The Effect Of Silver Diamine Fluoride On Bond Strength of Universal Adhesive To Demineralized Dentin

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Abstract

Aim: To evaluate the effect of silver diamine fluoride (SDF) on bond strength of universal adhesive to Sound and Demineralized dentin.

Materials and Methods: The 80 samples are divided into two main groups (40 teeth for each) according to the presence of the intervention (SDF); each main group is subdivided into two subgroups according to the substrate mineralization then moulded into self-cured acrylic resin with occlusal surface facing upward. The occlusal enamel is removed using wheel diamond stone under running water. Samples were wet ground against 600,800,1200 grit SC paper to obtain smooth occlusal surface. Half of the samples were immersed into lab made demineralizing solution to obtain dentin surface demineralization, then divided into 4 groups (n = 20), Group 1: Sound Dentin without SDF, Group 2: Demineralized Dentin without SDF, Group 3: Sound Dentin with SDF, Group 4: Demineralized Dentin with SDF. Microtensile bond strength was measured using Universal testing machine. Means and Standard deviation for each group were calculated, data were statistically analysed by Two-way ANOVA and Tukey Post Hoc test.

Results: Results revealed that Group 1 and Group 3 have highest micro-tensile bond strength values with no statistically significant difference between them. Group 1 showed the highest statistically significant mean micro-tensile strength values than Group 2. Group 2 showed no statistically significant difference in values than Group 3. The least statistically significant micro-tensile strength value was recorded for Group 4. The different surface treatment used have statistically significant effect on the micro-tensile bond strength (P<0.001). In addition, it showed that there is a statistically significant effect of the dentin substrate mineralization, on the micro-tensile bond strength (P<0.001). The interaction between surface treatment and substrate was also statistically significant (P<0.005).

Conclusion: Contamination of sound dentin surface by SDF does not affect bonding in case of using universal adhesive in etch and rinse mode. Treatment of caries affected dentin by SDF negatively affect bond strength in case of using universal adhesive in etch and rinse mode.

Keywords: SDF, Dentin Remineralization, Silver Diamine Fluoride, Microtensile Bond Strength, Caries Affected Dentin

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Introduction
Dental caries is a complex bacterial driven, multifactorial, dynamic disease that results from the imbalance between the tooth minerals and the plaque biofilm; that is, when the pH drop causes mineral loss over time from dental hard tissues.1 Traditionally, dental practitioners have been trained to manage cavitated lesions through a surgical approach where caries removal has been performed till reaching hard, dry dentine. In recent years, with an improved understanding of disease pathology and the need to decrease the risk of pulp exposure, more conservative approaches such as partial caries excavation has been developed, and complete removal of all carious dentin is no longer considered as the standard strategy.3 Silver diamine fluoride [Ag (NH3)2F] (SDF) is a colourless alkaline solution containing 25% silver, 5% fluoride, 8% amine and 62% water (AgNH2 F) and is the most concentrated fluoride product (44,800ppm) commercially available for caries management.4 and has proven to be effective in management of dental caries. 5 SDF is a non-invasive, simple, painless and low-cost approach, moving dentistry toward more frequent non-surgical management to arrest dental caries, lower rates of surgical care, tooth loss, and increase tooth-restoration cycle.4 Although the SDF solutions are generally regarded as safe and effective, the effect of its application on the dentin bond strength to direct restorative materials, is uncertain and evidence for the influence of previous application of 38% SDF on the bond strength value of direct restorative materials to sound and caries affected dentine is still needed.

MATERIALS AND METHODS
A total of 40 natural human molars (calculated using G power sample size determination). Teeth were collected from the department of Oral and Maxillo-facial surgery, faculty of dentistry, Ain Shams University, under the roles of the Ethical Committee of Faculty of dentistry, Ain Shams University. The collected teeth were placed in 6% sodium hypochlorite solution for 24 hours to remove all soft tissues, staining and plaque. The occlusal enamel is removed using diamond wheel stone with copious water irrigation. Each sample will be moulded into an acrylic block. Using custom made mould (2 cm diameter, 2.5 cm high), separating medium for inner surface and then, self-cured acrylic resin (Acrostone, Egypt) was mixed and poured into the mould till it became flushed with the upper rim of the mould, with occlusal surface facing upward and extended 1 ml above and leave them to set. Teeth were wet ground using silicon carbide paper grits #600, #800 and #1200 microns respectively till occlusal surface became flat with no irregularities. Using digital ultrasonic cleaner (CODYSON Ultrasonic cleaner CD-4860, Shenzhen Codyson co, Ltd. China) (5 minutes/400C ) remove any debris 6. Half of the specimens (n=40) will be separately submitted to immersion in acidic buffer solution of pH. 4.4 (2.2 mM CaCl, 2.2 mM NaH2PO4 , 0.05 mM Acetic Acid), each in a separate container at room temperature. Half of the specimen (n=40, 20 is totally sound and 20 is demineralized) will be properly air drying using triple way syringe. Silver diamine fluoride will be applied to dentin dried surface (one coat only) active application (10 sec) using micro-brush, leave it for 1 minute to insure its penetration. Then gentle air dryness (5 sec) followed by rinsing (15 sec) then gentle air dryness (5 sec) and leave 10 minutes before adhesive application. For all samples, 32% phosphoric acid etching application for 15 seconds without agitation and rinsed with water for 30 seconds. The excess water was blot dried using tissue paper to maintain a moist dentine surface. The surface was properly air drying, then 10 sec active application of single coat of single bond universal adhesive followed by 5 sec gentle air drying to remove excess adhesive and solvent, giving uniform thickness for adhesive layer. Then light curing for adhesive for 20 second using (3M Elipar LED curing light device intensity 1200 mW/cm2). Filtek z250 XT composite disc (3M- ESPE, USA) is prepared with 4 mm thickness then light curing for 20 second. Teeth fixed in acrylic resin blocks were then mounted in an automated diamond saw (Isomet 4000, Buehler Ltd., Lake Bluff, IL, USA), which was used for all sectioning procedures in this study. Occlusal surfaces
were flattened to the level of the dentino-enamel junction under copious water coolant (Cool 2 water-soluble anticorrosive cooling lubricant, Buehler Ltd., Lake Bluff, IL, USA), with a concentration of 1:33, lubricant: water.

The objective of longitudinal sectioning of restored teeth was to obtain composite-dentin beams of (0.9 mm x 0.9 mm) in area. Each beam was composed of composite and dentin with adhesive at the interface. Geraldeli’s jig was used to mount beams onto the universal testing machine (Instron, MA, USA). Each beam was aligned in the central groove of the jig and glued in place by its ends using cyanoacrylate based glue (Zapit, DVA Inc, USA). The jig was in turn mounted into the universal testing machine (Instron, MA, USA) with a load cell of 500 N. Tensile load was applied, at a cross-head speed of 0.5 mm/min, until bonding failure of the specimen occurred. Bond strength was calculated in MegaPascal (Bluehill Lite software, Instron, MA, USA). Specimen fragments were carefully removed from the jig with a scalpel and stored in their corresponding labelled plastic cones until examination of failure mode. The force required for failure (Newton) was divided by the surface area (mm²) to calculate the microtensile bond strength in MPa. Statistical analysis was performed using SPSS statistical analysis program for Windows (version 21.0). One-Way ANOVA and Tukey HSD post-hoc test was used after the evaluation of normal distribution of the collected data using Kolmogorov-Smirnov and Shapiro-Wilk tests and for homogeneity of variance using Levene statistic. Statistical power was determined using GPower program for Windows. The significant level was set at P=0.05. Data was collected and subjected to two-way analysis of variance (ANOVA) at P ≤ 0.05 was used to test the effect of the SDF surface treatment, the Dentin substrate mineralization and the interaction between them on the microtensile bond strength of universal adhesive to dentin

### Table 1 Materials Ingredients & specifications

| Material                                    | Composition                                                                 |
|---------------------------------------------|-----------------------------------------------------------------------------|
| **Demineralizing solution**                 | 2.2 mM CaCl , 2.2 mM NaH₂PO₄ , 0.05 mM Acetic Acid-pH 4.4                  |
| **Silver Diammine Fluoride (SDF)**          | (AgNH₂F) solution contains 25% silver 62% water , 5% fluoride, 8% amine-PH = 10 |
| **Single Bond Universal**                   | MDP: dimethacrylate resins, HEMA, Vitrebond copolymer, filler, ethanol, copolymer, water, initiators, silane-pH: 2.7 |
| **Filtek™ Z250 XT, 3M™ ESPE™**             | Surface-modified zirconia/silica with a median particle size of approximately 3 microns or less. Non-agglomerated / non aggregated 20nm surface-modified silica particles. Filler loading of 82% by wt (68% by volume). The resin system: Bis-GMA*, UDMA*, Bis-EMA*, PEGDMA* and TEGDMA*. |
| **Scotchbond Etchant**                      | 32% phosphoric acid pH: 0.5                                                  |

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RESULTS
Data was collected and subjected to two-way analysis of variance (ANOVA) at $P \leq 0.05$ was used to test the effect of the SDF surface treatment, the dentin substrate mineralization and the interaction between them on the microtensile bond strength of universal adhesive to dentin (Table 2). It revealed that different surface treatment used have statistically significant effect on the microtensile bond strength ($P<0.001$). In addition, it showed that there is a statistically significant effect of the dentin substrate mineralization, on the micro-tensile bond strength ($P<0.001$). The interaction between surface treatment and substrate was also statistically significant ($P<0.005$).

The results of One-Way ANOVA followed by Tukey’s HSD Post Hoc Test were shown in (Table 3). Results revealed that that Group 1(sound dentin without SDF) and Group 3(sound dentin treated with SDF) have highest micro-tensile bond strength values with no statistically significant difference between them. Group 1(sound dentin without SDF) showed the highest statistically significant mean micro-tensile stress values than Group 2(demineralized dentin treated with SDF). Group 2(demineralized dentin treated with SDF) showed no statistically significant difference in values than Group 3 (sound dentin treated with SDF). The least statistically significant micro-tensile stress value was recorded for Group 4 (demineralized dentin treated with SDF).

Table (2) : Two-way ANOVA of all tested variables for the effect of the SDF surface treatment, the dentin substrate mineralization and the interaction between them on the micro-tensile bond strength values

| Source                | Type III Sum of Squares | Degrees of Freedom | Mean Squares | F    | Sig    | Partial Eta Squared |
|-----------------------|-------------------------|--------------------|--------------|------|--------|--------------------|
| Corrected Model       | 2112.801**              | 3                  | 704.933      | 58.46| 0.001  | 0.60               |
| Intercept             | 3549.375                | 1                  | 3549.375     | 1722.364 | 0.001  | 0.78               |
| Treatment             | 643.940                 | 1                  | 643.940      | 33.362 | 0.000** | 0.03               |
| Substrate mineralization | 1710.711              | 1                  | 1710.711     | 88.475 | 0.000** | 0.06               |
| Treatment * Substrate mineralization | 150.00     | 1                  | 150.020      | 0.375 | 0.561  | 0.01               |
| Error                 | 1677.690                | 60                 | 28.0096      |      |        |                    |
| Total                 | 60760.348               | 69                 | 913.997      |      |        |                    |
| Corrected Total       | 60760.348               | 69                 | 913.997      |      |        |                    |

P-Value < 0.05 is statistically significant (*)

Table (3): Means ± Standard Deviation for the effect of SDF application on micro-tensile bond strength (MPa) to sound and demineralized dentin.

| Groups                  | Sound dentin without SDF | Demineralized dentin without SDF | Sounded dentin with SDF | Demineralized dentin with SDF |
|-------------------------|--------------------------|----------------------------------|-------------------------|-------------------------------|
| Micro-tensile Bondstrength | 26.57 ± 5.2              | 20.13 ± 4.9                      | 23.72 ± 3.6             | 11.61 ± 3.3                  |

Means with same superscript letters, indicates not statistically significant difference at P ≤ 0.05

Figure (1): bar chart representing the mean and standard deviation in MPa of all tested groups
DISCUSSION
The contemporary caries management philosophy has changed from the traditional surgical approach to a medical model, which often includes the use of fluoride therapy (Chu et al. 2010). Among the fluoride agents, silver diamine fluoride (SDF). SDF will provide a new caries management approach, moving dentistry toward more frequent non-surgical management of dental caries. This study conducted to examine if pretreating non-caries and caries dentin with SDF adversely affects the bond strength of composite resin to dentin or not.

Universal adhesive in etch and rinse mode used in the current study, was attributed to Markham et al who reported that the bond stability of universal adhesives to dentin in a self-etch mode would be adversely affected by SDF application, where SEM images of dentin clearly showed the remaining precipitate of SDF, especially in dentinal tubules. Phosphoric acid was responsible for removing the precipitate formed by the SDF application thus recovering some bond strength. Rinsing off the SDF-treated dentin surface in the current study methodology, seems to be able to eliminate the excess of silver precipitate from peritubular and intertubular dentine, favoring the adhesion.

The results of the current study revealed that group 1 showed the highest statistical significant micro-tensile strength than group 2. This result was supported by other studies that showed a decrease in bond strength value of caries affected dentin in regard to sound dentin.

Kucukyilmaz et al. reported that the intermittent remineralization cycles occur during the caries process, leading to occlusion of dentinal tubules with crystals of minerals, and as a result, the bond strength values of this altered dentin can be affected negatively.

Additionally, Spencer et al reported that acid-etching step (as used in the present study) could promote phase transition of this disorganized collagen to a gel so, they postulated that the lack of adhesive penetration in the caries-affected dentin is due to a phase transition in the collagen that has been disordered by caries.

Spencer et al. attributed the decrease in bond strength of caries affected dentin bonded with commercial total-etch BisGMA / HEMA adhesive as used in the present study to the evidence of phase separation into hydrophobic and hydrophilic phases when bonded to wet, demineralized dentin matrices.

As a result of phase separation, the majority of demineralized dentin matrix is primarily infiltrated by HEMA. HEMA has a low crosslink density and thus, it will tend to absorb extraneous water, leading to plasticization and breakdown of the adhesive.

Results revealed that there was no statistically significant difference in microtensile bond strength mean values between group 1 and group 3. Meaning that silver diamine fluoride (SDF), a caries arresting and preventing agent, does not adversely affect the bond strength of resin composite to non-carious dentin.

A systematic review and meta-analysis proposed to evaluate the influence of SDF on bonding, it showed that the adverse effect on the bond strength of adhesive systems seems to be eliminated by the rinsing SDF off after the application time. The rinsing seems to be able to eliminate the excess of silver precipitate from peritubular and intertubular dentine 9 favouring the adhesion.

Studies conducted by Selvaraj et al. (2016), Quock et al. (2012) and Wu et al., (2016) reported no decrease in the bond strength to SDF-treated dentin, this may be attributed to the methodology used in which after the lesions have been treated with 38% SDF solution for the determined period of time (for three minutes), the SDF is then followed by rinsing for 30 seconds with distilled water.

Kucukyilmaz et al also reported a decrease in micro-tensile bond strength of sound dentin treated with SDF. This was attributed to the methodology used in which SDF was applied without rinsing off the precipitant; consequently the dentinal tubules
were occluded by silver precipitant that can prevent deep infiltration of resin tags.15

Regarding the effect of SDF application on demineralized caries affected dentin comparing to sound dentin; results revealed that the least statistically significant micro-tensile strength value was recorded for demineralized dentin treated with SDF.

This may be attributed primarily to the intermittent demineralization/remineralization cycles occur during the caries process, leading to occlusion of dentinal tubules with crystals of minerals.15 In caries affected dentin surfaces, the Ca/P value was found to be significantly lower compared with sound dentin surfaces.15

The composition of the adhesive used also help to explain the results. Scotchbond universal contains 10 MDP. The acidic monomer 10 MDP showed higher chemical affinity with calcium in dental hard tissues because it can bond ionically and form a more stable chemical bond.27,28 But unfortunately the lower Ca and P content and a lower Ca/P ratio in the caries affected dentin was insufficient to enhance a stable chemical bond especially with etch and rinse approach.

Sayed et al revealed that silver ions released from SDF have a high affinity for protein (collagen), so the more collagen exposed the more silver uptake. Therefore, the greater amount of exposed collagen in demineralized dentin will lead to more silver uptake and its reduction to metallic silver.29 Silver ions can infiltrate demineralized dentin lesion completely with further penetration into the underlying sound dentin. Additionally, Fröhlich et al showed a higher levels of silver precipitated were formed on the demineralized compared with non-demineralized dentine that may negatively influence the bond strength on this substrate.31

Etching demineralized dentin after SDF application, responsible for removing a significant amount of calcium 30 and the remaining hydroxyapatite at the bonded interface is critical for generating a sufficient chemical interaction.27 A study conducted by Kucukyilmaz et al who showed that the lowest mTBS values were obtained for the caries affected dentin group after SDF application, and are thought to be caused by dense accumulation of Ag ions.

**CONCLUSION**

Under limitation of this study, the following conclusion could be suggested:

1- Contamination of sound dentin surface by SDF does not affect bonding in case of using universal adhesive in etch and rinse mode.

2- Treatment of caries affected dentin by SDF negatively affect bond strength in case of using universal adhesive in etch and rinse mode.

**CONFLICT OF INTEREST**

The authors declare that they have no conflict of interest regarding the materials used in this study.

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