Evaluation Of Shear Bond Strength Of Two Dentin Bonding Agents With Two Desensitizers – An In-Vitro Study

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Abstracts: Background & Objective**:** The aesthetic quality of a restoration may be as important to the mental health of the patient as the biological and technical qualities of the restoration are to his physical or dental health. In Conservative Dentistry; to mimic, repair and reconstruct the natural tooth structure for the long term, would be based on the use of a restorative material retained only by an adhesive system, whether in load bearing or non-load bearing environments. To achieve high strength, bonds between tooth structure and restorative materials have been a long term goal of dental profession. Objective is to compare the shear bond strength of two different dentin bonding agents with two different desensitizers. Methodology: Eighty molars were taken, which were ground to expose dentin. The teeth were divided into two major groups. Each major group was subdivided into four subgroups of 10 samples each. Groups Ia and IIa were treated as dry bonding groups, groups Ib and IIb were treated as moist bonding groups, group Ic and IIc were rewetted with Gluma desensitizer, and groups Id and IId were rewetted with Systemp® desensitizer. Major group I was treated with Gluma comfort bond and Charisma. Major group II was treated with 3M ESPE Adper™ Single Bond 2 and 3M ESPE Filtek™ Z250. The samples were thermocycled and shear bond test was performed using Instron machine. The data was analyzed using one-way analysis of variance and Tukey's significant different test. Results: The results revealed that the specimens rewetted with Gluma desensitizer showed the higher shear bond strength compared to all other groups, irrespective of the bonding agent or composite resin used. Conclusion: It can be concluded that the moist or rewetting technique could preserve the micro-morphological integrity of the collagen resulting in the optimum penetration of adhesive resin into the demineralized layer, thus, giving higher bond strength. [Joshi P NJIRM 2016; 7(3):67 - 74]

Key Words: 2-hydroxyl ethyl methacrylate, glutaraldehyde, rewetting agents, shear bond strength.

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Introduction: Adhesive dentistry has advanced greatly over the last decade. Composite resins are replacing dental amalgam as an esthetic alternative and posterior restorative material. Strong durable bonds between dental biomaterials and tooth structures are essential to achieve mechanical as well as biologic and esthetic properties. This has led to various developments in the field of adhesive restorative dentistry.

Bonding of resin to enamel is due to the micromechanical bond between the resin bonding agent and the highly inorganic substrate of enamel, which is achieved by the acid etching procedure. True adhesion has been the "holy grail" of dental restorative materials for many decades.[1]

Wetting is essential for the success of all other adhesion mechanism. An adhesive cannot form micromechanical interlocks, chemical bonds, or interpenetrating networks with a surface, unless it can intimately contact the surface, spread onto the surface and fill microscopic irregularities. These conditions are achieved if the adhesive wets the surface. Enamel bonding has shown tremendous clinical success; however, dentin bonding cannot be predictably relied upon for long-term interfacial integrity, because of its complex biological structure.[1]

Marshall et al stated that various structural components and properties of dentin could directly affect the adhesive bond[5]. Bonding of resin to enamel is due to the micromechanical bond between the resin bonding agent and the highly inorganic substrate of enamel, which is achieved by the acid etching procedure. However, bonding of composite resin to dentin is comparatively difficult due to the complex structure of dentin with a low inorganic content randomly arranged in an organic collagen matrix and the presence of dentinal fluid.

Acid etching removes the supporting inorganic matrix of dentin, leaving the organic substance, but the collagen in the organic substance shrinks and collapses easily when it is dried with air syringe after being rinsed with water.[2] The moist or wet bonding technique is one way to preserve the micro morphological integrity of the collagen, and studies

have reported optimum infiltration of adhesive resin into the demineralized layer occurs, giving higher bond strength values.[3]

Over wetting and over drying conditions may have undesirable effects on the bonding performance.[4] To achieve a balance between the two conditioned dentine, various desensitizing solutions have been suggested as rewetting agents. Desensitizing agents reduce the postoperative sensitivity and also enhance the bond strength associated with composite restoration.[7,8,9]

This in vitro study was designed to evaluate the effect of rewetting dentin with two desensitizers on the dentin shear bond strength and to compare them with moist dentin, dry dentin and dentine with different rewetting agents.

Material and Methods: *Materials:*

Desensitizers: Table 1

- Systemp Desensitizer (Ivoclar Vivadent, Shann, Liechenstein)
- Gluma Desensitizer (Heraeus Kulzer)

Bonding Agent: Table 2

- 3M ESPE Adper™ Single Bond 2
- Gluma Comfort Bond

Composition of Composites: Table 3

- 3M ESPE Filtek™ Z250
- Charisma (Heraeus Kulzer)

Table 1: Composition of Desensitizers

SYSTEMP® DESENSITIZER (IVOCLAR VIVADENT)	GLUMA DESENSITIZER (HERAEUS KULZER)
Polyethylene glycol Dimethacrylate 35.0 Maleic acid < 0.01 Glutaraldehyde (50 %) 10.0 Water 55.0(in wt. %)	2-hydroxylethyl methacrylate 36.1% Glutaraldehyde 5.1% Water

Table 2: Composition of Bonding Agents

3M ESPE Adper™ Single Bond 2	GLUMA COMFORT BOND
Bis-GMA* HEMA** Dimethacrylates Polyalkenoic acid Copolymer Initiator Ethanol and Water	HEMA** 4META*** Methacrylate Polycarboxylic acid Glutaraldehyde Solvent Ethanol and water

*Bis-GMA: Bisphenol A-glycidyl methacrylate
**HEMA: 2-Hydoxylethyl methacrylate
***META: methyloxy ethyl trimellitic anhydride

Table 3: Composition of Composites

3M ESPE Filtek™ Z250	CHARISMA (HERAEUS KULZER)
Surface modified zirconia/silica with a medium particle size of approx.3 um or less Non-agglomerated / Non-aggregated 20 nanometer surface modifier silica particles Filler (82% by wt, 68% by vol)	Bis-GMA and TEGDMA**** 64% filler (by volume) Barium aluminium fluoride glass (0.02 – 0.07) Silicon dioxide (0.01 – 0.04 um)

****TEGDMA: triethylene glycol dimethacrylate

Methodology

Eighty freshly extracted, caries-free human permanent molars stored in 10% buffered formalin solution for 2–4 weeks were debrided using periodontal curettes. After cleaning, the teeth were stored in distilled water until use.

Preparation Of The Specimens

Flat dentin surfaces were created on the extracted teeth, with slow-speed diamond disk under water coolant. Then, each tooth was mounted in a chemically cured acrylic resin, such that 3-4 mm of the coronal dentin was exposed. The coronal dentin was finished and polished with 600-grit wet silicon carbide paper. The specimens were placed in distilled water until ready for use. The occlusal surface of specimens in each group was treated with 37% phosphoric acid for 15 sec and the treated surface was thoroughly rinsed with water for the same time with water spray.

These specimens were divided into two major groups, depending upon different bonding agents used. Each group was further divided into four subgroups:

Group A: Gluma Comfort Bond

Group B: 3M ESPE Adper™ Single Bond 2

In all groups, the dentin surface was etched with 37% phosphoric acid for 10 seconds and the treated surface was thoroughly rinsed with water, followed by the application of the respective bonding agent, according to the manufacturer's instructions and light cured

under different dentin surface conditions like dry, wet or dry and rewetted with different rewetting agents.

SUBGROUPS:
Further each major group was divided into four subgroups of 10 samples each, depending upon different etched dentin surface conditions like dry, wet(control group) and rewetted with two different rewetting agents.

Subgroup A I: Dry
After rinsing of the etched dentin surface, it was dried for 5 seconds with an air syringe, positioned 2cm from the dentin surface, followed by the application of GLUMA COMFORT BOND with a disposable brush and light cured for 10 seconds.

Subgroup A II: Moist
Samples were treated same as in subgroup A I except that after rinsing, the etched dentin surfaces were blotted with blotting paper, leaving the surface visibly wet, as evidenced by a glistening appearance. Compressed air was not used.

Subgroup A III: Rewet with SYSTEMP® DESENSITIZER
Samples were treated same as in subgroup A I, but after air drying the dentin surface was re-wetted with SYSTEMP® DESENSITIZER Ivoclar Vivadent(rewetting agent), prior to the application of GLUMA COMFORT BOND.

Subgroup A IV: Rewet with GLUMA DESENSITIZER
Samples were treated as in Subgroup A I but after air drying the dentin surface was re-wetted with GLUMA DESENSITIZER (rewetting agent) prior to the application of GLUMA COMFORT BOND.

Subgroup B I: Dry Subgroup
After rinsing of the etched dentin surface, it was dried for 5 seconds with an air syringe positioned 2cm from the dentin surface followed by the application of 3M ESPE Adper™ Single Bond 2 with a disposable brush and light cured for 10 seconds.

Subgroup B II: Moist subgroup
Samples were treated same as in Subgroup B I except that after rinsing, the etched dentin surfaces were blotted with blotting paper, leaving the surface visibly wet , as evidenced by a glistening appearance. Compressed air was not used.

Subgroup B III: Rewet with SYSTEMP® DESENSITIZER
Samples were treated same as in Subgroup B I, but after air drying, the dentin surface was re-wetted with disposable brush tip saturated in SYSTEMP® DESENSITIZER Ivoclar Vivadent (rewetting agent) prior to the application of 3M ESPE Adper™ Single Bond 2.

Subgroup B IV: Rewet with GLUMA DESENSITIZER
Samples were treated as in Subgroup B I, but after air drying the dentin surface was re-wetted with a disposable brush tip saturated in GLUMA DESENSITIZER (rewetting agent) prior to the application of 3M ESPE Adper™ Single Bond 2.
The results were analyzed by one way ANOVA for multiple comparison and pair wise comparison was made using Post Hoc Tukey test for significance.

FABRICATION OF COMPOSITE CYLINDER:
For this step, tygon tubes were used with internal diameter of 4.5mm and 2mm height. The tygon tube was placed over the sectioned dentin surface. Resin composite was placed over the adhesive in the tygon tube. The composite was polymerized for 40 seconds using light curing unit MONITEX Ti-Lite GT-1500, then the tube was removed with help of No. 11 BP blade.

THERMOCYCLING PROCEDURE:
The specimens were stored in distilled water at 37°C for 24 hours. Thermocycling unit was custom fabricated. It consisted of thermocouple and a heating element. A temperature sensor kept in the water bath was connected to a digital display unit. Temperature of 8–48°C was set with this unit. The specimens were thermocycled in a water bath set between 8 and 48°C for 2500 cycles with a 30-sec dwell time and 10-sec transfer time. Then, the specimens were stored for 1 week in distilled water.

TESTING PROCEDURE: Each specimen was loaded into Universal testing machine using software for testing. The long axis of the specimen was perpendicular to the direction of the applied force. The knife-edge was located at the interface between the composite and dentin surface. The shear bond strength was measured in the shear mode at a crosshead speed of 0.5 mm/min until fracture occurred.

Evaluation of shear bond strength:
The breaking load values were recorded through a computer connected to Instron testing machine.

The values obtained were in ''kg" and bond strength was calculated in Mpa using the formula given below:

Newton = kg × 9.81

$$\text{Bond strength (Mpa)} = \frac{\text{Load (N)}}{\text{Surface area (mm}^2\text{)}}$$

Results:

Subgroup A I Gluma Bond (Dry)		Subgroup B I 3m Espe Adper™ Single Bond 2 (Dry)	
Average	7.05	Average	6.99

Subgroup A Ii Gluma Bond (Moist)		Subgroup B Ii 3m Espe Adper™ Single Bond 2 (Moist)	
Average	8.48	Average	8.98

Subgroup A Iii Gluma Bond & Systemp Desensitizer (Rewet)		Subgroup B Iii 3mespe Gluma Bond & System P Desensitizer (Rewet)	
Average	9.29	Average	9.63

Subgroup A Iv Gluma Bond & Gluma Desensitizer (Rewet)		Subgroup B Iv 3mespe Gluma Bond & Gluma Desensitizer (Rewet)	
Average	10.00	Average	10.68

Statistical Analysis:

Subgroup	Ai	Aii	Aiii	Aiv
Shear Bond Strength	7.05±0.80	8.48±1.32	9.29±1.08	10.00±1.36

There Was A Significant Difference In The Shear Bond Strength Between The Four Groups Using One Way Anova (F (3,36) =11.87, P<0.01). The Post Hoc Using Tukey's Hsd Test Indicated No Significant Difference Within Subgroups.

Subgroup	Bi	Bii	Biii	Biv
Shear Bond Strength	6.99±1.37	8.98±1.24	9.63±1.27	10.68±1.75

There Was A Significant Difference In The Shear Bond Strength Between The Four Groups Using One Way Anova (F (3,26) =9.18, P<0.01). The Post Hoc Using Tukey's Hsd Test Indicated No Significant Difference Within Subgroups.

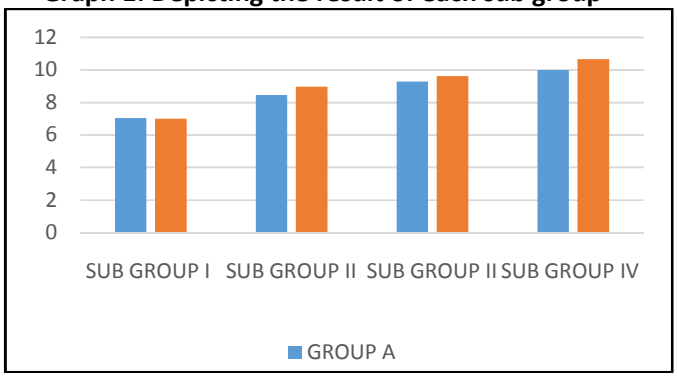
Subgroup	A I	B I	P Value
Shear Bond Strength	7.05±0.80	6.99 ±1.37	=0.95

Subgroup	A Ii	B Ii	P Value
Shear Bond Strength	8.48 ±1.32	8.98± 1.24	=0.75

Subgroup	A Iii	B Iii	P Value
Shear Bond Strength	9.29 ±1.3	9.63± 1.27	=0.80

Subgroup	A Iv	B Iv	P Value
Shear Bond Strength	10.00 ±1.36	10.68± 1.75	=0.78

Graph 1: Depicting the result of each sub group



Discussion: Way back in 1955, Buonocore introduced the concept of acid-etching, i. e. chemically treating the enamel to alter its surface characteristics to allow for adhesion of acrylic resins to the enamel surface of the tooth[6]. After nearly three decades of experience, adhesive techniques are routinely incorporated into clinical practice[10].

Composite restorations are extensively used in the field of restorative dentistry; the mechanical durability of the material still remains an area of concern. Enhancement of bond strength, without compromising the biologic and structural integrity of the tooth can provide a solution to the problem.[11] Acid-etching transforms the smooth enamel into a very irregular surface. After rinsing off the etchant with water and drying the enamel surface with air, a fluid resin is applied on the enamel surface. This resin penetrates into the subsurface, drawn by capillary action. Monomers in the fluid resin polymerize and become interlocked with the enamel surface. The formation of

resin microtags within the enamel structure is the fundamental mechanism of adhesion of resin to enamel. As opposed to enamel, dentin is an intrinsically wet organic tissue penetrated by a tubular maze containing the odontoblastic process, which communicates with the pulp. The dynamic nature of dentin as a substrate is responsible for inconsistent bond strength and marginal leakage, which still occurs with all resin-based adhesives.[25]

When etched dentin is dried using air syringe, the collagen fibers collapse and result in molecular arrangement changes[1]. Excessive drying with air blast to the dentin surface, followed by acid conditioning results in desiccation of the dentin, causing a collapse of the dentin demineralized zone, making it difficult for the hydrophilic resin primer to penetrate completely to the depth of the etched zone. Therefore, bonding to dry dentin results in an incomplete formation of the hybrid layer by compromising the resin infiltration and impregnation of this acid conditioned layer.

This causes decrease in bond strength of dry dentin.[6,14,15] After removal of the hydroxyapatite crystals by acid etching, it is essential to keep the tissue moist, when rinsing off the etchant, to prevent the collapse by air drying of the fibrillary structure of the collagen scaffold, at the superficial demineralized zone. It has been reported that the infiltration of the adhesive monomers, through the nanospaces of the moist, dense collagen web, enhances bond strength. Dentin bonding agents contains hydrophilic monomers as primers along with a solvent such as acetone or ethanol and an adhesive resin, these organic solvents can displace water from dentin surface and from the moist collagen network to allow the monomers to intermingle with the exposed collagen fibres and form a “hybrid layer”.[25]

Hybridization theory originally advanced by Nakabayashi (1991) is the most commonly accepted basis for adhesion to dentin. Acid demineralization of superficial dentin exposes a collagen fibril network having inter and intra-fibrillar microporosities. Low-viscosity monomers placed on this surface diffuse into the demineralized region to form a resin-dentin interdiffusion zone. On polymerization, entanglement of the fibrils by the resin occurs to create a hybrid layer of resin-reinforced dentin. Formation of this hybrid layer is thought to be the primary bonding mechanism for many adhesive systems.[5] The resin-impregnation creates a transitional hybrid layer, that is neither resin nor tooth, but a hybrid of the two. The thin layer of resin-reinforced dentin locks the two dissimilar substances together on a molecular level, sealing the surface against leakage and imparting a high degree of acid resistance. This direct chemical interaction with the inner tubular dentin is the key to bond strength.[2]

Solvents like ethanol and acetone act as a carrier and water chaser, increasing the dentin wettability. Ethanol and water are polar solvents and have the potential to expand the collagen matrix that has collapsed because of drying. As the native conformation of hydrated collagen is maintained by hydrogen bonding, the loss of water would cause collagen collapse, the introduction of polar solvents to re-expand collagen is possible because of their high hydrogen bonding potential.[25]

To keep the exposed collagen scaffold penetrable to resin, it has been recommended that the conditioned dentin surface be maintained in a visibly moist condition, a clinical technique commonly referred to as wet bonding or as moist bonding, recommended by Gwinnett and Tay.[15]The benefit of the wet bonding technique is derived from the ability of water to maintain the collagen framework and intertubular porosity patent for monomer infiltration[15,16].

Moist dentin produced higher bond strength than dry dentin[3,13,14] (group A II and group B II).Most studies have reported that with a moist surface, higher bond strength values are achieved. The risk of moist dentin is an over wet condition resulting in excessive water, which appears to cause phase separation of the hydrophobic and hydrophilic monomer components[3] which leads to blister and globule formation spaces at the resin–dentin interface.

Therefore, it appears that a difficulty exists in achieving a balance between two extreme conditions, which may have undesirable effects clinically on the bonding performance. Here, the dentin surface is left visibly moist (glistening) after etching and rinsing prior to application of the dentin bonding agent. The studies by Kanca et al (1992) suggested, that wet dentinal surface exhibited significantly higher bond strengths than did the dry surfaces.[3] It has been suggested that the inclusion of water in the adhesive may re-expand the collapsed fibrils and facilitate the infiltrations of etched dentin by the resin monomers. To overcome

this problem, various rewetting agents have been tried[1,4,6] Rewetting following acid conditioning not only expands the dematerialized collagen network, but also favors the diffusion of the hydrophilic resin monomers into the etched zone[17] Some of the rewetting agents are used to expand the dematerialized collagen network; they are water, Gluma desensitizers, aqua-prep, 5% glutaraldehyde in water, Tubilicid, MS coat, Vivasens Hurriseal, and Protect.[18]

Pilo R[4], Perdigao J et al[8], F. R. Tay and Gwinnett[15], Van Meerback B et al[16] in their study concluded better bond strength with moist dentin as compared to dry dentin.

The application of Gluma desensitizer after etching of dentin has been shown to improve the efficacy of dentin bonding system (group A III and group B III). Similar results have been reported in a few other studies.[19,20,21,22] Improved bond strength may be due to the use of glutaraldehyde and 2-hydroxyl ethyl methacrylate (HEMA). Glutaraldehyde is a known fixative and flocculating agent that crosslinks collagenous biomaterials[23]. The aldehyde group of glutaraldehyde cross-linking primarily with the ε-amino groups of lysine and hydroxylysine residues in dentin collagen resulting in protein fixation demonstrates that glutaraldehyde may bond to dentin collagen fibrils[7]. This process could possibly stabilize the collagen layer and thus contribute to improved bond strength.

HEMA plays an important role as a stiffening agent preventing any subsequent shrinkage and undergoes a potential reaction (chemical) between its ester functional group and dentin collagen. It also has the ability to promote dentin adhesion and helps in facilitating diffusion of resin monomer and the formation of hybrid layer.[15,16]

In this study, Systemp was the other desensitizer used as a rewetting agent in subgroups AIII and B III. It gave slightly higher shear bond strength value than the moist groups (subgroup A II and subgroup B II). Systemp desensitizer contains polyethylene glycol dimethacrylate (PEG-DMA) and glutaraldehyde. Their combined effectiveness ensures optimal sealing of the tubules[24]. The combination of polyethylene glycol dimethacrylate, which precipitates proteins and thus leads to local concentrations; and glutaraldehyde, which establish stable, covalent bonds of proteins, results in the formation of firm plugs of protein that seals tubules.

Statistically significant difference in dentin shear bond strength between dry dentin, moist dentin and rewetting with dentin desensitizer is observed in the 8 subgroups which were tested.

According to this study, moist dentin gave better bond strength as compared to dry dentin. It could reduce sensitivity and dehydration of dentin, as air spray is not applied. Other results suggested that, the application of desensitizer as a rewetting agent resulted in higher bond strength, if clinically used it could give better post-operative prognosis.

Figure 1: Dentin etched surface

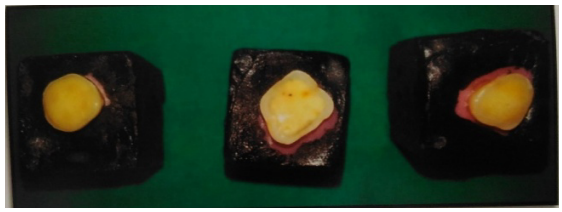


Figure 2: GLUMA COMFORT BOND

Figure 3: 3M ESPE Adper™ Single Bond

Figure 4: Group A - Dentin surface bonded with composite cylinder

Figure 5: Group B - Dentin surface bonded with composite cylinder

Figure 6: Aluminium moulds and Tygon tubes

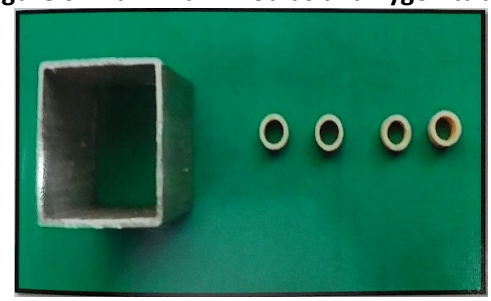
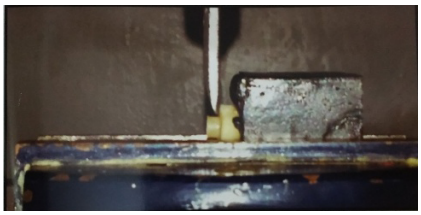


Figure 7: sample sheared with a knife edge on a



universal testing machine

Conclusion:

1. This study stated moist dentin will give better bond strength than dry dentin
2. The use of rewetting agents will improve the bonding of contemporary resin adhesive system to dentin.
3. The use of desensitizers as a rewetting agent is mandatory as it could reduce post-operative discomfort. It also increases the bond strength.

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