glass (at 1043 cm^{-1}).

4. DISCUSSION

The proposed hypothesis that agitation would not lead to differences in DD and DC in the A/D interface for the self-etch adhesives was partially rejected. The results indicated that for the mild self-etch adhesive AEB, agitation showed no significant influence on the obtained DD and DC. However, the DD/DC values of the strong self-etch adhesive APLP were to a great extent dependent on the application mode. Agitation could enhance the DD and DC of APLP in the A/D interface. The data might be correlated to the interfacial morphology of the specimens. SEM results indicated that compared to the hybrid layer/interface for the APLP-WA, APLP-WOA lost integrity at the interface following a regular SEM preparatory procedure. The relatively lower DC at the interface of APLP-WOA with dentin might have

contributed to the less optimal bonding performance as shown from the morphological observations.

The previous study suggested that agitation was one of the key factors to determine the DC of the APLP adhesive layer as applied on dentin.²⁷ The data showed that agitation could improve the DC of APLP in the adhesive layer. The observation was to a great extent associated with the increased chemical interaction (with agitation) of acidic monomer with hydroxyapatite in dentin. The chemical interaction could provide a buffering effect to the acidic monomer, which otherwise would consume more coinitiator of amine to compromise photopolymerization efficacy.^{35, 36} The chemical interaction was also believed to be responsible for the observed decreasing trend of DC from the inner to outer APLP adhesive layers, since the availability of HAp involved in the chemical reaction also decreased with the distance from dentin. The current study disclosed that APLP showed distinct DCs even in the interface if different application modes were employed. Chemical interaction at the A/D interface might have also played an important role in the process. Raman spectral comparison (Figure 9) indicated that the vibration of ν_1 PO₄ at around 960 cm⁻¹ of the APLP in interface appeared differently in both intensity and wavenumber position depending on the application mode. Generally the Raman vibration of ν_1 PO₄ is contributed from two components during the demineralization process at the interface: one is the undemineralized dentinal mineral and the other is the calcium phosphate complex produced from chemical interaction of hydroxyapatite with acidic monomers.^{34, 37} The ν_1 PO₄ for the undemineralized dentinal mineral appears at 959 cm⁻¹ as a sharp and high-intensity peak in contrast to a broad peak at 960–962 cm⁻¹ for the calcium phosphate complex. The ν_1 PO₄ for undemineralized dentinal mineral likely transforms to that of the calcium phosphate complex as the demineralization and chemical interaction proceed. It can be found from Figure 9 that the interface of APLP-WA exhibited a broader and lower-intensity of ν_1 PO₄ band than APLP-WOA. Meanwhile, the ν_1 PO₄ of APLP-WA in the interface appeared at a higher wavenumber (960 cm⁻¹) than that of APLP-WOA (959 cm⁻¹). The current result probably suggested that a higher content of calcium phosphate complex produced at the interface of APLP-WA than APLP-WOA, thus a greater DC was also anticipated. Another important Raman peak related with structural change of APLP during the chemical interaction with HAp was associated with the ν_3 PO₄ located at 1043 cm⁻¹ (as appeared in the spectrum of APLP on glass). This peak might shift to a lower wavenumber in case calcium phosphate complex is formed. Figure 9 depicted that the ν_3 PO₄ peaks in spectra of the interface and adhesive layer at the 1st micron for both APLP-WA and APLP-WOA appeared at a lower wavenumber of 1037 cm⁻¹. In contrast, the same peaks for the adhesive layer at the 15th micron were located at 1039 and 1042 cm⁻¹ for APLP-WA and APLP-WOA, respectively. This result suggested that the A/D chemical interaction occurred more intensively in the interface and adhesive layer close to the interface (such as at the 1st micron from interface) than a further position (such as at the 15th micron from the interface). As a result, higher DCs were observed (Figure 8).

It was interesting to notice that in the interface, the DC distribution of APLP specimens with and without agitation exhibited slightly increasing and decreasing trends, respectively. Even if the trends were not statistically significant, they may imply the discrepancy of polymerization processes of APLP-WA and APLP-WOA in a specific environment like the hybrid layer. Previous results have shown that the highest level of suboptimal polymerization and nanoleakage of self-etch adhesives occurred at the bottom of interface and at the peritubular level.^{20, 25, 26} These areas represent the most hydrophilic regions of the interface as a result of intimate contact with dentinal fluids. In the current study, the strong etching function of APLP also allowed the dentinal fluids release and to be involved in polymerization. It is reasonable to speculate that polymerization of adhesives at the bottom of the interface was more interfered by water than that at the top. The DC result of

APLP-WOA in interface (Figure 8 (B)) correlated well with this speculation. The DC of APLP-WOA at the bottom of the interface was even lower than that in adhesive layer close to the interface (Figure 8 (B)). As agitation was applied, the dentinal fluids would be mixed with APLP more evenly throughout the interface area. Therefore a reduction of DC from the top to bottom of the interface was not observed for the APLP-WA. Conversely, the DC of APLP-WA showed a slightly increasing trend. This was probably related to the chemical interaction of APLP with dentinal mineral, which improved the DC of APLP.

The rationale behind the self-etch approach assumes that acidic monomers will penetrate beyond the smear layer into mineralized dentin through partially dissolving HAP to generate a resin-infiltrated zone with mineral incorporated.^{3, 38, 39} If the simplified self-etch adhesives fail to penetrate beyond the smear layer and to demineralize the dentinal mineral to form a hybridized zone, the bond strength might be compromised.⁴⁰ In the attempt to improve bonding effectiveness of the self-etch adhesives, active application or agitation could be used as a simple and rapid technique. Agitation can provide a consistent etching effect and enhance the interaction of acidic monomers with etched dental substrate.⁴¹ This procedure can also increase the moieties kinetics and allow for better monomer diffusion.^{38, 42} Recent studies have demonstrated that agitation could improve the immediate resin-dentin bond strength as well as reduce the degradation rate of the self-etch adhesive systems.^{28–30} The findings were in agreement with our present SEM result of the strong self-etch adhesive APLP that agitation could help to achieve more optimized bonding interface, while the specimens without agitation displayed a debonding feature. More importantly, the micro-Raman spectroscopic data presented detailed information of the monomer conversion at the A/D interface during polymerization of self-etch adhesives. The calculated DCs of the APLP at the A/D interface provided important evidence regarding the significant role of the agitation in improving the bonding performance of strong self-etching adhesive in terms of their polymerization behavior. The results might benefit the current understanding on the mechanism of active application mode improving the bonding performance of self-etch adhesives.

The DC at the A/D interface of the mild self-etch adhesive AEB did not show significant dependence on the application mode. This might have been related to its favorable composition and pH value for achieving a fairly high DC.³¹ Also there was no apparent morphological difference of the interface detected between AEB-WOA and AEB-WA. The present result suggested that polymerization efficacy and morphology of the AEB at interface were less dependent on the application mode if compared with those of the strong adhesive APLP. Nevertheless, the comparison of SEM images of the two groups might also reveal that more resin tags were produced for the AEB-WA than AEB-WOA. This fact suggested that agitation produced slightly more demineralization to facilitate diffusion of the monomers, which consequently led to formation of more AEB resin tags. Further investigation is needed to find out the relationship of mechanical properties with the application mode of the mild self-etch adhesive AEB.

Based on the micro-mechanical interlocking mechanism,² successful adhesion to dental substrate is strongly dependent on the capacity of the acidic monomers penetrating and demineralizing dental mineral. However, a deeper demineralization and thicker hybrid layer are not necessarily associated with better bonding interface or properties. As shown in the current results, even if the thicknesses of both demineralization and hybrid layer produced by APLP were greater than those of AEB, the morphological result disclosed that the formed A/D interface of APLP was even more inferior to that of AEB in the case of inactive application. Actually, both laboratory and clinical data have shown that strong self-etch adhesives such as APLP generally underperform at dentin as compared to mild self-etch adhesives, in particular with regard to bond durability and restoration longevity.^{43, 44} As

revealed by the current study, this was partially related to the suboptimal polymerization performance of strong self-etch adhesives, which was arisen from interference of water in dentin tubules due to profound etching of acidic monomers, as well as negative influence of monomer acidity on initiating efficacy of the coinitiator in the self-etch adhesive systems.^{35, 36} The present result revealed that the DC of the strong self-etch adhesive APLP might be further compromised by an inappropriate application mode (without agitation). Therefore, as a method to improve DC of the strong self-etch adhesives within the interface area, active agitation is especially necessary. So that this “weak link” to dental substrate can be strengthen.

It is worth mentioning that although chemical reaction plays an important role in the DC difference between APLP-WA and APLP-WOA, other factors especially solvent evaporation induced by active agitation should have also contributed,³⁸ since solvent (especially water in APLP) likely impairs polymerization of the monomers within the demineralized substrates. For AEB, it contains both water and considerable amount of ethanol. Ethanol evaporates easily and meanwhile produces a desiccation effect within the hybrid layer, helping water to evaporate. Thus, solvent evaporation for AEB was faster and less dependent on agitation as compared to APLP. This might have also contributed a higher DC of AEB than APLP.

In summary, the present study has disclosed the role of acidic monomers/dental mineral chemical interaction in determining the curing performance of the self-etch adhesives. The chemical interaction with dentin is especially important for improving DC of the strong self-etching adhesive at the A/D interface. Agitation could benefit polymerization efficacy of the strong self-etch adhesive through enhancing the chemical interaction with tooth substrate. In addition, solvent evaporation and its influence on polymerization efficacy depending on the application mode of self-etch adhesives should be also considered.

Acknowledgments

This investigation was supported by Research Grants 5T32DE7294-15 and R15-DE021023 from the National Institute of Dental and Craniofacial Research, National Institutes of Health, Bethesda, MD 20892. The authors do not have a financial interest in the products, equipment, and companies cited in the manuscript.

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Figure 1. Representative scanning electron micrographs of the adhesive/dentin (A/D) interface for (A) Adper Easy Bond applied without agitation (AEB-WOA), and (B) Adper Easy Bond applied with agitation (AEB-WA).

Figure 2. Representative scanning electron micrographs of the adhesive/dentin (A/D) interface for (A, C) Adper Prompt L-Pop applied without agitation (APLP-WOA), and (B, D) Adper Prompt L-Pop applied with agitation (APLP-WA). (C, D) present an extended field of interest from which some overall features such as the debonding of APLP-WOA specimens could be clearly displayed.

Figure 3. Representative Raman spectra of dentin, Adper Easy Bond, and Adper Prompt L-Pop. The two adhesives were 10s-light-cured on glass.

Figure 4.

Representative micro-Raman mapping spectra acquired at 1 μm interval across the adhesive/dentin (A/D) interface for (A) Adper Easy Bond applied without agitation (AEB-WOA), and (B) Adper Easy Bond applied with agitation (AEB-WA). The spectra for AEB, interface, and dentin were identified using colors of green, red, and black, respectively.

Figure 5.

Representative micro-Raman mapping spectra acquired at 1 μm interval across the adhesive/dentin (A/D) interface for (A) Adper Prompt L-Pop applied without agitation (APLP-WOA), and (B) Adper Prompt L-Pop applied with agitation (APLP-WA). The spectra for APLP, interface, and dentin were identified using colors of green, red, and black, respectively.

Figure 6.

The degree of demineralization (DD) as a function of location for (A) Adper Easy Bond applied without and with agitation (AEB-WOA, AEB-WA), and (B) Adper Prompt L-Pop applied without and with agitation (APLP-WOA, APLP-WA). In each figure, means with different letters are significantly different ($P < 0.05$).

Figure 7.

Illumination of spectral subtraction in order to eliminate the spectral interference from the organic component of dentin at 1667 cm^{-1} on the C-C double bond at 1640 cm^{-1} . The employed spectrum was that of Adper Prompt L-Pop applied with agitation (APLP-WA) in the interface of the 1st micron.

Figure 8.

The degree of conversion (DC) as a function of location for (A) Adper Easy Bond applied without and with agitation (AEB-WOA, AEB-WA), and (B) Adper Prompt L-Pop applied without and with agitation (APLP-WOA, APLP-WA). In each figure, means with different letters are significantly different ($P < 0.05$).

Figure 9.

Representative Raman spectra in the wavenumber region of 900–1200 cm^{-1} for the Adper Prompt L-Pop applied without and with agitation (APLP-WOA, APLP-WA) in the interface (at the 1st micron), and in the adhesive layer at positions of 1th and 15th microns from the interface. The spectrum of the APLP cured on glass was also given for reference.

Table 1

Compositions of one-step self-etch adhesives used in this study.

Adhesive	Composition
Adper Easy Bond (AEB, 3M ESPE, Seefeld, Germany)	2-hydroxyethyl methacrylate (HEMA), bis- GMA, methacrylated phosphoric esters, 1,6 hexanediol dimethacrylate, polyalkenoic acid (Vitrebond copolymer), finely dispersed bonded silica filler, ethanol, water, initiators based on camphorquinone, stabilizers
Adper Prompt L-Pop (APLP, 3M ESPE, Seefeld, Germany)	Methacrylated phosphoric esters, bis-GMA, initiators based on camphorquinone, stabilizers, water, 2-Hydroxyethyl methacrylate (HEMA), polyalkenoic acid