MICRO SHEAR BOND STRENGTH OF UNIVERSAL DENTAL ADHESIVES TO DENTIN USING DIFFERENT ETCHING MODES (IN VITRO STUDY)

Samaa M. Morsy\textsuperscript{1+}\textit{BDS}, El Sayed Moustafa\textsuperscript{2} \textit{PhD}, Mahmoud Elsharkawy\textsuperscript{2} \textit{PhD}.

ABSTRACT

INTRODUCTION: Patient demand for tooth colored restorations have made composites an important part of the restorative process. An important factor affecting the intraoral performance of composite restorations is bonding. Therefore, a strong bond at the tooth-restoration interface is necessary for long-term success of a composite restoration.

OBJECTIVES: To compare the micro shear bond strength of a resin composite (Filtek Z250) to dentin using three universal adhesives in self-etching and total-etching modes and perform fractographic analysis using scanning electron microscope.

MATERIALS AND METHODS: 60 dentin specimens were used in this study. They were divided into two main groups (self-etch mode & total-etch mode). Each group was subdivided into three subgroups according to the bonding agent used (Tetric N bond universal, All bond universal and Single bond universal). Bonding of adhesives and composite buildups were done according to manufacturer’s instructions. All specimens were thermocycled for 500 cycles from 5 to 55°C. The micro shear bond strength was measured using universal testing machine. Specimens were further sectioned, gold sputtered and evaluated fractographically using scanning electron microscope (SEM). Data obtained were analyzed using F-test (ANOVA), two-way ANOVA and Spearman’s correlation tests.

RESULTS: In self-etch mode, the micro shear bond strength of the three tested adhesives didn’t show a significant difference. Meanwhile, etching significantly improved the micro shear bond strength of Single bond universal, while Tetric N bond universal and All bond universal showed no significant difference.

CONCLUSIONS: An etching step prior to the application of the universal adhesives didn’t significantly affect their micro shear bond strength, except for Single bond universal.

KEYWORDS: Adhesives, dentin, bonding agents, etch mode, micro shear bond strength, fractographic analysis.

INTRODUCTION

The goal in adhesive dentistry is to achieve an adequately strong bonding of the restorative resin to the tooth structure so that there is optimum retention, decreased microleakage and hence, superior color stability and clinical longevity of the restoration (1).

The main challenge for a dental adhesive is the ability to bond effectively to substrates of different nature. Adhesion to dentin poses a difficult challenge. This is partly due to the biological characteristics of dentin, namely its highly organic content, its tubular structure, and the presence of the dentin smear layer that forms immediately after cavity preparation (1, 2).

The preliminary use of phosphoric acid in total-etch adhesives increased the probability of clinical errors due to the need of rinsing and adequate management of dentin moisture (3). Contrary to the etch and rinse approach, self-etch adhesives do not remove but incorporate the smear layer in the hybrid layer and are capable of etching the tooth surface, while simultaneously preparing it for adhesion (4, 5). It is thought that the incidence of postoperative sensitivity appears to be lower in deep cavities when using self-etch relative to etch and rinse systems thanks to chemical bonding and reduced demineralization of dentin (6).

Manufacturers are constantly introducing new adhesive systems with claims of simplicity in use, improvement in their composition and ability to bond to tooth structure. Scientists and researchers feel the obligation to substantiate these claims. Previous studies have shown that bonding effectiveness of some adhesives appears dramatically low, whereas the bonds of other adhesives are more stable (7, 8). A new type of single step self-etch adhesives has been introduced. This type of self-etch adhesive is categorized as ‘universal’ or ‘multi-mode’ as they can be used either with the total-etch mode or the self-etch mode or as ‘selective’ enamel etching mode; self-etch on dentin and etch and rinse on enamel (9). Universal adhesives

\textsuperscript{1} Bachelor of Dentistry, Faculty of Dentistry, Alexandria University, Egypt.
\textsuperscript{2} Professor of Conservative Dentistry, Faculty of Dentistry, Alexandria University, Egypt.

\*Corresponding author
E-mail: samaone20@gmail.com

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are ‘universal’ in two main ways: First, they have an ability to bond to different types of dental materials without pre-treatment of the adherent surface, so they may be helpful for repair of resin composite restorations that involve different adherent substrates in the same restoration (10). Second, they are recommended by dental manufacturers for use both with and without acid pretreatment of tooth surfaces, can be used in total-etch mode with phosphoric acid without any negative effect on dentin bonding performance (11).

Because universal adhesives are marketed since a short time, relatively little information is available on their performance apart from those provided by the manufacturers, especially for the more recently introduced versions (12). Therefore the purpose of this study was to determine the dentin bond quality of universal adhesives in different application modes. The null hypothesis was that the tested universal adhesives would show no difference in bond strength when applied in different etching modes to dentin substrate.

**MATERIALS AND METHODS**

Eighteen extracted human permanent molars were selected for testing the micro shear bond strength of composite resin to dentin. The teeth were collected from the out-patient clinics at the Faculty of Dentistry, Oral Surgery Department, Alexandria University. The study was conducted after receiving the approval of the ethical committee at Faculty of Dentistry, Alexandria University, Egypt. Selected teeth were extracted due to periodontal reasons and were free of caries, attrition, abrasion, erosion, cracking or previous restoration and were selected from patients of age range 25-45. The teeth were thoroughly washed with running water, immersed in 0.5% solution of chloramine T for one week for sterilization (13), then polished with pumice rubber cups and stored in isotonic saline at room temperature which was changed weekly. The teeth were used within three months after extraction to avoid changes in dentin permeability which may affect bond strength testing (14).

The occlusal surface of the teeth were ground flat under running water by means of a diamond disk on a low speed motor with a cooling system to remove enamel and prepare flat superficial dentin surface. Then the dentin specimens were mounted vertically into a rubber mold with 14 mm internal diameter; filled with auto polymerizing acrylic resin, 2 mm below cement–enamel junction, with their occlusal surface facing upwards. After setting of the acrylic resin, the specimens were removed from the molds. The prepared specimens were assessed for absence of enamel and/or pulp tissue using stereomicroscope (OLYMPUS stereomicroscope sz11, Japan). All of these preparations were done by the same operator. (14, 15).

Sixty dentin specimens were randomly divided into two main groups: (Group A: self-etch mode and Group B: total-etch mode ) each consisting of 30 specimens which were then subdivided into three subgroups according to bonding agent used (Tetric N-bond universal, All bond universal and Single bond universal) with each subgroup containing 10 specimens; divided on three teeth. Three types of universal adhesives were applied in this study. The standard composition and manufacturer of these adhesives are shown in table 1.

**Table 1:** Composition and manufacturer’s instructions of adhesives and resin composite investigated in the present study.

| Tested materials and manufacturers | Composition | Self-etch mode | Total-etch mode |
|-----------------------------------|-------------|----------------|-----------------|
| N-Etch (Ivoclar Vivadent Inc., Schaan, Liechtenstein) | 37% phosphoric acid | Wash dentin for 5 s Dry with cotton pellet Rubbing the bond gently for 20 s Gentle air drying for 5 s Light cure for 10 s | Etch for 15 s Rinse for 5 s Drying with cotton pellets Apply adhesive as mentioned in the self-etch mode |
| Tetric n bond universal (Ivoclar Vivadent Inc., Schaan, Liechtenstein) | HEMA, MDP, Bis-GMA, D3MA, ethanol, water, highly dispersed silicon dioxide, CQ. | Wash dentin for 5 s Dry with cotton pellet Apply 2 coats of adhesive Rubbing the bond gently for 15 s Gentle air drying for 5 s Light cure for 10 s, No light cure between coats. | Etch for 15 s Rinse for 5 s Drying with cotton pellets leaving surface visibly moist Apply adhesive as mentioned in the self-etch mode |
| All bond universal (Bisco, Schaumburg, USA) | HEMA, MDP, Bis-GMA, Ethanol. | Wash dentin for 5 s Dry with cotton pellet Apply 2 coats of adhesive Rubbing the bond gently for 15 s Gentle air drying for 5 s Light cure for 10 s | Etch for 15 s Rinse for 5 s Drying with cotton pellets without over drying Apply adhesive as mentioned in the self-etch mode |
| Single bond universal (3M ESPE St. Paul, MN, USA) | HEMA, MDP, dimethacrylate resins, silane, initiators (CO), ethanol, water, filler | Wash dentin for 5 s Dry with cotton pellet Rubbing the bond gently for 20 s Gentle air drying for 5 s Light cure for 10 s | Etch for 15 s Rinse for 5 s Drying with cotton pellets |
| Filtek z 250 universal restorative (Ivoclar Vivadent Inc., Schaan, Liechtenstein) | UDMA and Bis-EMA (6) | One millimeter diameter plastic cylindrical tubes filled with resin composite molds were attached to the conditioned dentin surfaces and cured for 20 s | |

For the self-etch mode, the dentin bonding systems were applied directly following manufacturer’s instructions. But for the total-etch mode, the acid etching step was done before applying the dentin bonding systems. The resin composite was built up using a polyethylene tube (BioFlon IV cannula, India). The height of the tube was 2 mm and its inner and outer diameters were 0.9 and 1 mm respectively to allow the maximum number of specimens to be bonded to the same dentin substrate (16). All composite tubes were cured using the same LED curing unit at zero distance. According to manufacture instructions, each composite tube was cured for 20 seconds (Table 1). Tubes around the composite cylinders were removed by gently cutting each tube using a surgical scalpel blade no.11. Two up to four cylinders were placed on each molar; perpendicular to the prepared dentin surface (17).

After 1 week storage in artificial saliva at 37°C, the specimens were thermocycled 500 cycles between 5 °C and 55 °C with a dwell time of 5 seconds at each temperature to simulate the effect of thermal changes in the oral environment as recommended by the ISO TR 11450 standard in 2003 (18). Each acrylic-embedded tooth with the bonded composite micro cylinder was secured with tightening screws to the
lower fixed compartment of the universal testing machine (Instron model 3345, England) with a load cell of 500 Newtons. Data were recorded using computer software (Bluhiill 3; Instron). A loop prepared from an orthodontic wire (0.14 mm in diameter) was wrapped around the bonded micro cylinder assembly as close as possible to the base of the micro cylinder. The wire was aligned with the loading axis of the upper movable compartment of the testing machine. A shearing load was applied through the specimens. The shear load was applied at a cross-head speed of 0.5 mm/min until bonding failure occurs. The micro shear bond strength values (expressed in MPa) were calculated from the maximum load at failure divided by the bonded surface area (16).

The fracture load was recorded and the shear bond strength was calculated according to the following equation: \[ \sigma = \frac{F}{A}, \]
where: \( \sigma \) is the micro shear bond strength in Mega Pascals (MPa), \( F \) is the failure load in Newtons (N), \( A \) is the surface area in square millimeters (mm\(^2\)), where: \( A = \pi r^2 \) and \( r = 3.14 \times r = 0.9 \text{ mm} \).

After micro shear testing, the debonding sites of the fractured dentin specimens selected from all groups representing different modes of failure, were prepared for SEM; their roots were cut off up to the cemento-enamel junction, by a high speed motor with cooling system, to create dentin slices. Then the dentine slices were air-dried. They were not dehydrated using methods that involve passing the specimens through organic solvents, to avoid the possibility of extracting uncured monomers from the fractured surfaces. Completely air-dried specimens were secured to metallic stubs. They were sputter-coated with gold to reduce charging in electron beam of SEM upon scanning and then examined using a scanning electron microscope (JEOL JSM - 5300 operating, Japan) at 25 kV, at magnification from x100 up to x5,000 and showed different ultra-morphological characteristic features. The resin-dentin surfaces were photo-micrographed to be evaluated (19).

**Statistical analysis**

Data were fed to the computer and analyzed using Social Science Statistics, 2019. Descriptive statistics for shear bond strength in dentin were displayed as mean and standard deviation. Comparison of mean values of shear bond strength in dentin between the studied groups and the correlations between micro shear bond strength values and mode of failure were calculated. Statistical analysis was done using one-way (F-test), two-way ANOVA and Spearman’s correlation tests. Significance level was set at the 5% level. Bar chart was used for graphical presentation.

**RESULTS**

The mean shear bond strength of specimens in group A (self-etch) showed higher bond strength value for Tetric n bond universal ‘A1’ (16.53 ± 6.45 MPa) followed by Single bond universal ‘A3’ (14.82 ± 5.89 MPa) and All bond Universal ‘A2’ (12.83 ± 1.81 MPa). Although statistically, there was no significant difference between the three subgroups, as calculated using the F test of ANOVA.

As for the total-etch specimens in group B, the mean shear bond strength of Single bond universal ‘B3’ (26.94 ± 5.89 MPa) was significantly higher (\( P < 0.05 \)) than both Tetric n bond universal ‘B1’ (18.98 ± 8.22 MPa) and All bond Universal ‘B2’ (15.96 ± 4.18 MPa), with All bond Universal showing the lowest mean shear bond strength, as also calculated using the F test of ANOVA.

The mean shear bond strength of Tetric n-bond universal in total-etch mode (18.98 MPa) was slightly higher than in self-etch mode (16.53 MPa). Also, All bond universal in total-etch mode (15.96 MPa) was higher than in self-etch mode (12.83 MPa). Generally these differences were not significant. Meanwhile, the mean shear bond strength of Single bond universal in total-etch mode (26.94 MPa) was significantly higher (\( P < 0.05 \) and \( F = 16.25 \)) than in self-etch mode (14.82 MPa). These results were presented using bar chart (Figure 1).

A two-way analysis of variance (ANOVA) at \( P \leq 0.05 \) was used for analysis of the mean shear bond strength data. It revealed that the factor of adhesive system significantly influenced the mean shear bond strength values (\( P \leq 0.001 \)). Also, the factor of etching mode (total-etch vs. self-etch) significantly influenced the mean shear bond strength values (\( P \leq 0.001 \)). But the interaction between the two factors was not significant.

The overall failure mode percentages of the micro shear bond strength test were recorded after microscopic examination as 45% adhesive failures, 45% mixed failures and 10% cohesive failures.

The failure mode in the debonded specimens varied between subgroups, with the variation of the adhesive system used and the etching mode. These data are illustrated in (Table 2). The subgroups A1, A2 and A3 where acid etching was not used represented higher percentage of adhesive failure, than B1, B2 and B3 subgroups; in which acid etching was used. Meanwhile B1, B2 and B3 had higher percentage of mixed failure. Furthermore, cohesive failure represented the least percentage; occurred only in subgroups A3, B1, B2 and B3.

**Failure modes of the study subgroups.**

| Failure mode          | Adhesive System |
|-----------------------|-----------------|
|                       | A1       | A2       | A3       | B1       | B2       | B3       |
| Adhesive failure      | 50%      | 80%      | 30%      | 20%      | 60%      | 10%      |
| Cohesive failure      | 0%       | 0%       | 30%      | 10%      | 10%      | 10%      |
| Mixed failure         | 30%      | 20%      | 40%      | 10%      | 30%      | 80%      |

The Spearman’s rho correlation test results revealed a significant negative correlation between the mean shear bond strength and adhesive failure with (\( P = 0.20 \) and \( r_s = -0.7714 \)). While for the mixed failure, the results revealed a significant positive correlation with the mean shear bond strength with (\( P = 0.20 \) and \( r_s = 0.8143 \)). As for the cohesive failure, the results revealed a weak positive correlation with the mean shear bond strength with (\( P = 0.50 \) and \( r_s = 0.1857 \)).

The fractured composite-dentine specimens examined by SEM for fracture surface analysis are shown (Figures 2, 3).
Dentin when it is subjected to loading (2).

Etching modes achieve equivalent bonding performance to and there is limited information as to whether the different type of adhesive was only recently introduced to the market, without acid pretreatment of dentin surfaces. However, this is recommended by dental manufacturers for use both with and introduced for patient care. These bonding systems are categorized as "universal" or "multi-mode" has been recently  

Smear layer by using an etching agent, and a self-etching infiltration morphology, by generating thicker hybrid layers universal adhesives significantly improves the interface hybridization (i.e. resin tags and hybrid layer), has been proposed to improve the bond strength of universal adhesives. Phosphoric acid etching of dentine prior to application of universal adhesives significantly improves the interface infiltration morphology, by generating thicker hybrid layers and longer resin tags. Removal of the smear layer and smear plugs by this pretreatment facilitates the adhesive penetration, especially in mild universal adhesives (15).

Based on the above considerations, the purpose of this study was to evaluate the bonding ability of these three new commercially available bonding agents; Tetric N bond universal (ivoclar vivadent), All bond universal (Bisco) and Single bond universal (3MSPE) in self and total-etch modes. And analyze the fractured composite-dentin surfaces fractographically using SEM.

Recently, the micro shear bond strength test has been advocated as a modified method for evaluating the bonding ability of dentin-adhesive systems. Compared to the macro shear bond strength test, the micro shear bond strength test is more advantageous; it has fewer internal defects as well as more homogeneous stress distributions at the interface due to the use of smaller specimens. Furthermore, the small size of the tested specimen permitted several specimens to be bonded to the same dentin substrate; thereby promoting the conservation of extracted teeth. In addition, the micro shear bond strength test does not require an additional specimen trimming process after the bonding procedure as in micro tensile bond test, which conserves the integrity of the specimens and avoids pre-testing failures (17).

In the present study, the results revealed that etching significantly improved the micro shear bond strength of Single bond universal, while Tetric N bond universal and All bond universal showed no significant difference, this comes in consistent with a previous study by Takamizawa et al in 2016 (20). The composition of Single bond universal is similar to Tetric N bond universal and All bond universal; containing 10-MDP (10-Methacryloyloxydecyl dihydrogen phosphate) as a functional monomer. While the components in those adhesives are similar, there may be differences in the quantities and proportions of water, solvent, MDP and dimethacrylate resins among the adhesives. There is a possibility that such differences may influence viscosity and wettability of each bonding agent, affecting the ability of resin monomers to penetrate into demineralized dentin (21).

Self-etch adhesives partially demineralize dentine, leaving a substantial amount of hydroxyapatite crystals around the collagen fibrils. Yoshida et al (21) showed that an effective chemical interaction occurs between MDP and hydroxyapatite forming a stable Nano layer that could form a stronger phase at the adhesive interface, which increases the mechanical strength of the adhesive. In addition, stable MDP-Ca salt deposition along with Nano layering may explain the high bond stability, which has previously been proven by both laboratory and clinical research (22). According to the adhesion-decalcification concept, the MDP-Ca salt complex is highly insoluble, and stable, and forms strong molecular bonds to hydroxyapatite-based substrates (20).

Single bond universal is considered a mild self-etch adhesive. It has a pH of approximately 2.7 (16). In addition to 10-MDP in Single bond universal, it also contains methacrylate modified polyalkenoic acid copolymer; Vitrebond Copolymer (VBCP). Vitrebond copolymer bonds chemically and spontaneously to tooth substrates. Mitra et al reported a higher bond strength for adhesives containing Vitrebond copolymer than for those which did not contain it. It is likely that the presence of the polyalkenoic acid copolymer

Figure 1: Bar chart showing the comparison between the different subgroups in the self-etch and total-etch modes according to mean shear bond strength (MPa).

Figure 2 (I): SEM micrograph of one of the debonding sites showing fracture of the adhesive which exposed the underlying dentin. Failure probably started at the dentin-adhesive interface as a corner flaw and propagated through the adhesive, x100 (A: adhesive layer; D: dentin).

Figure 2 (II): SEM magnification at of the marked area in figure (I) showing adhesive partially covering the dentin. No trace of composite was found on the surface, x1000 (A: adhesive layer; D: dentin).

Figure 3 (I): SEM micrograph of the dentine side of a fracture beam showing different substrates. The arrow probably represents the area where the crack was initiated, x100.

Figure 3 (II): SEM of the marked area in figure (I) showing cohesive failure both in composite and dentin, x 1000 (D: dentin; C: composite resin).

DISCUSSION

A new type of single-step self-etch adhesive that is categorized as “universal” or “multi-mode” has been recently introduced for patient care. These bonding systems are recommended by dental manufacturers for use both with and without acid pretreatment of dentin surfaces. However, this type of adhesive was only recently introduced to the market, and there is limited information as to whether the different etching modes achieve equivalent bonding performance to dentin when it is subjected to loading (2).

An etch and rinse system is characterized by removing the smear layer by using an etching agent, and a self-etching system is characterized by eliminating the technique-sensitive rinsing step and reducing the operation time. On the other hand, micromechanical interlocking by means of good dentine hybridization (i.e. resin tags and hybrid layer), has been proposed to improve the bond strength of universal adhesives. Phosphoric acid etching of dentine prior to application of universal adhesives significantly improves the interface infiltration morphology, by generating thicker hybrid layers and longer resin tags. Removal of the smear layer and smear plugs by this pretreatment facilitates the adhesive penetration, especially in mild universal adhesives (15).

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favors additional bonding of universal adhesives to pre-etched dentin (23).

In this study, the results of micro shear bond strength of the three tested adhesives didn’t show a significant difference in self-etch mode. This may be due to that VBCP; in Single bond universal, in combination with MDP has shown contradictory results in the literature because this combination improved the bonding ability of Single bond universal in some studies, while in other studies was proven to be insignificant in comparison with other adhesives which didn’t contain this combination (4, 9, 21). The VBCP may compete with the MDP monomer for Ca-bonding sites located in hydroxyapatite and due to its high molecular weight, could even prevent VBCP approximation during polymerization. This comes in agreement with Wagner et al (9); who stated that there was not a statistically significant difference in mean micro bond strength when Single bond universal in self-etch mode was compared to All bond universal; which only contains MDP.

Also, Yoshida et al (21) hypothesized that VBCP may compete with the MDP present in Single bond universal. However, Muñoz et al (4) compared the longevity results of Single bond universal (contains both MDP-VBCP) with Adper single bond 2 (contains only VBCP), two materials with similar compositions, the only difference being the presence of MDP in the former, it seemed that the association MDP-VBCP enhanced the bonding ability, since Single bond universal in both etching modes showed stable bonds even after 6 months of water storage (4).

While there was some variation in the results of the present study, depending upon the adhesives and the etching mode, the total-etch mode groups of both Tetric n bond universal and All bond universal did not show substantially different micro shear bond strength values than the self-etch mode groups. Therefore, using universal self-etch adhesives in total-etch mode may have had no negative impact on dentin bonding. Similar results have been found by Takamizawa et al (20). And also Wagner et al (9) who stated that similar bond strength values were observed for the universal adhesives regardless of application mode, which makes them reliable for working under different clinical conditions.

Thus, the hypothesis was refused for Single bond universal only; which demonstrated an improvement in the dentin bond strength with prior phosphoric acid etching, compared to the other two universal adhesives evaluated. The two other adhesive systems tested had the same versatility of being used in both the etch and rinse and self-etch approaches; the differences in their compositions have been the reason for their different performances in bond strength, as evaluated by this study and other in vitro studies (22). This conclusion comes in agreement with Rosa et al (22), Wagner et al (9) and Takamizawa et al (20) who stated that using universal self-etch adhesives in total-etch mode may have no negative impact on dentin bonding. And the universal adhesives might be used for bonding resin composite to dentin with either the etch and rinse or the self-etch approach in clinical situations.

Various patterns of failure were observed among the tested subgroups. The bonding systems subgroups where etching agent was not used, showed a large percentage of adhesive failure between the dentin and bonding resin combined with a small percentage of the more complicated mixed failure in most specimens. When the dentin surface was treated with the etching agent, the complex mixed failure pattern was predominant in most specimens; with a small percentage of adhesive failure. Meanwhile cohesive failure was only observed in six out of the sixty specimens used in this study.

Overall, both adhesive and mixed fracture rates were higher than cohesive failures in resin composite, which could be attributed to the good bonding properties of the adhesives used in this study, and the adhesives’ ability to resist flaw propagation, such as crack growth or peeling resistance from the substrate. For the shear bond test, if dentin pull-out was observed in the failure surface, then the calculated nominal bond strength was no longer based on the cross sectional area. Thus, the bond test could not discriminate between good and very good bonding agents (24). These results were similar to those of the studies conducted by De Munck et al (25), Martinez et al (26) and Muñoz et al (27). The results of the study conducted by Firat et al (28) were inconsistent with those of this study because cohesive and mixed failure patterns were predominantly observed in the acid-etched groups, indicating that the adhesive interface was preserved.

Microscopic observations also revealed that higher bond strength values were generally associated with mixed failures and lower bond strength values with adhesive failures. This was especially true for All bond universal adhesive bonded to non-etched dentin, which revealed 80% adhesive failures while also showing the lowest bond strength of all groups (12.83 ± 1.81 MPa), meanwhile Single bond universal bonded to etched dentin, which showed 80% mixed failures while also showing the highest bond strength of all groups (26.94 ± 5.89 MPa). These results were similar to that of the study conducted by Sabatini (29) who also stated that mixed failures were associated with highest bond strength values while adhesive failure modes were associated with lowest bond strength values.

Fracture surface analysis (fractography) is well established as a mean of failure analysis. It has been recognized as a powerful analytical tool in dentistry. The application of fractography is based on the principle that the entire history of the fracture process is encoded on the fracture surface of tested materials. Fractography has been used to relate the stress at failure, the nature of the stress state, and the sizes of the initial crack and surrounding topography. It shows that there are characteristic markings on the surfaces that are implying the fractal analysis. In addition, the microstructure characterization is necessary to analyze relevant mechanical properties and to support further arguments on fracture and bonding phenomena. It is difficult to discuss materials behavior without proper material characterization, which should be the first step of any research proposal involving materials (30).

Mechanical failure occurs when the applied stress becomes greater than the strength of the material. In fact, forces propagating from the application point can be expected to run along the path of least resistance, that is, along the dentinal tubules, as in fig. 3; where the crack probably initiated at a weak point on the border of the composite-dentin complex, and propagated leading to cohesive failure both in composite and dentin. As for fig. 2; failure probably started at the dentin-adhesive interface as a corner flaw and propagated through the adhesive (31, 32).So, the null hypothesis was rejected because each different etching mode created different weak regions at the interface between resin composite and adhesive layer or between adhesive layer and decalcified dentin; which varied according to the universal adhesive used.
CONCLUSION
Noting the limitations of our study, the following conclusions can be drawn:
1. An etching step prior to the application of the universal adhesives, did not significantly affect their micro shear bond strength, except for Single bond universal.
2. In self-etch mode, there was not a significant difference between the micro shear bond strengths of the three tested adhesives.
3. Fractographic principles should be applied for the analyses of fractured surfaces, improving the understanding of the fracture phenomenon, which is, at the end, the most common failure cause of dental restorations.

CONFLICT OF INTEREST
The authors declare that they have no conflicts of interest.

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