

EVALUATION OF MICROTENSILE BOND STRENGTH OF ARTIFICIAL CARIES AFFECTED DENTIN-ADHESIVE INTERFACE AFTER USING DIFFERENT CARIES REMOVAL METHODS (IN-VITRO STUDY)

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### **ABSTRACT**

Aim: This study was subjected to investigate the effect of different caries removal methods on immediate and delayed microtensile bond strength of universal adhesives to artificial caries affected dentin.

**Materials and methods:** 60 anonymous extracted human molar teeth were used in this study. The teeth were mounted into acrylic blocks then the coronal enamel and dentin was removed exposing a flat surface of dentin. Then all the specimens were submitted to a pH-cycling model to simulate artificial caries-affected dentin. Two universal adhesives were used GPDM and 10-MDP containing universal adhesive. Teeth were divided into three groups (n=20) according to the type of caries removal method gp 1: diamond bur, gp 2: smart bur, gp 3: laser. Then each group was further subdivided into two subgroups (n=10) according to the type of universal adhesive used. Half of the specimens from each group were tested for Microtensile bond strength ( $\mu$ TBS) immediately and the other half were tested for ( $\mu$ TBS) after 6 months of storage.

**Results:** The diamond bur recorded significantly higher values of  $\mu$ TBS than other methods, while smart prep bur recorded the lowest bond strength. 10-MDP containing universal adhesive showed significantly immediate stronger microtensile bond strength but showed non -significant higher bond deterioration in delayed  $\mu$ TBS than GPDM containing universal adhesive

**Conclusion:** The bond strength changes under various circumstances and at different times between different types of dentin bonding systems and different caries removal methods

**KEYWORDS**: pH cycling method, conservative caries removal methods, universal adhesives, functional monomers

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# INTRODUCTION

The most prevalent non-communicable disease is dental caries, which is more common among people from poor socioeconomic backgrounds. Dental caries has several underlying causes and progresses gradually. The main causes of dental caries development are host vulnerability and dietary choices, which have an impact on the oral cavity's natural flora. Dental caries is a serious public health concern due to its great prevalence and significant effect on general health, even though it is not usually life-threatening.<sup>1</sup>

High-speed drills are considered one of the conventional caries removal techniques, and their efficacy has been demonstrated for a very long time. On the other hand, it has a number of drawbacks, including the excessive removal of healthy tooth structure.<sup>2</sup> Through the avoidance of unnecessary cutting of healthy tooth structure, minimally invasive treatments seek to reduce the size of the cavity preparation. Only friable enamel and soft dentin are removed while minimizing unnecessary cutting of sound dentin<sup>3</sup>, which can be achieved by using adhesive materials.<sup>4</sup> To get over the drawback of high-speed drills and achieve minimal invasive dentistry, it has been suggested to use a variety of techniques for caries removal such as air abrasion, lasers, polymer burs and chemomechanical caries removal methods.5

Methods used for cavity preparation have an impact on the morphology and thickness of the smear layer to the underlying dentin.<sup>6</sup> The theoretical bond strength seems to be limited when adhesive resins are bonded to smear layers unless the smear layers are removed or partially removed. Therefore, adhesive systems can be divided into two main classes based on how they interact with the smear layer: smear layer dissolving self-etch adhesive technique and smear layer removal etchand-rinse technique.<sup>7</sup>

Using an additional acid gel, usually phosphoric acid, the etch-and-rinse strategy involves

completely removing the smear layer and superficial hydroxyapatite.<sup>8</sup> Next, adhesive monomers are infused into the micro-porosities, creating hybrid tissue known as the resin-dentin inter-diffusion zone, or the "Hybrid Layer."<sup>9</sup> However, because there is no rinse phase in the self-etch bond technique, the smear layer is only dissolved, not removed, and the dissolved products are embedded in the interfacial transition zone.<sup>10</sup>

"Universal" or "multi-mode" one-step self-etch adhesives are a new class of adhesives that have emerged in recent years. The concept is that, without sacrificing the bonding efficacy, these adhesives can be applied simultaneously using the etch-and-rinse and self-etching procedures.<sup>11</sup> Dentin substrates tend to be complex and difficult to adhere to<sup>12</sup> The weakest component of tooth-colored restorations is still the bonded interface between dental hard tissues and resinous restoratives and luting agents., despite notable advancements in adhesive strategies.<sup>13</sup> The properties of dentin surface can be altered by various caries removal methods, and this can have an impact on bond strength of dental materials.<sup>14</sup>

Artificial approaches to manufacture in vitro caries-like lesions have been offered as a way around challenges encountered when employing natural caries-affected dentin. Since it was first suggested, the pH-cycling dynamic model has been frequently used to mimic artificial caries dentin as well as the development of enamel lesions. <sup>15</sup> pH-cycling induces a dentin layer damaged by caries that has surface demineralization and hardness values comparable to those of naturally occurring CAD in primary teeth.<sup>16</sup> Also different studies showed that the intertubular nanohardness of permanent teeth with natural and artificial caries-affected dentin tissues was superficially similar.<sup>17</sup>

Therefore, the purpose of this study is to assess how various caries removal techniques alter and affects the  $\mu$ TBS (microtensile bond strength) of universal adhesives to artificial caries affected dentin.

## MATERIALS AND METHODS

### **Ethical Aspects:**

The study has received approval by Beni Suef University's FDBSU-REC research ethics committee, with approval number REC-FDBSU/02112023-03/KS.

## Sample Size Calculation:

Using statistics from previous studies, the sample size was determined using the G\*Power 3.1.9.4 software. The method of calculation of sample size was based on a precision level of  $\pm 5$ , a confidence level of around 95%, and a desired level of confidence interval of 95%.

# 1. Teeth Selection:

Sixty extracted anonymous permanent human molars, which were extracted due to orthodontic needs or periodontal disease were used in this study. Extracted teeth were collected from oral surgery clinic in Beni Suef University.

After extraction, collected teeth were cleaned from soft tissue remnants using hand scaler (Zeffiro; Lascod, Florencem, Italy) and polished using rubber cups and pumice (Shofu one gloss PS, Shofu Dental Corp. Japan). Teeth were then washed and disinfected in 5% chloramine-T solution for 24 hours then stored in distilled water at room temperature till use. Every week, the storage media was changed to reduce deterioration, and all the specimens were used within a month.

Material	Description	Composition	Manufacturer	Batch no.
OptiBond universal	One step universal adhesive	HEMA (hydroxy ethyl methacrylate), GDMA (glycidyl dimethacrylate), GPDM (glycerophosphate dimethacrylate), Acetone, water, ethanol.	Kerr medical, Middleton, WI, USA	7875665
OptiShade	Nano hybrid composite material	Pre-polymerized filler, silica, ytterbium trifluoride, and chemically infused mixed oxides make up the fillers (about 81.5% by weight or 65.1% by volume for the Light, Medium, Dark, and Universal Opaque shades; approximately 78.5% by OptiShade contains Bis-EMA, Bis-GMA, and TEGDMA resins).	Kerr medical, Middleton, WI, USA	8193352
Prime & Bond universal	One step universal adhesive	The ingredients are water, PENTA (dipentaerythritol pentacrylate phosphate), 10- MDP (10-methacryloyloxydecyl dihydrogen phosphate), CQ tertiary amine, and Active GuardTM Technology cross linker.	Dentsply Sirona, USA	2107000661
Ceram.x Spectra ST- HV	Nano-ceramic filled composite material	Matrix: ethyl-4-(dimethylamino) benzoate, dimethacrylate resin, and methacrylic modified polysiloxane nanoparticles Filler: non-agglomerated barium glass, CQ', ytterbium fluoride, spherical, pre-polymerized SphereTEC fillers (particle size & 15 um). Content of filler: 60–62 vol%, 78–80 weight%	Dentsply Sirona, USA	1907000289

TABLE (1) The materials' names and product specifications that were utilized in the current study

# 2. Specimen Preparation:

Each tooth was positioned vertically within a self-curing (Acrostone, Cairo, Egypt) acrylic resin block by means of cylindrical plastic syringe with a diameter of 10 mm. To replicate the alveolar bone level in a healthy tooth, the resin was placed 2 mm below the cemento-enamel junction (CEJ). A scalpel was then used to separate the PVC cartilage once the acrylic resin has fully set.

Under water lubrication, a low-speed diamond saw (Isomet; Buehler, Lake Bluff, IL, USA) was used to remove the coronal enamel and dentin perpendicular to the tooth's long axis until elimination of occlusal enamel and exposure of DEJ. To expose intermediate dentin and standardize the depth, after exposure of DEJ, the periodontal probe was inserted and drilled with a round bur to 1mm depth as a guidance, then the Isomet was used to expose intermediate dentin until the periodontal probe depth landmark. To create a consistent smear layer, 600 grit silicon carbide paper was utilized for 60 seconds under running water. Then further dentin deepening for 1 mm was made by different caries removal methods to mimic morphological and smear layer formation after using different methods.

### 3. Induction Of Artificial caries affected dentin:

To create artificial caries affected caries dentin lesions (CAD), we followed the pH-cycling method which is a suitable method to simulate artificial CAD for several purposes, mainly for bond strength analysis.<sup>15</sup> To replicate artificial CAD, the specimens were subjected to a pH-cycling model. All specimens were submerged for eight hours in 10 ml of demineralizing solution (2.2 mM CaCl2, 2.2 mM NaH2PO4, 50 mM acetic acid; adjusted pH of 4.8) and sixteen hours in the same amount of remineralizing solution (1.5 mM CaCl2, 0.9 mM NaH2PO4, 0.15 mM KCl; adjusted pH of 7.0). After immersions, the teeth were rinsed with deionized water. This process was done without agitation at room temperature for a duration of 14 days. <sup>15, 16</sup> The pH-cycling method provided similar hardness values in comparison to dentine naturally caries-affected until the depth 40  $\mu$ m, and lower than sound dentine until depth of 200  $\mu$ m.<sup>16</sup>

# 4. Specimen grouping and Dentin Surface Preparation:

The specimens were divided into three major groups randomly (n = 20) based on the method employed for caries removal. Each group was further split into two subgroups (n=10 for each) A and B, depending on the kind of universal dentin adhesive that was employed.

*For Group* 1, dentin surface was prepared using a regular-grit diamond bur (Prima Dental Manufacturing Ltd, UK) in a high-speed hand piece with water cooling for 1 mm depth, the diamond bur was moved across the dentin surface five times and the bur was changed with a new one after preparing 5 teeth.

*For Group 2* a SmartPrep bur (SS White, Lakewood, NJ, USA) was used and was connected to a water-cooled, low-speed hand piece and moved five times over the dentin surface. The bur was extended for 1 mm depth. Since the SmartPrep burs are self-limiting and intended for single use only, a new bur was utilized for every tooth.

*In Group 3,* dentin surface was prepared using an Er,Cr:YSGG laser beam (Waterlase IPLUS®, Ain Shams University) with a gold handpiece and MZ8 tip. The parameters—energy density, frequency, water cooling percentage, and aircooling percentage—were set in accordance with the manufacturer's instructions as follows: energy density 4.50 wat, frequency 50 Hz, water cooling 80%, and air cooling 60%. After 30 seconds of moving the beam over the whole dentin surface, the teeth were cleansed with an air water spray and allowed to air dry for 10 seconds.

### 5. Adhesive Application:

Both universal adhesives OptiBond (Kerr medical, Middleton, WI, USA) and Prime and Bond (DENTSPLY Sirona, Konstanz, Germany) were used with self-etch approach according to the assigned study design groups following the manufacturer's recommendations.

Regarding subgroup A, in which OptiBond universal adhesive was used, the adhesive bond was applied to dentin surface with a disposable brush (Microbrush, Young Innovations Europe, Heidelberg, Germany), and with rubbing movement of dentin surface for 20-seconds followed by 10 seconds of a gentle air stream followed by 20-second light cure with an LED curing unit (Demi, Kerr, Middleton, WI, USA; light power density: 800 mW/ cm<sup>2</sup>)

Whereas For subgroup B, Prime&Bond universal adhesive was applied and slightly agitated for 20 seconds followed by solvent evaporation with air for at least 5 seconds and finally light cured with an LED curing unit (Demi, Kerr, Middleton, WI, USA; light power density: 800 mW/cm<sup>2</sup>) for 20 seconds following the manufacture instructions.

### 6. Resin composite build up:

In subgroup A, OptiShade universal composite (Kerr Medical, Middleton, WI, USA) was packed incrementally (each increment 2 mm thickness) to the bonded dentin surfaces of all specimens using a plastic composite applicator (Larident, Italy). The bonded surface was subsequently exposed to light for 20 seconds (light power density: 800 mW/cm2; Demi, Kerr, Middleton, WI, USA). Finally, a resin composite block with a height of around 4 mm was created on each specimen.

The resin composite build up for subgroup B was carried out utilizing Ceram.x Spectra ST-HV nanofilled composite (Dentsply Sirona, USA) in the same manner as for subgroup A.

# 7. Storage of the specimen:

Half of the specimens from each group were evaluated for Microtensile bond strength  $\mu$ TBS after been preserved in distilled water at 37°C for 24 hours in an incubator (BTC, Model: BT1020, Egypt) to complete the polymerization process. The other half of the specimens were tested for delayed  $\mu$ TBS after 6 months of storage in distilled water, and the water was changed every 3 days <sup>19</sup>.

### **Microtensile Bond Strength Test**

Restored teeth were longitudinally sectioned along the buccolingual and mesiodistal planes across the bonded interface with Isomet cutting machine (Buehler, Lake Bluff, IL, USA) under water cooling, to obtain bonded beams per tooth, each with a crosssectional area of approximately 0.9\*0.9 mm<sup>2</sup>. The cross-sectional area of every beam was measured to the nearest 0.01 m using a digital caliper (Absolute Digimatic, Mitutoyo, Tokyo, Japan) to determine the actual bond strength values.

To perform  $\mu$ TBS, four centralized beams of the dentin-resin complex were selected from every specimen. The bond strength test was carried out instantly following the cutting. The beam specimens were fixed to the testing designed microtensile Geraldeli's jig using cyanoacrylate gel (Zapit, DVA Inc, USA). The jig was then fitted and mounted into the 500 N load cell universal testing machine (Instron, MA, USA) and subjected to tensile stress.

The specimens were exposed to a tensile load at a crosshead speed of 0.5 mm/min until it fractured. Consequently, a digital calliper with a precision of 0.01 mm (Model CD-6BS Mitutoyo, Tokyo, Japan) was used to measure the cross-sectional area of each failed specimen. The value of  $\mu$ TBS was determined and given in MPa by dividing the force at debonding [N] by the specimen's bonded surface area [mm<sup>2</sup>]. The findings for each of the four beams under examination were averaged, and the resulting mean was then utilized to make statistical conclusions. Using a software program (Bluehill Lite software, Instron, MA, USA), the bond strength was determined in megapascal units

### **Statistical Analysis:**

After being tabulated and examined in Microsoft Excel, the data were imported into SPSS version 25 (Statistical Package of Social Science) for additional analysis. Shapiro-Wilk's test (all P > 0.05) was applied. The three factors (adhesive resin type, caries removal technique, and storage period) and their relationship with the  $\mu$ TBS test were assessed using a three-way analysis of variance (ANOVA). For multiple testing, the Bonferronie-Holm correction was used to modify all P-values. The minimum value needed for statistical significance was established at P < 0.05. For each bond type, the results were compared across the various caries removal techniques either short-term (after 24 hours) or long-term (after six months of water

storage) using a oneway ANOVA test and a post hoc multiple comparison test.

#### **RESULTS**

Three-way ANOVA showed that  $\mu$ TBS (MPa) was significantly affected by all the variables, P less than 0.001. Also, the interaction between the three variables was statistically significant, P less than 0.001.

The diamond bur recorded significantly higher values of  $\mu$ TBS than other methods, smart prep bur recorded the lowest bond strength among all subgroups with a significant difference, as shown in table (2). The threshold for statistical significance was established at P=0.05. It was found that aging, caries removal technique, and adhesive type all had a significant impact on  $\mu$ TBS P < 0.001. Also, the interaction between the three variables was statistically significant P < 0.001.

TABLE (2) Showing one way ANOVA and post hoc test of immediate and delayed µTBS strength test results between different caries removal methods

Caries removal method	Time factor	Optibond	Prime & bond	Test of significance
Group 1 Diamond bur	Immediate	6.68±1.27	15.54±3.49	t=9.23 p<0.001*
	delayed	3.82±1.24	6.26±1.85	t=4.25 p<0.001*
Unpaired test		t=6.36 p<0.001*	t=5.74 p<0.001*	
% of change		42.8%	59.7%	p=0.35
Group 2 Smart prep bux	Immediate	4.41±1.02	10.60±3.73	t=6.20 p<0.001*
	delayed	3.75±0.89	4.51±1.44	t=1.72 p=0.096
Unpaired test		t=1.87 p=0.070	t=5.91 p<0.001*	
% of change		14.9%	57.5%	p=0.01
Group 3 Laser	Immediate	5.82±1.61	9.19±3.23	t=3.61 p=0.001*
	delayed	4.05±1.15	4.81±2.06	t=1.24 p=0.224
Unpaired test		t=3.47 p=0.002*	t=4.43 p<0.001*	
% of change		30.4%	47.7%	P=0.340



Fig. (1) A chart showing mean immediate microtensile bond strength among different group

### Effect of caries removal method on µTBS

The one-way ANOVA outcome showed that the caries removal method has a significant effect on bond strength The results of various caries removal techniques, either short-term after treatment (after 24 hours) or long-term (after six months), were compared for each bond type used using a one-way ANOVA test and a post hoc multiple comparison test.

### Effect of adhesive type on µTBS

Tukey post-hoc multiple comparison test revealed that there was no statistically significant difference in  $\mu$ TBS between OptiBond universal and Prime&Bond universal, (P>0.05) in group 1, 2 and 3 as shown in table 2. Prime&Bond universal showed significantly stronger bond strength than OptiBond universal (P<0.05) in group 1,2,3 on immediate testing as shown in figure 1.

### Effect of aging on µTBS

Un-Paired sample t-test was used to compare  $\mu$ TBS results in all groups after 24 hours and after six months of water storage. Prime&Bond universal showed significantly higher percentage of bond deterioration from immediate to delayed than OptiBond universal in group 3, (P<0.001) as shown in Table 2.



Fig. (2) A chart showing mean µTBS after 6 months among different groups

### DISCUSSION

A conservative and preventive strategy is currently the foundation of dental caries management.<sup>16</sup> Caries-affected dentin is the main substrate that remains for interaction with dental restorative materials.<sup>20</sup>

Studies indicate that the adhesive strength to caries-affected dentine is lower than that to sound dentine, making adhesion to this substrate a topic of study. Reduced hardness of caries affected dentin, a thicker zone of exposed collagen following the application of the adhesive system, and the obliteration of dentinal tubules by acid-resistant mineral crystals have all been implicated in the decreased bond strengths.<sup>16</sup>

Various models have been developed to create caries-like lesions due to the irregular characteristics of natural caries affected dentin and the challenge of getting this substrate in a standardized method for in vitro experiments.<sup>16</sup> Several methods have been proposed to generate in vitro caries affected-like lesions in an attempt to address standardization concerns when employing substrates impacted by caries. These consist of utilizing buffered solutions, incubating with natural plaque, and using acidified gels and pH cycling method.<sup>20</sup>

When pH-cycling method is considered, the demineralization of the substrate is characterized by the absence of bacteria. The fundamental advantage of this approach is its capacity to produce carious lesions with greater speed and consistency, leading to dentin surfaces with more homogeneity. The pH-cycling approach seems to be an effective way to represent CAD for several reasons, particularly for bond strength investigations.<sup>16</sup>

In 1949, Oskar Hagger created the first adhesive system by combining GPDM with a chemically cured resin-based restorative substance (Sevriton®) in a liquid cavity sealer. GPDM has been used in the most recent generations of universal adhesives as well as in two- and three-step ER adhesive systems. Unlike 10-MDP, GPDM consists of a hydrophilic phosphate group and a short molecule with two hydrophobic methacrylate groups; these functional groups are not separated by a lengthy carbon spacer group.<sup>21</sup> Few data exsist about the effectiveness of adhesive systems containing GPDM.

In this study the results showed high immediate microtensile bond strength  $\mu$ TBS in all caries removal groups bonded with prime and bond universal adhesive containing 10-MDP. The universal adhesive containing GPDM exhibited notably reduced bond strength values as compared to 10-MDP. This could be attributed to the chemical interaction between the functional monomer and dentin structure. Despite being adsorbed to the HAp, GPDM does not create a calcium salt that is stable.<sup>22</sup>

Furthermore, because GPDM is very hydrophilic, GPDM-Ca salts are more vulnerable to hydrolytic breakdown. Owing to its high hydrophilicity, GPDM seems less suitable as a monomer for a "mild" selfetch adhesive that interacts with the HAp ionically in addition to micro-retention. However, it might be a suitable monomer for an etch-and-rinse adhesive.<sup>22</sup>

The results also coincide with Siddarth B et al,<sup>23</sup> in which the chemistry of acidic functional phosphate

monomers with hydroxyapatite was explained by the adhesion/decalcification hypothesis (also known as the "AD concept"). According to this AD idea, phase 1 is the first contact where all acidic monomers link to the Ca ions of hydroxyapatite. This is the phase where phosphate (PO43-) and hydroxide (OH-) ions are released from hydroxyapatite to reach electron neutrality in its solution. Depending on the stability of the monomer-Ca salt produced, the functional monomer will either adhere (phase 2, adhesion method) or dissociate along with an extensive decalcification (phase 2, decalcification route). When it comes to calcium salt stability, 10-MDP is somewhat more stable than GPDM; although GPDM decalcifies, 10-MDP adheres to the adhesion route.23

Carrilho et al.<sup>24</sup> found in the results of systematic review that the 10-MDP monomer can create an acid-base resistant zone on the adhesive interface, thereby enhancing the adhesive stability and bond strengths of dental adhesives containing it when compared to other systems/functional monomers.

When it comes to using lasers to remove dental cavities, erbium-doped yttrium, aluminum, and garnet (Er:YAG) lasers are particularly prominent. This laser works in a pulsed mode, and the handpiece has a water spray to keep tissue from drying up and heat from building up, which promotes effective energy absorption.<sup>25</sup> Numerous studies have assessed the application of lasers in dentistry, emphasizing the use of the erbium laser in hard tissues, which has been used in this research. Enamel and dentin surfaces etched with (Er, Cr: YSGG) lasers show micro-irregularities and no smear layer which could help in deeper resin infiltration of self-etch adhesives and enhance the bond strength.<sup>22</sup>

The results indicated the lowest immediate bond strength was found in the laser caries removal method. De Munck et al.<sup>26</sup> conducted a comparison of bond strength following laser irradiation and conventional acid-etch techniques, their results aligned with these findings. They claimed that since smear layers are not created by laser ablation, adhesive systems applied to laser-irradiated surfaces are not affected by smear layer.

However, various surface alterations were observed as result from laser ablation. There was a noticeable relative decrease in organic tissue within the dentin, which could mean that organic tissue is removed by the Er:YAG laser only in some cases. All these surface alterations may affect hybridization and thus the bonding effectiveness.<sup>26</sup>

Numerous variables, including temperature, duration, and mechanical, chemical, and chemical interactions, might impact how durable dentin bonding is. Several comparative studies assessing the variations in dentin bonding strength have been published. In addition, various artificial aging methods were employed to mimic the alterations in the oral cavity. The most popular technique involves storing the bonded specimens in water at 37°C for a predetermined amount of time<sup>22</sup>, therefore specimens were stored in distilled water for evaluation of delayed microtensile bond strength in this study.

One of the biggest challenges in dentistry has been to promote an effective and long-lasting bond between dentin substrate and adhesive restorative materials because adhesive deteriorates when immersed in water for extended periods of time. As a result, the adhesive strength may gradually decrease over time.<sup>27</sup>

Regarding the impact of polymer bur on bond strength, there was statistically significant difference in bond strength between it and standard diamond bur. Toledano et al. <sup>28</sup> examined the impact of polymer burs as caries removal techniques on both healthy and caries-affected dentin, their findings were in line with the results of this investigation. They concluded that using polymer burs reduced the bonding efficacy of self-etch adhesives to dentin. According to their hypothesis, tested self-etching agents were unable to penetrate the thick

smear layer formed by polymer burs. Additionally, using polymer burs reduced the ability of self-etch adhesives to bind to dentin.<sup>28</sup>

Although MDP has stronger chemical affinity with calcium ions than GPDM, the delayed  $\mu$ TBS of samples treated with Optibond containing GPDM showed lesser bond deterioration than that of those treated with samples containing 10-MDP (Prime & Bond). This indicates that the chemical binding force resulting from the chemical affinity between the functional monomers and HAp contributed less to the overall dentin–resin bond strength than micromechanical retention.<sup>27</sup>

Optibond has a further higher concentration of monomers such as glycerol phosphate dimethacrylate (GPDM) for resin penetration, despite being adsorbed to the HAp, GPDM does not create a calcium salt that is stable. Furthermore, because GPDM is very hydrophilic, GPDM-Ca salts are more vulnerable to hydrolytic breakdown.<sup>29</sup>

In this study the delayed microtensile bond strength decreased more in Prime and bond group than Optibond group although is not statistically significant. These results may be attributed to partial dimeneralization of CAD, where water replaces the minerals lost from the dentin matrix throughout the carious process; this can make up as much as 53% of the volume in CAD. A decrease in MDP-dentin bonding sites and an impact on the chemical bonding of adhesives to CAD could result from partial demineralization in CAD<sup>30</sup> which may be attributed to the decrease in microtensile bond strength by aging.

The solvent in the Optibond group is Acetone, which is very hydrophilic and volatile, so it can quickly remove water and evaporate, leaving a larger concentration of monomers such as glycerol phosphate dimethacrylate (GPDM) enabling resin penetration<sup>23</sup>, which causes increase in bond strength by time in contrast to prime and bond group which bond strength depend mainly on MDP-dentin bonding sites which is low in CAD <sup>29</sup>

## CONCLUSIONS

The following outcomes were reached within the constraints of this in vitro investigation.

- 1. While the literature supports the pH-cycling model for artificial caries induction, it is important to remember that in vitro studies cannot replicate all physiological changes that occur in a clinical setting, including dentin-pulp complex responses such as dentin sclerosis and tertiary dentin formation.
- 2. The bonding performances can also be influenced by the smear layer when applied in self-etching approach. In addition, the functional monomer in adhesives affects the durability of the bond strength.
- 3. The GPDM-based universal adhesive can provide higher micromechanical retention potential, bond strength, and durability than the MDP-based universal adhesive in selfetch mode, although the MDP-based universal adhesive provides better chemical retention.
- The bond strength changes under various circumstances and at different times between different types of dentin bonding systems and different caries removal methods.
- Finally, we concluded that the conventional caries removal methods using diamond burs yields better bond strength to caries-affected dentin than with smart burs or laser.

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