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# EFFECT OF BROMELAIN ENZYME VERSUS SODIUM HYPOCHLORITE ON SHEAR BOND STRENGTH OF RESIN COMPOSITE TO DENTIN USING TWO ADHESIVE SYSTEMS

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## *ABSTRACT*

**Background:** Proper bond strength of composite restoration contributes to endurance and survival; thus, dentin surface preparation for increased bond strengths is required.

**Objectives:** This study is designed to assess the effect of bromelain enzyme and Sodium hypochlorite (NaOCl) on shear bond strength of resin composite to dentin using two adhesive systems; etch and rinse and self-etch.

**Subjects and methods:** Sixty complete (caries-free) maxillary and mandibular premolar teeth from patients aged (25-45) were used in this in-vitro experimental study. It had to be removed for orthodontic purposes. The prepared premolars were allocated into two main groups of 30 specimens according to the pretreatment agent and the bonding system used. The pretreatment agent used for dentin was divided into 3 subgroups of 10 each. specimens each Bromelain (A1), sodium hypo chlorite (A2) and control group (A3). The type of bonding system used; in the first subgroup is the self-etch bonding system (B1). The second subgroup was the total-etch bonding system (B2). Both shear bond strength (SBS), micro SBS, ultra morphology.

**Results:** Regarding the control group, the self-etch system (12.88±1.15) had a significantly higher value than the total-etch system (8.98±0.01) (p<0.001). Respecting NaOCl (10%), the total-etch system (24.58±2.31) had a significantly higher value than the self-etch system (15.46±2.30) (p<0.001). Concerning Bromelain, the total-etch system (27.50±2.31) had a significantly higher value than the self-etch system (18.38±2.41) (p<0.001).

**Conclusion:** The prior treatment of premolar dentin with NaOCl and bromelain enzyme results in composite resins with similar SBS. In other words, after acid etching, bromelain, a safe solution with no side effects, may effectively dissolve the collagen network as effectively as NaOCl.

**KEYWORDS:** Bromelain, shear bond strength, bonding system.

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## INTRODUCTION

Adhesion was first used in dentistry in 1955 by Buonocore. Each subsequent generation of adhesive was substituted by the newer, which addressed the problems of the previous generation as well as the practitioners' preferences. Self-Etch, total-etch adhesives, and resin-modified approach are the adhesives now in use because they have special tooth-self-adhesion capabilities. Fusayama first proposed the idea of total etching or the etch and rinse method in the early 1980s [1].

A paradigm shift in dentistry practice can be seen in the establishment of a reliable bonding to enamel and dentin substrates along with various bonding agents. Dentists may frequently place direct and indirect restorations because they can attach to both enamel and dentin substrates in a fashion that is generally predictable [2]

Dental materials that attach to dental tissues as well as possible are constantly being developed as part of restorative dentistry. As a result, there is growing interest in the study of dental adhesives. The chemical properties of an adhesive system and the therapeutic techniques that must be used in conjunction with clinical usage to be effective are both crucial components of an adhesive system [3]

Modern restorative dentistry has been inspired by advancements in dentin bonding technologies. The design of a conservative cavity, which essentially depends on the success of existing dentin bonding technologies, has been produced as a result of the need for the least invasiveness. Etch-and-rinse and self-etch are two systems that define how adhesives interact with enamel and dentin. Acidic functional monomers are used in self-etch systems to impregnate and demineralize tooth structures at the same time. The benefits and drawbacks of etch-and-rinse and self-etch systems are mostly connected to the streamlined bonding processes necessary in healthcare settings. When choosing the best adhesive materials for clinical usage, understanding the composition, properties, and mechanisms of adhesion for each adhesive system is essential [4]

This study is designed to evaluate the effect of bromelain and NaOCl on SBS of resin composite to dentin using two adhesive systems; etch and rinse and self-etch.

## SUBJECTS AND METHODS

### Teeth selection:

Sixty complete (caries-free) maxillary and mandibular premolar teeth from patients aged (25-45) were used in this in-vitro experimental study. It had to be removed for orthodontic purposes. To prevent the development of cracks, the safest way of extraction is acquired by maintaining the alveolar bone structure by slipping with luxation until the tooth has free vertical mobility at which time it has been picked up with tweezers. Teeth surfaces were scaled and polished using a rubber cup and pumice paste right after extraction. Using a diamond disk and water cooling, it was possible to section the crowns at a depth of 2 mm from the dentin enamel junction (DEJ), parallel to the occlusal surface.

### Ethical regulations:

Before having their teeth extracted for research purposes, participants were informed of the study's goal and given an illustration of it by one or more members of the surgical department at the Minia Dental Hospital. The undamaged (caries-free) maxillary premolar teeth that had been extracted for orthodontic treatment served as the subject of this in-vitro experimental study. All of the teeth used in the study were safely destroyed after use. The ethical committee for the Faculty of Dentistry at Minia University gave its approval to this investigation.

### Specimen preparation

The tooth was Placed into a saline solution for 7 days before the experiment. Acrylic blocks were prepared at 20 mm diameter using a 20 cm plastic disposable syringe. The 20 cm syringe was cut by the end leaving a hollow opened piston area. The inner syringe area was lubricated by separating the medium and the cold cure acrylic resin was mixed following manufacturer instructions. Piston of the syringe is aspirated at the 10 mm area.

Acrylic resin was packed in the syringe in the doughy stage and the root area of each tooth is immersed in self-cure acrylic resin same occlusal level until the acrylic resin setting reaction occurs tooth inside the cylindrical syringe block, The teeth are left fixed in their position until the acrylic resin set, then removed by pressing the piston to extract the acrylic specimen and then stored in distilled water to avoid dehydration of the teeth until the next step.

**Grouping of the specimens :**

The prepared premolars were allocated into two main groups of 30 specimens according to the pretreatment agent and the bonding system used. The specimens were divided into 3 subgroups of 10 according to the pretreatment agent used specimens each; Bromelain (A1), sodium hypo chlorite (A2) and control group (A3). The type of bonding system used; in the first subgroup was, the self-etch bonding system (B1). The second subgroup was the total-etch bonding system (B2).

**Sample etching technique :**

Teeth in Group 1 were etched with phosphoric acid gel (37%). Teeth in Group 2 were etched and deproteinized with NaOCl (10%). Teeth in Group 3 were etched and deproteinized using bromelain (8%). Group 4 teeth were carved using a self-etch mechanism in universal adhesive. Group 5 teeth were deproteinized with NaOCl (10%). Group 6 was deproteinized with bromelain (8%). The samples were stored for a week at room temperature prior to SBS [5].

**Application of the adhesive:**

3m per Single bond and 3m universal single bond were used as self-etch and total-etch systems.

Bonding agent were used according to manufacturer instructions . Each sample bonding agent was applied by micro brush for one layer . Additional layer was applied after air thinning by gentle air pressure then light cured for 20 seconds .

**Light curing :**

Fanta light cure system was used with the following specifications : 35,000AMD, Wavelength: 358nm-515nm, Turbo mode: light intensity 2300mw/cm² – 2500mw/cm², Normal mode: illumination 1000mw/cm² – 1200mw/cm², Power input: 100-240VAC 50Hz/60Hz, Power output: DC 5V/1A, Battery; 2600mA.

**Microshear bond Strength test:**

These tests were carried out using Instron®'s Bluehill Lite Software. Each tooth with its own bonded micro-cylinders was mounted into the centre hole of a specially designed sample holder jig, which was subsequently mounted horizontally with tightening screws to the lower fixed section of a materials testing machine. The shearing test was carried out using a chisel-shaped mono-bevelled metallic rod mounted on the testing device's upper movable parts and moving at a cross-head speed of 0.5 mm/min. The needed force to cause debonding was determined in Newtons.

## RESULTS

Descriptive statistics for SBS (MPa) values in different groups were presented in Table (1).

There was a significant interaction between both tested variables (pretreatment agent and bonding system) (Table 2).

For self-etch, there was a marked variance between groups ($p<0.001$). Bromelain had the highest value (18.38±2.41), followed by NaOCl 10% (15.46±2.30), then the control group (12.88±1.15), while the lowest value was found in NaOCl 5.25% (9.60±1.10). Bromelain had remarkably higher values than other groups ($p<0.001$). The control

TABLE (1) Descriptive statistics for SBS (MPa) values in different groups:

| Pretreatment agent | Bonding system | Mean | 95% confidence interval | | SD | Min | Max |
|---|---|---|---|---|---|---|---|
| | | | Lower | Upper | | | |
| **Control** | **Self etch** | 12.88 | 12.03 | 13.73 | 1.15 | 11.08 | 14.74 |
| | **Total etch** | 8.98 | 8.97 | 8.99 | 0.01 | 8.96 | 8.99 |
| **NaOCl 5.25%** | **Self etch** | 9.60 | 8.79 | 10.42 | 1.10 | 7.78 | 11.14 |
| | **Total etch** | 18.12 | 16.97 | 19.27 | 1.55 | 16.18 | 20.81 |
| **NaOCl 10%** | **Self etch** | 15.46 | 13.75 | 17.16 | 2.30 | 12.49 | 18.48 |
| | **Total etch** | 24.58 | 22.87 | 26.29 | 2.31 | 20.91 | 27.61 |
| **Bromelain** | **Self etch** | 18.38 | 16.60 | 20.17 | 2.41 | 15.15 | 21.73 |
| | **Total etch** | 27.50 | 25.79 | 29.21 | 2.31 | 24.08 | 30.44 |

TABLE (2) Effect of different variables and their interactions on SBS (MPa):

| Source | Sum of squares | df | Mean square | f-value | p-value |
|---|---|---|---|---|---|
| **Pretreatment agent** | 1275.31 | 3 | 425.10 | 127.37 | **<0.001*** |
| **Bonding system** | 457.25 | 1 | 457.25 | 137.01 | **<0.001*** |
| **Pretreatment agent * Bonding system** | 432.69 | 3 | 144.23 | 43.22 | **<0.001*** |

***df=degree of freedom*; significant (p ≤ 0.05) ns; non-significant (p>0.05)***

group and NaOCl 10% have notably higher values than NaOCl 5.25%. There was a marked variation between groups (p<0.001). Concerning Total-etch, Bromelain had the highest value (27.50±2.31), followed by NaOCl 10% (24.58±2.31), then NaOCl 5.25% (18.12±1.55), while the control group had the lowest value (8.98±0.01). All post hoc pairwise comparisons were remarkable (Table 3).

Regarding the control group, the self-etch system (12.88±1.15) had a remarkably higher value than the total-etch system (8.98±0.01) (p<0.001). Respecting NaOCl (10%), the total-etch system (24.58±2.31) had a remarkably higher value than the self-etch system (15.46±2.30) (p<0.001). Concerning Bromelain, the total-etch system (27.50±2.31) had a remarkably higher value than the self-etch system (18.38±2.41) (p<0.001) (Table 4).

## Statistical Analysis

For numerical data, the mean and standard deviation were displayed. The Shapiro-Wilk test was done to verify if they were normal. The data, which had a parametric distribution, was evaluated using two-way ANOVA. Following that, Tukey's post hoc analysis was performed. Simple effect comparisons were performed using the pooled error term from the primary ANOVA model and the Bonferroni correction. The significance level was set at p 0.05 for all tests. The statistical study was carried out using the R statistical analysis program for Windows, version 4.1.3.

TABLE (3) Mean of SBS (MPa) for different pretreatment agents within each bonding system:

| Bonding system | SBS (MPa) (mean±SD) | | | | p-value |
|---|---|---|---|---|---|
| | Control | NaOCl 5.25% | NaOCl 10% | Bromelain | |
| **Self etch** | 12.88±1.15$^{B}$ | 9.60±1.10$^{C}$ | 15.46±2.30$^{B}$ | 18.38±2.41$^{A}$ | **<0.001*** |
| **Total etch** | 8.98±0.01$^{D}$ | 18.12±1.55$^{C}$ | 24.58±2.31$^{B}$ | 27.50±2.31$^{A}$ | **<0.001*** |

***Values with different superscript letters within the same horizontal row are significantly different *; significant (p ≤ 0.05) ns; non-significant (p>0.05)***

TABLE (4) Mean of SBS (MPa) for different bonding systems within each pretreatment agent:

| Pretreatment agent | SBS (MPa) (mean±SD) | | p-value |
|---|---|---|---|
| | Self etch | Total etch | |
| **Control** | 12.88±1.15 | 8.98±0.01 | **<0.001*** |
| **NaOCl 5.25%** | 9.60±1.10 | 18.12±1.55 | **<0.001*** |
| **NaOCl 10%** | 15.46±2.30 | 24.58±2.31 | **<0.001*** |
| **Bromelain** | 18.38±2.41 | 27.50±2.31 | **<0.001*** |

****; significant (p ≤ 0.05) ns; non-significant (p>0.05)***

## DISCUSSION

The crucial component for adhesion is the close connection that forms between the adhesive and the substrate. Adhesion to enamel is quite simple, but adhesion to dentin is a challenging task. Dentin's biological characteristics, such as its tubular shape, its linear structure, and its dentin smear layer, all contribute to its difficulty in adhesion [6].

Natural teeth were used in this study. Concerning tooth design and morphology, this option was closer to the clinical circumstances. To reduce potential differences and errors, teeth of average sizes and shapes were chosen before testing. Premolars were utilized because of their small size and restricted surface area for bonding. The premolars that were gathered included both maxillary and mandibular premolars, as well as first and second premolars. To reduce the biases brought on by the variation in mineralization and morphology of the teeth, it would have been desirable to collect only one type of tooth, however, this was nearly impossible due to the challenges faced during specimen collection [7].

In this study, self etch control group had better micro SBS than the total etch because of the use of 10-MDP and it as the acidic monomer of the self-etch system which altered the smear layer rather than removing the 37% phosphoric acid in the total etch. Self-etch adhesives are recommended for direct composite resin restorations, especially when dentin is the primary support material because they provide a greater and more consistent bond strength to dentin [9].

It was worthy to mention that the Depth was confined in this study below the DEJ by 1 mm. Too deep dentin layers give weaker bonding than superficial dentin due to the widening of dentinal tubules and the more complex collagen fibres [10]. According to reports, bond strength is significantly influenced by dentin depth, and as dentin depth rises, bond strength declines [15].

NaOCL is a significant proteolytic compound for removing organic compounds. It is thought that the proteolytic action of NaOCL causes significant degradation of complicated peptide chains. With exposed hydroxyapatite crystals, NaOCL-treated dentin benefits and over time produces a more stable interface. However , There are numerous drawbacks of deproteinizing acid-etched dentin using NaOCl. It creates a zone of fragility, is cytotoxic, and has a foul taste and odour. The adverse consequences of NaOCl increase as dentinal depth grows. Due to these drawbacks, efforts are being made to find more effective ways to deproteinize dentin [11].

In this study, two concentrations of sodium hypochlorite were used 5.25% and 10% to investigate the concentration effect on bond strength. sodium hypochlorite in a concentration of 10 % produced micro SBS higher than the 5.25% concentration .

This study investigated and compared the Micro SBS. Cohesive failures and overestimations of bond strengths were the results of macro-bond strength testing. Micro-bond strength tests were introduced to lessen the defects. The majority of bond-strength tests are conducted using them [14].

Micro Shear bond tests were used because of their simple procedures and minimal equipment as well as specimen preparation requirements. The use of the bromelain enzyme as a dentin treatment, before the use of total-etch and self-etch adhesive systems, had an impact on the composite's ability to form a strong micro shear bond to superficial dentin. The use of adhesive systems and the interplay of bonding systems and dentin treatments had a substantial impact on the bond strength results in this investigation.

This study agrees with Kochhar et al., that after acid etching, the elimination of collagen fibre with the bromelain enzyme improves SBS and strengthens the binding between composite resin and tooth structure [16].

This study agrees with Chauhan et al., that bond strength is increased when unsupported collagen is removed with the bromelain following acid etching. Application of the bromelain enzyme had a substantial impact on the results for bond strength. No remarkable differences between the control and the NaOCl could be found. The group treated with the bromelain enzyme displayed the maximum bond strength [8].

Moreover This study agrees with Sharma et al., that application of bromelain enzymes had a substantial impact on the bond strength result. SBS of NaOCl (5 and 10%) and in the bromelain were found to differ noticeably. Group II (sodium hypochlorite 5% and 10%) and Group I both had worse results than the bromelain group [11].

## CONCLUSION

The prior treatment of premolar dentin with bromelain and NaOCl results in composite resins with similar SBS. In other words, after acid etching, bromelain is a safe solution with no side effects, and may effectively dissolve the collagen network as effectively as NaOCl.

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