

AN IN VITRO EVALUATION OF INFLUENCE OF SALIVA DECONTAMINATION TECHNIQUE ON THE SHEAR BOND STRENGTH OF FIBER REINFORCED CORE BUILD UP MATERIAL TO DENTIN USING 'UNIVERSAL' ADHESIVE AND 'EIGHTH-GENERATION' ADHESIVE.

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Abstract Page

Background: Over the last decade, adhesive dentistry has been expanding at a rapid rate. Universal bonding agent and eighth-generation bonding agent are widespread in dentistry today. Saliva is the main contaminant in oral cavity which hinder the bonding.

Aim: To evaluate the influence of saliva decontamination technique on the shear bond strength of fiber reinforced core build up material to dentin using ‘universal’ adhesive and ‘eighth-generation’ adhesive.

Materials and Methods: Total sixty freshly extracted premolars were randomly distributed into two separate groups of adhesives. Group A- 3M Scotchbond Universal adhesive (3M ESPE, St Paul, MN, USA) and Group B - Futurabond DC (Voco, Germany). Samples were again subdivided into three subgroups (n=10): subgroup-1 Control (without contamination), subgroup-2 Contamination, subgroup-3 Decontamination. Samples were restored with short fiber reinforced composite restoration. Universal Testing machine was used to evaluate the Shear Bond Strength (SBS).

Statistical analysis: The obtained data were examined statistically using one-way ANOVA, followed by Tukey’s *post-hoc* test HSD test ($p < 0.05$)

Results: Group B shown statistically higher mean shear bond strength compared to Group A ($p < 0.05$). In both adhesive groups, maximum mean shear bond strength was obtained in subgroup-1 whereas the lowermost mean shear bond strength was seen in subgroup-2.

Conclusion: Salivary contamination significantly affects the shear bond strength (SBS) of short fiber reinforced composite to dentin using universal adhesive and eighth generation self-etch adhesive. Bond strength significantly improved after reapplication of bonding agent.

Keywords: Futurabond DC; Saliva Contamination; Scotchbond Universal Adhesive; Short Fiber Reinforced Composite; Shear Bond Strength.

INTRODUCTION:

Adhesive dentistry is intensely evolving concerning both, the dentist and patient’s perspective. The dentin adhesives together with the use of dental composite restorative materials have replaced the larger cavity preparations with smaller preparation and more conservative technique.^[1,2] Composites are becoming the preferred material for coronal restoration, due to their conservative properties, good aesthetics, and ease of workability.^[3,4]

Ever X Posterior is a fiber-reinforced composite (SFRC) (GC Europe, Leuven, Belgium), which was introduced for using as a reinforcement under conventional composites because of its high fracture resistance. It is the only available composite resin that structurally corresponds to the dentine.^[6] Garoushi et al. was found a significant enhancement in the material's physical properties when using ever X posterior composite as posterior reinforcement material.^[5,6] It contains 3-4 mm short fibers that can obstruct crack propagation through the restoration and act as a load-bearing barrier under high occlusal forces.^[7-10] Compared with conventional posterior composite resins the ever x posterior has been shown

to resist the polymerization shrinkage stress due to its different fiber orientation and hence reduced the marginal microleakage. [11,12]

Buonocore in the year 1955 introduced the role of adhesion in the field of dentistry. [13,14] Application of dental adhesives has moved from a multistep bonding process to a single-step process, that is from a total-etch to self-etch system. [15] Self-etch adhesive can be applied in a single step and provide adequate adhesion to enamel and dentin, and most importantly comfort for patients. [14,16]

Universal adhesive can be applied either as total-etch or as self-etch adhesives. [16-17] Scotchbond universal (SBU) (3M ESPE, USA) is a single bond universal adhesive composed of 10-methacryloyloxydecyl dihydrogen phosphate (MDP) and methacrylate modified polyalkenoic acid copolymer (PAAC). [18] MDP is a hydrophobic molecule that may impart hydrophobicity to an adhesive, decreasing its water permeability, and chemical bonding between PAAC to calcium in hydroxyapatite results in high bond strength. [18,19]

Combination of nanotechnology to adhesive dentistry leads to the advancement of eighth-generation self-etch bonding agents (Futurabond DC, Voco, Germany). [20-22] As per the manufacturer's instructions Futurabond DC is self-etch adhesives with two components that are mixed before application and are applied as a single-step procedure. These nanofiller reinforced self-etch adhesives have well enamel and dentin bond strength, stress absorption, and longer shelf life. [15,16]

The persistent evolution of adhesive material is now focused on gaining a better understanding of factors affecting adhesion. [21] To get an effective cohesive and adhesive bond, adhesives should not be contaminated. [15] Contaminants like saliva, blood, gingival fluid can cause the failure of the bonding between the resin and the tooth substrate which leads to complications like microleakage, sensitivity, tooth discoloration, secondary caries, and eventual loss of the restoration. [23,24]

Until now, no studies are available evaluating outcome of saliva decontamination technique on the shear bond strength of fiber reinforced core build up material to dentin using 'universal' adhesive and 'eighth-generation' adhesive. Thus, the aim of this study was to evaluate outcome of saliva decontamination technique on the shear bond strength of fiber reinforced core build up material to dentin using 'universal' adhesive and 'eighth-generation' adhesive.

The null hypothesis of this study was that saliva decontamination would not affect the shear bond strength of short fiber reinforced core build up material to dentin using universal adhesive and eighth-generation adhesive.

MATERIALS AND METHODS:

Sixty single rooted human mandibular premolars without carious and fracture were collected according to the protocol approved by the state health University and Institutional Ethical Review Board. The materials used in this study are listed in Table 1.

Sample Preparation and Mounting of Specimens: Using Vernier Calliper, distance of 1.5 mm was measured from the cuspal tip of tooth. The samples were then sectioned using diamond disc at measured point to obtained flat dentin surfaces [figure 1a]. Silicon carbide paper #600 was used to create a homogeneous smear layer. Later all samples were mounted till the cementoenamel junction in aluminum molds with self-cure acrylic resin. Unstimulated human saliva was obtained from a single person in a sterile beaker just before the procedure and was used instantly.

Division of Samples: Sixty samples were divided at random into two groups of adhesives (n=30)

Group A- 3M Scotchbond Universal adhesive (3M ESPE, USA)

Group B- Futurabond DC (Eighth Generation Self-etch adhesive) (Voco, Germany).

Each group was then further divided into three experimental subgroups (n=10) as follows:

Subgroup 1: Control group- Samples were not contaminated with saliva. Application of adhesive was done on prepared flat dentin surface as per the manufacturer's instructions and curing was done for 10 seconds using light-emitting diode unit (3M ESPE).

Subgroup 2: Contamination group- Application of adhesive was done as per manufacturer's instructions on flat dentin surface and samples were contaminated with fresh saliva using a disposable brush for 20 seconds [figure 1b]. Surface was dried with gentle stream of air for 3 seconds and curing was done as in subgroup 1.

Subgroup3: Decontamination – Application of adhesive and salivary contamination were achieved similarly as in subgroup 2. Rinsing of the contaminated surface was carried out for 30 seconds with water from an air-water syringe. Surface was then dried with gentle stream of air for 3 seconds and reapplication of adhesive was done as part of decontamination process followed by light curing.

Composite Placement: Following adhesive application, the Teflon tube of dimensions 3mm x 4mm was placed on the prepared dentin surfaces and filled with ever x posterior composite using horizontal incremental technique [figure 2a and 2b]. Each increment was light-cured for 20 seconds using a Light-emitting diode (3M ESPE). After removal of the Teflon tube, the resin cylinder was additionally cured for 20 seconds. [figure 2c].

Shear bond strength Analysis: Shear bond strength test has been carried out using an Instron Universal Testing Machine (Unitek, 9450 PC, FIE, India) at a continuous crosshead speed of 0.5 mm/minute.

Statistical analysis: Statistical analysis was done by using descriptive and inferential statistics using one-way ANOVA, followed by multiple comparison: Tukey test and student's unpaired t test. Software used in the analysis was SPSS 22.0 version and level of significance is considered as p- value (p<0.05).

Results: The results of the study (Tables 2) (Graph 1) revealed that the 8th generation Futurabond DC showed the greater mean shear bond strength in comparison with Scotchbond universal adhesive in all three conditions that is with contamination, without contamination and decontamination. After contamination with saliva substantial bond strength reduction was seen in both groups.

Mean shear bond strength in (Graph 2) subgroup A1 was 8.89 ± 3.28 , in subgroup A2 it was 3.56 ± 1.23 and in subgroup A3 it was 8.56 ± 1.99 . By using One-way ANOVA statistically significant variation was seen in mean shear bond strength in three subgroups of Group A ($F=16.40$, $p=0.0001$). By using multiple comparison: tukey test (Table 3) statistically significant variance was seen in mean shear bond strength between subgroups A1 and A2 ($p=0.0001$) and between subgroups A2 and A3 ($p=0.0001$) and there was no significant difference was seen between subgroups A1 and A3 ($p=0.949$)

Mean shear bond strength in (Graph 3) subgroup B1 was 10.97 ± 2.62 , in subgroup B2 it was 5.52 ± 1.70 and in subgroup B3 it was 8.60 ± 2.77 . By using One-way ANOVA statistically significant variation was seen in mean shear bond strength in three subgroups of group B ($F=12.74$, $p=0.0001$). By using multiple comparison: tukey (Table 4) test statistically significant difference was seen in mean shear bond strength between subgroups B1 and B2 ($p=0.022$) and between subgroups B2 and B3 ($p=0.0001$) and there was no significant difference was seen between subgroups B1 and B3 ($p=0.092$)

Mean shear bond strength in (Table 5) (Graph 1) subgroup 1 of group A was 8.89 ± 3.28 and in group B it was 10.97 ± 2.62 . By using Student's unpaired t test statistically no significant difference was seen in subgroup 1 of group A and B ($t=1.56$, $p=0.13$). Mean shear bond strength in subgroup 2 of group A was 3.56 ± 1.23 and in group B it was 5.52 ± 1.70 . By using Student's unpaired t test statistically significant difference was seen in subgroup 2 of group A and B ($t=2.95$, $p=0.009$). Mean shear bond strength in subgroup 3 of group A was 8.56 ± 1.99 and in group B it was 8.60 ± 2.77 . By using Student's unpaired t test statistically no significant difference was found in subgroup 3 of group A and B ($t=0.03$, $p=0.97$)

Discussion:

The ideal restoration should not only form a structurally adhesive bond with the tooth but also can resist repetitive masticatory forces over an extended period.^[6] Successful coronal restoration should have good physical and mechanical properties and also provide the fluid tight coronal seal. There are some shortcomings of resin composites in high stress-bearing areas due to polymerization shrinkage, less wear and fracture resistance.^[3] To overcome this, in year 2013, short fiber reinforced composite (SFRC) (Ever X Posterior; GC, Tokyo, Japan) having higher mechanical and physical properties as compared to conventional composite was introduced. It also mimics the stress absorbing properties similar to dentine.^[6] This fiber reinforced composite composed of resin matrix, E glass fibers and barium glass fillers. This filler improves durability of the polymer matrix and prevent cracks in restoration.^[11,12] Garoushi et al. confirmed that short fibre-reinforced composite resin exhibited excellent fracture resistance in high stress bearing areas.^[8] Studies comparing the fracture resistance of

endodontically treated teeth restored with ever X posterior showed the maximum fracture resistance.

The mechanism of bonding to tooth structure involves the flow of resin monomers into the micro porosities and form an effective micromechanical bond.^[16] In restorative dentistry it is very challenging to achieve moisture free working field when proper isolation techniques are not followed. To have strong bonding between composite resin and the tooth substrate, the adherent and adhesive surface mustn't be contaminated.^[17,25] Studies have shown that shear bond strength of composite to dentin was affected by contamination of adhesives with saliva and blood.^[13] A statistically significant weakening in bond strength was seen when salivary contamination occurred after the application of adhesives. Saliva may create a physical barrier declining the effective copolymerization.^[15] This is in accordance with a study done by Taneja et al. who stated that reduced bond strength is due to the deposition of salivary glycoprotein over the unpolymerized adhesive layer.

In this study, natural human saliva from a single donor was used as the contaminant to reduce variability in PH, enzymes, and protein content. Taneja S et al. said that saliva is composed of more than 99% water and glycoprotein which sooner or later lowers the bond strength of dentin adhesives.^[15,26] Glycoproteins in saliva acts by preventing polymerization and the primary factors responsible for a reduction in bond strength.^[23]

In the present study, eighth generation self-etch adhesive Futurabond DC shown better shear bond strength than Scotchbond universal in all the three subgroups. Eighth generation self-etch adhesive Futurabond DC have highly functionalized silica oxide (SiO₂) nanoparticles (0.20 nm). Nano-fillers increases the penetration of resin monomers and the thickness of hybrid layer by crosslinking of resin components. It has acidic hydrophilic monomers used to etched enamel after contamination with saliva or moisture.^[15,16,27] Munoz MA et al. observed that “etch and rinse adhesives are more prone to biodegradation over time compared to self-etch adhesives which prevents degradation of bonds even after long term water storage”.^[13]

Scotchbond universal adhesive contains Vitrebond™ copolymer, which is resistant to moisture.^[13] Despite that, it was found that there is a reduced in bond strength after salivary contamination. This is in agreement with Kim J et al. who said that reduced into bond strength might be due to the thin layer of the biofilm and opposition of the monomer in process of hybridization. Also, hydrolytic enzymes of saliva cause the breakdown of molecules of Bis-GMA (Bisphenol Glycerol Methacrylate) which can further compromises the bonding.^[13,17] Due to the acidic nature of universal adhesive, it is likely to have removed the salivary proteins without difficulty.^[17]

The hydrophilic property of the newer generations acts by dispersing through it and polymerize within the exposed collagen bundles of demineralized superficial dentine.^[29] Saliva decontamination procedures by Erickson et al. and Cobanglu N et al. showed that reapplication of adhesive as decontamination procedure increases the bond strength.^[28,30]

The limitations of the present study are that simulating the oral environment in vitro has shown an increase in physical property, which might not same for in vivo conditions. More studies are needed to evaluate microscopic bond failure.

Conclusion:

Keeping limitations in mind, we can conclude that

1. Salivary contamination significantly decreases the shear bond strength of short fiber reinforced core build up material to dentin in both eighth generation self-etch adhesive and universal adhesive group.
2. 8th generation self-etch (Futurabond DC) adhesive showed better shear bond in comparison to universal adhesive (Scotchbond universal) in both contamination and decontamination group.
3. Reapplication of adhesive system after the salivary contamination resulted in improved bond strength of short fiber reinforced core build up material to dentin in both eighth generation self-etch adhesive and universal adhesive group.

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Table 1: Materials Used In This Study

Table 1: Materials Used

Serial No.	Product Name	Type	Manufacturer	Composition
1	Futurabond DC	Eighth Generation adhesive	Voco,Germany	Organic acid, Bis-GMA, HEMA, TMPTMA, BHT, Ethanol, Fluorides, amines, Catalyst
2	Scotchbond universal	Universal adhesive	3M ESPE, St Paul, MN, USA/ 527687	MDP Phosphate Monomers, Dimethacrylate Resins, HEMA, Methacrylate-Modified Polyalkenoic Acid Copolymer, Fillers, Ethanol, Water, Initiators, Silane
3	Ever X posterior composite	Short fiber reinforced composite	GC Co, Tokyo, Japan/1309121	Bis-GMA, TEGDMA, PMMA, Triethylene Glycol Dimethacrylate, Glass Fillers and Inorganic Granular Fillers

Bis-GMA: Bisphenol A glycidyl methacrylate; HEMA: Hydroxyethyl methacrylate;

TMPTMA: Trimethylolpropane trimethacrylate

BHT: Butylated hydroxytoluene

MDP: Methacryloyloxydecyl dihydrogen phosphate; UDMA: urethane dimethacrylate;

TEGDMA: triethyleneglycol dimethacrylate; PMMA: polymethylmethacrylate

Table 2: Comparison of Shear Bond Strength (MPa) in Group A and Group B

Mean shear bond strength (in MPa) for each subgroup (Mean±SD).		
	Group A	Group B
Subgroup-1: Control (n=10)	8.89	10.97
Subgroup-2: Contamination (n=10)	3.56	5.52
Subgroup-3:Decontamination (n=10)	8.57	8.60

Group A: Scotchbond Universal adhesive; Group: Futurabond DC adhesive

Table 3: Multiple Comparisons: Tukey Test in Group A (Scotchbond universal adhesive)

Subgroup		Mean Difference	Std. Error	p-value	95% Confidence Interval	
					Lower Bound	Upper Bound
A1	A2	5.33	1.04	0.0001,S	2.74	7.91
	A3	0.32	1.04	0.949,NS	-2.26	2.90
A2	A3	-5.00	1.04	0.0001,S	-7.59	-2.42

Group A: Scotchbond universal adhesive; Group B: Futurabond DC adhesive; NS: non-significant difference; S: significant difference;

Table 4: Multiple Comparisons: Tukey Test in Group B (Futurabond DC adhesive)

Subgroup		Mean Difference (I-J)	Std. Error	p-value	95% Confidence Interval	
					Lower Bound	Upper Bound
B1	B2	5.44	1.08	0.0001,S	2.76	8.12
	B3	2.36	1.08	0.092,NS	-0.31	5.04
B2	B3	-3.07	1.08	0.022,S	-5.75	-0.39

Group B: Futurabond DC adhesive; NS: non-significant difference; S: significant difference;

Table 5: Comparison of Shear Bond Strength (MPa) in group A and B

Subgroups	Group A	Group B	t-value	p-value
1	8.89±3.28	10.97±2.62	1.56	0.13,NS
2	3.56±1.23	5.52±1.70	2.95	0.009,S
3	8.56±1.99	8.60±2.77	0.03	0.97,NS

Group A: Scotchbond universal adhesive; Group B: Futurabond DC adhesive; NS: non-significant difference; S: significant difference; SD: Standard Deviation

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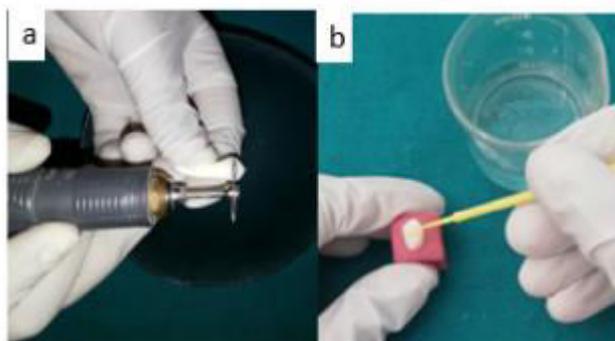


Figure 1: (a) preparing flat dentin surface
(b) Application of saliva

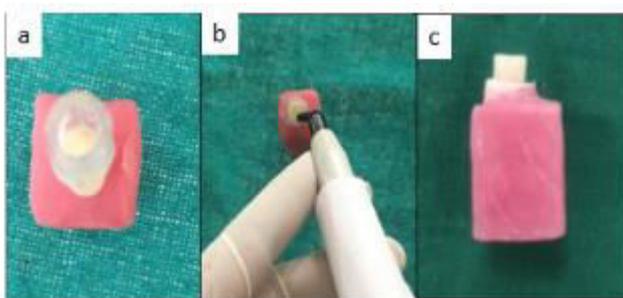
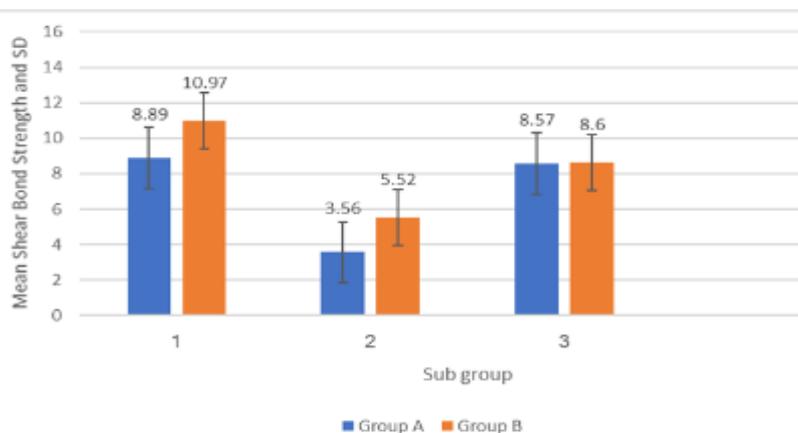


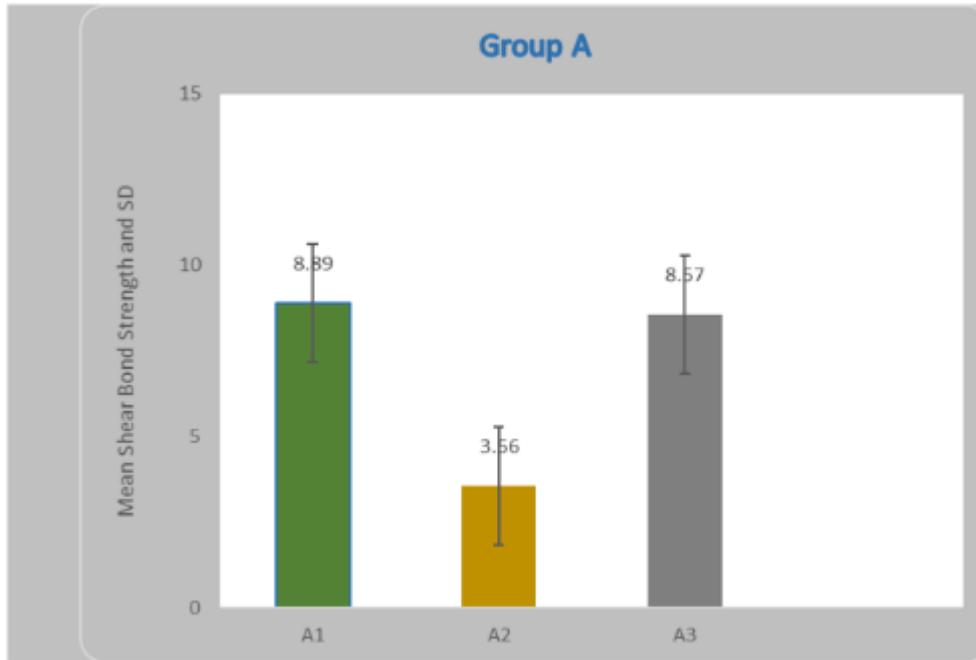
Figure 2: (a) Placement of Teflon tube
(b) Teflon tube was filled with ever x posterior
(c) prepared final specimen

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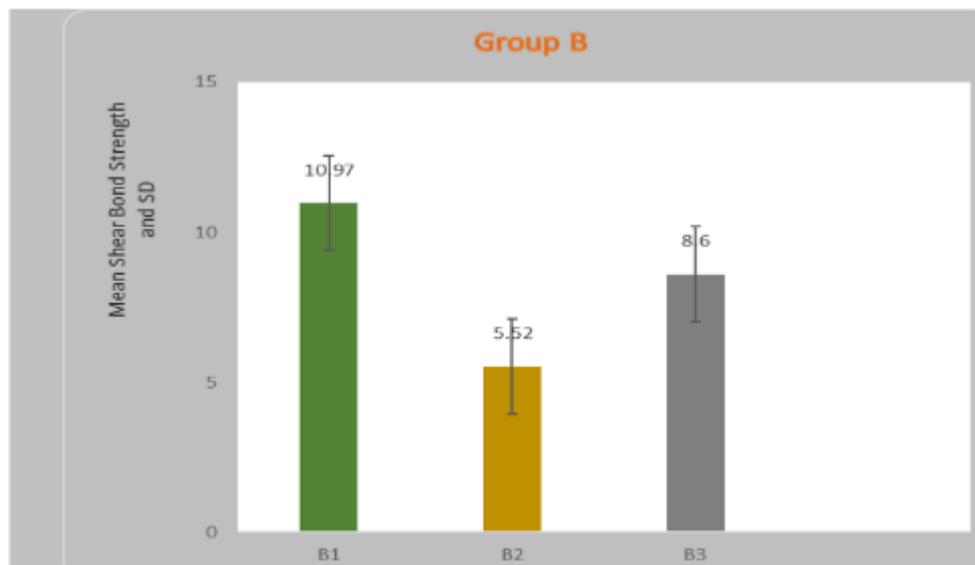
Group A: Scotchbond universal adhesive; Group B: Futurabond DC adhesive; SD: standard deviation

Graph 1: Comparison of shear bond strength (MPa) between Group A: Scotchbond universal adhesive and Group B: Futurabond DC adhesive



A 1: Control; A2: Contamination; A3: Decontamination group; SD: Standard Deviation

Graph 2: Comparison of shear bond strength (MPa) in Group A: Scotchbond universal adhesive



B 1: Control; B 2 : Contamination; B 3 : Decontamination group; SD: Standard Deviation

Graph 3: Comparison of shear bond strength (MPa) in Group B: Futurabond DC adhesive