Effect of Er, Cr: YSGG, Nd: YAG, and diode laser against different photosensitizers on tensile and shear bond strength of bonded composite to caries affected dentin

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Abstract. – OBJECTIVE: This study aimed to assess the impact of various cavity disinfection methods on the adhesive bond integrity of composite resin to caries-affected dentin (CAD). Additionally, it will evaluate the micro tensile bond strength (µTBS) of different dentin substrates [CAD and sound dentin (SD)] using various adhesive agents.

MATERIALS AND METHODS: The sample consisted of twenty human mandibular molars with sound dentin (SD) and eighty with CAD. All samples were positioned in a group of polyvinyl pipes with an internal diameter of 3 mm and were positioned perpendicularly up against the cementoenamel junction (CEJ). A total of 60 CAD samples (n=10) were used for shear bond strength (SBS) testing. CAD samples were disinfected with erbium chromium-doped yttrium, scandium, gallium and garnet (Er, Cr: YSGG) laser in group 1, Diode laser in group 2, neodymium-doped yttrium aluminum garnet (Nd: YAG) laser in group 3, riboflavin in group 4, curcumin in group 5, and chlorhexidine in group 6. Sixty CAD samples were treated with Scotchbond™ Etchant and Scotchbond™ multi-purpose primer and bonded with composite for SBS testing. On the twenty remaining CAD samples that did not undergo any type of disinfection, as well as the twenty samples that had sound dentin (SD), two different types of adhesive systems were used for micro tensile testing. For ten of each CAD and SD sample, 3c™ Adper™ Scotchbond™ multi-purpose adhesive was applied to the dentin surfaces. For the remaining ten CAD and SD samples, the All-Bond 2 adhesive system was used. The samples were prepared for µTBS testing. In all specimens, bond failure was assessed using a stereomicroscope. Analysis of variance (ANOVA) and Tukey Honestly Significant Difference (HSD) tests were used to compare the means of multiple groups, at a significance level of *p*<0.05.

RESULTS: CAD disinfected with chlorhexidine (CHX) (17.19±1.02 MPa) exhibited the highest SBS values. Samples in group 5 disinfected with curcumin activated by photodynamic therapy (PDT) showed the lowest SBS (12.49±1.11 MPa). Scotchbond adhesive displayed comparable µTBS (*p*>0.05) when applied on CAD and SD. Moreover, All-Bond 2 adhesive, when applied on CAD, exhibited µTBS significantly lower than All-Bond 2 adhesive on SD (*p*<0.05). Analysis of debonded CAD surface after SBS showed that a cohesive type of failure was dominant in different experimental groups, followed by adhesive

CONCLUSIONS: CAD disinfection with Er:Cr: YSGG, Diode Laser, and Riboflavin activated by photodynamic therapy have the potential to be used as an alternative to CHX for acceptable shear bond strength. The use of Adper™ Scotchbond™ multi-purpose adhesive on CAD and SD did not significantly compromise µTBS.

Key Words:
Caries affected dentin, Micro tensile bond strength, Shear bond strength, Er, Cr: YSGG laser, Diode laser, Nd: YAG laser, Photosensitizers.

Introduction

Tooth cavity preparation involves removing the infected dentin to eliminate bacteria and decay before restoring the tooth with a restorative material[1]. The goal is to remove all the infected dentin while preserving as much healthy tooth structure as possible[2]. However, studies[3] have shown that even after the infected dentin is removed, there may still be bacterial fragments present in the affected dentin. Bacterial fragments left behind in the tooth after treatment can increase the risk of recurrent infections[3].

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Various cavity disinfectants are effective in eliminating bacteria from caries-affected dentin (CAD). However, these disinfectants can also have negative effects on the stability of the resin-dentin bond by affecting the hybrid layer[3,4]. The alteration or loss of this layer can lead to decreased bond strengths and an increased risk of restoration failure[5]. To mitigate these issues, several alternative treatment modalities have been suggested, such as the use of different remineralizing agents, low-intensity laser therapy, and photosensitizers activated by photodynamic therapy (PDT)[3,4,6].

Alternative therapies such as PDT and laser therapies using Er, Cr: YSGG, Nd: YAG, and diode laser have shown promising results in improving bond strength and creating a rough and receptive surface for bonding[7,8]. Er, Cr: YSGG, Nd: YAG, and diode laser work on the principle of ablation and micro-abrasion, which creates a rough surface similar to the etched pattern created by acid etching[9]. This rough surface enhances the adhesion of the composite material to the dentin by increasing the surface area available for bonding. In addition, the lasers are bactericidal by nature[10].

On the other hand, Riboflavin and Curcumin are photosensitizers that are activated by PDT and that have been shown to improve the shear bond strength (SBS) of glass fiber posts to radicular dentin[11,12]. PDT works by using a light source to stimulate the photosensitizer, resulting in the formation of reactive oxygen species (ROS) that can improve the bond strength[11]. However, the effect of PDT on composite bonding is not well understood, and further research is needed to determine its effectiveness for this application. Additionally, the safety and long-term durability of these alternative conditioning therapies need to be thoroughly evaluated before they can be widely adopted in clinical practice.

The use of chlorhexidine (CHX) as a cavity disinfectant and its effect on SBS has been debated in the literature due to heterogeneous outcomes. Some studies[13,14] have reported that CHX can reduce SBS, while others have found no significant negative effect. Currently, there is insufficient comparative data available to evaluate the effectiveness of PDT *vs.* conventional disinfectant CHX on caries-affected dentin (CAD). The available data regarding the effectiveness of cavity disinfectants, such as Er: Cr, YSGG, Nd: YAG, and diode laser, compared to CHX, is limited and often contradictor[5,8]. Moreover, the adhesive properties of CAD surface and sound dentin are debatable[15,16]. It is hypothesized that CAD disinfected with CHX will show similar shear bond strength (SBS) as compared to curcumin and riboflavin activated by PDT, Er, Cr: YSGG, Nd: YAG, and diode laser. Moreover, it is further postulated that the micro tensile bond strength (μTBS) will be significantly lower in CAD surface compared to sound dentin (SD) with different adhesive systems. In light of these gaps in knowledge, this study aims to assess the impact of various cavity disinfection methods on the adhesive bond integrity of composite resin to caries-affected dentin. Additionally, it will evaluate μTBS of different dentin substrates (CAD and SD) using various adhesive agents.

Materials and Methods

The current study adhered to the guidelines of the Checklist for Reporting In-Vitro Study (CRIS) to ensure transparency and reproducibility. The sample consisted of twenty human mandibular molars with sound dentin (SD) and eighty with caries-affected dentin (CAD), which were extracted in an atraumatic fashion. Several techniques were employed to determine the presence of affected and infected dentin, including, visual examination, surface hardness assessment with a dental explorer, dentin staining (0.5% basic fuchsin and 1.0% acid red), and digital micrographic examination. Any dentin that appeared dark stained, mushy, and soft was categorized as infected and subsequently removed. Dentin that appeared hard and pink after staining was categorized as CAD[3].

A radiographic examination was conducted to select only teeth where caries were still within the middle third of dentin ICDAS criteria 3 and 4. Soft inorganic tissues adhering to the tooth surface were removed using ultrasonic scalers (Superior Instruments Co, New York, NY, USA), and chloramine-T trihydrate solution (Merck, Germany) was used at 4°C for 48 hours to disinfect all the teeth. A group of polyvinyl pipes with an internal diameter of 3 mm were positioned perpendicularly up against the cementoenamel junction (CEJ) to receive the tooth samples[4]. The tooth samples were polished using 320-grit silicon carbide paper under running water.

Power analysis was performed using G*Power 3.1, (Heinrich Heine University in Düsseldorf, Germany). Sample size calculation determined the minimum sample size required to achieve 80% power, to detect significant differences at a significance level of 0.05. The analysis yielded 9 samples for each group and the sample size was rounded up to account for any potential loss during the experiment; therefore, a total of 60 CAD samples (10 samples per group) were used for shear bond strength testing[6].

Group 1 – CAD disinfection using Er,Cr:YSGG laser (ECYL): Waterlase ECYL (Biolase, California, USA) was used for CAD sample disinfection. The laser tip, MZ8, was placed 2 mm away from the CAD surface and operated at a frequency of 30 Hz and a power of 0.5 W for 60 seconds. During the treatment, the air/water pressure was maintained at a ratio of 65% to 55%. Following the disinfection procedure, the dentin surface was rinsed with distilled water and dried using air[7,17].

Group 2 – CAD disinfection using diode laser: this experiment involves using a Diode laser (SIROLaser Advance, Sirona, Chicago, IL, USA) to treat teeth specimens. The laser was set to a power of 2 Watts at 10 Hz, with a pulse duration of 10 µs, and will be delivered *via* a fiber-optic tip with a diameter of 200 microns and an incidence angle of 90 degrees. CAD samples were irradiated for 60 seconds, divided into four quarters with 15 seconds of treatment on each surface, and a 10-second rest period between each surface will be applied using a digital timer. After the treatment, the dentin surface was rinsed with distilled water and dried with air[18].

Group 3 – CAD disinfection using Nd: YAG laser: Nd: YAG laser was used to irradiate CAD surfaces using a 400 µ quartz micro-tip in a noncontact circular motion. The power output of the laser was set to 1.5 W, and the tip was kept at the right angle to the CAD surface during the irradiation process. The CAD was irradiated for 60 seconds, with the treatment time divided into four quarters of 15 seconds each, with a 10-second rest period between each surface. This timing was facilitated using a digital timer[19].

Group 4 – CAD disinfection using Riboflavin: a sterile applicator brush was utilized to apply 150 µg/ml of riboflavin photosensitizer (PS). Riboflavin PS was then activated using LED light with a wavelength of 660 nm for approximately 60 seconds, at a power level of 150 mW. After the treatment, the CAD surface was rinsed with distilled water and dried with air[11].

Group 5 – CAD disinfection using curcumin: curcumin photosensitizer at a concentration of 500 mg/L was applied to CAD, and an LED curing unit (Bluephase; Ivoclar-Vivadent, Schaan, Liechtenstein) was employed. The LED was set to a wavelength range of 385-515 nm, with the tip placed vertically at a fixed distance from the cavity. The irradiation intensity was set to 1200 mW/cm^2. After the treatment, the dentin surface was rinsed with distilled water and dried with air[20].

Group 6 – CAD disinfection using CHX (control): a clean application brush and chlorhexidine digluconate solution were used (Consepsis, Ultradent, Istanbul, Turkey) to disinfect the CAD for 10 seconds. Subsequently, the surface was rinsed with distilled water and left to air dry for 5 seconds without desiccation.

After the disinfecting treatments described above 60 CAD samples were treated similarly, as follows: Scotchbond™ Etchant (3M™, USA) (35% phosphoric acid) was applied for 15 seconds on CAD, the etchant was rinsed, and the dentin surface was dried. Scotchbond™ multi-purpose primer was applied for 5 sec and was dried, followed by the application of Adper Scotchbond™ (3M™, USA) and light-curing with a LED curing light (Bluephase, Liechtenstein, Germany) for 15 seconds.

On the twenty remaining CAD samples that did not undergo any type of disinfection, as well as the twenty samples that had sound dentin (SD), two different types of adhesive systems were used for micro tensile testing. For ten of each CAD and SD samples, 3M™ Adper™ Scotchbond™ multi-purpose adhesive was applied to the dentin surfaces as stated previously. For the remaining ten CAD and SD samples, the All-Bond 2 adhesive system (BISCO, USA) was used according to the manufacturer's instructions.

All the treated and untreated CAD and SD surfaces were then restored with incremental resin composite (Tetric N–Ceram, Ivoclar, Liechtenstein) using a 4 mm diameter mold. Following restoration, the samples were exposed to total humidity in an incubator at a temperature of 37°C for 24 hours and then subjected to an aging process using a thermocycler (Bio-Rad, UK). The thermocycler performed 8,000 cycles between 5°C and 60°C, with each cycle having a dwell time of 45 seconds[2,5,21].

Microtensile, SBS Testing, and Failure Analysis

Twenty CAD samples and twenty SD samples (10 of each were bonded with All-Bond 2 and 10 with Adper™ Scotchbond™ multi-purpose) were sectioned vertically along their long axis both mesially-distally and buccally lingually using a slow-speed diamond saw (Isomet 1000, Buehler, Plymouth, MN, USA) to obtain three stick-shaped tensile specimens measuring 1 mm² each from each sample. Specimens were mounted on a micro-tensile testing device (BISCO; Schaumburg, IL, USA) and were subjected to tensile stress at a crosshead speed of 1 mm/min until they failed. The resulting tensile bond strength was expressed in MPa. The formula used to calculate the tensile strength was the force (in N) at the time of fracture divided by the bond area (in mm²).

μTBS = F/A

Where:

μTBS is the micro-tensile bond strength, F is the force at fracture (this is measured in Newtons), A is the cross-sectional area of the bonded interface (measured in square millimeters). CAD specimens disinfected with CHX, Curcumin, and Riboflavin activated by PDT, Er, Cr: YSGG, and Nd: YAG were tested for bond strength between a composite material and CAD. The testing was performed using a universal testing machine (Zwick/Reoll, model 205). The specimens were positioned using a jig, that was used to hold the samples in place during testing. The composite cylinder and blade were positioned at a right angle to each other in the universal testing machine. The bonded samples were subjected to a force applied parallel to their surface, at a speed of 1 mm/min. The experiment was carried out until the failure between the two surfaces occurred. The force required for failure was expressed in MPa[22,23].

Failure Analysis of Debonded Surface

The specimens underwent an examination under a stereomicroscope at 40x magnification (Clinidock model Guangdong, China) to establish their failure mode. Three types of failures were observed: adhesive, cohesive, and admixed. Adhesive failures occur when the bond between the composite material and dentin fails without any damage to the composite or dentin itself. Cohesive failures occur when the composite material or dentin fractures, leaving the other material still attached to the restoration. Admixed failures occur when both adhesive and cohesive failures are present.

Statistical Analysis

The study's outcomes were analyzed using Statistical Packages for Social Sciences (SPSS-20.0, IBM Corp., Armonk, NY, USA). Analysis of variance (ANOVA) and Tukey Honestly Significant Difference (HSD) tests were used to compare the means of multiple groups, at a significance level of $p<0.05$.

Results

Table I and Figure 1 reveal the SBS (Mean±SD) of the CAD surface after different methods of disinfection were applied, before bonding to the restorative material. CAD disinfected with CHX (17.19±1.02 MPa) exhibited the highest SBS values. Samples in group 5 disinfected with curcumin activated by PDT showed the lowest SBS (12.49±1.11 MPa). CAD samples sanitized with

Table I. SBS (mean ± SD) of CAD surface after different methods of disinfection bonded to the restorative material.

Investigated groups	*N*	*Mean ± SD (MPa)*	*p-value*$^{!}$
Group 1: CAD disinfection with ECYL	10	$16.87 \pm 1.21^{\Omega}$	< 0.05
Group 2: CAD disinfection with a diode laser	10	$16.25 \pm 0.28^{\Omega}$	
Group 3: CAD disinfection with Nd: YAG	10	$13.44 \pm 0.91^{\infty}$	
Group 4: CAD disinfection with Riboflavin activated by PDT	10	$16.54 \pm 0.49^{\Omega}$	
Group 5: CAD disinfection with Curcumin activated by PDT	10	$12.49 \pm 1.11^{\infty}$	
Group 6: CAD disinfection with CHX (control)	10	$17.19 \pm 1.02^{\Omega}$	

CAD: Caries affected dentin, ECYL Erbium, chromium-doped yttrium, scandium, gallium, and garnet, Nd:YAG: neodymium-doped yttrium aluminum garnet, PDT: Photodynamic therapy, CHX: chlorhexidine. *Different superscript symbols denote statistically significant differences (Tukey Honestly Significant Difference (HSD) tests. !Showing significant differences among study groups (ANOVA).

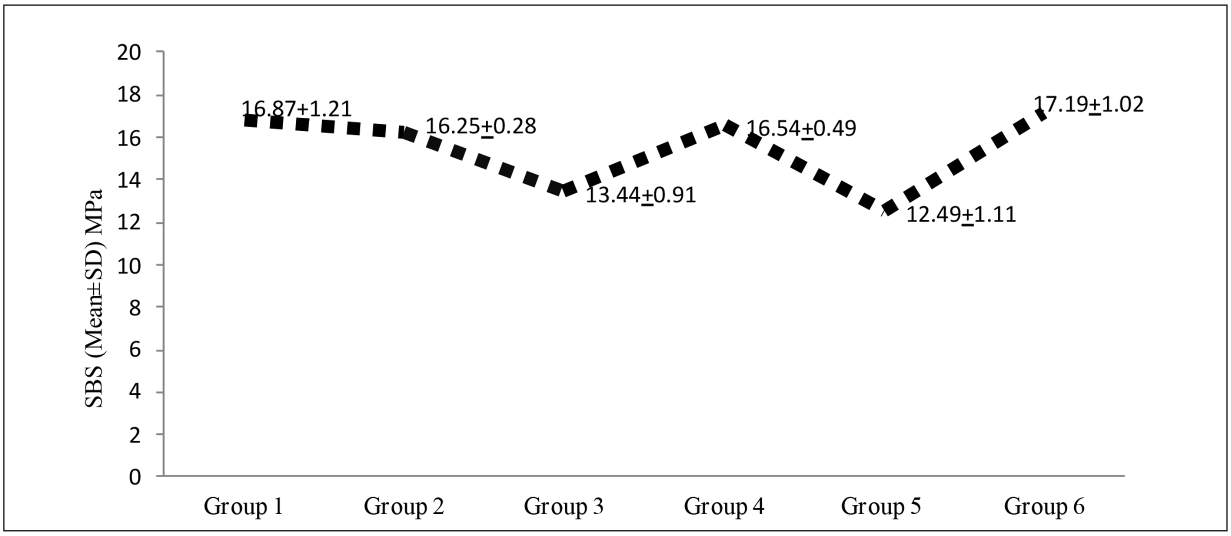

Figure 1. SBS (Mean±SD) of CAD surface after different methods of disinfection bonded to the restorative material. Group 1: CAD disinfection with ECYL (n=10), Group 2: CAD disinfection with a diode laser (n=10), Group 3: CAD disinfection with Nd: YAG (n=10), Group 4: CAD disinfection with Riboflavin activated by PDT (n=10), Group 5: CAD disinfection with Curcumin activated by PDT (n=10), Group 6: CAD disinfection with CHX (control) (n=10).

ECYL, Diode Laser, and riboflavin activated by PDT established comparable bond strength values to CAD surface disinfected with CHX control ($p>0.05$). Similarly, CAD specimens disinfected with Nd: YAG and curcumin activated by PDT exhibited comparable ($p>0.05$) but significantly lower bond values to CAD samples disinfected with ECYL, Diode Laser, Riboflavin, and CHX ($p<0.05$).

Table II and Figure 2 present µTBS (Mean±SD) of CAD and SD surfaces after using two different adhesive systems. Scotchbond adhesive displayed comparable µTBS ($p>0.05$) when applied on CAD and SD. Moreover, All-Bond 2 adhesive, when applied on CAD, exhibited µTBS significantly lower than All-Bond 2 adhesive on SD ($p<0.05$). Scotchbond adhesive demonstrated comparable µTBS ($p>0.05$) when applied on CAD and SD and was similar to All-Bond 2 adhesive on SD.

Analysis of debonded CAD surface after SBS showed that a cohesive type of failure was dominant in different experimental groups, followed by adhesive (Figure 3). The same tendency was noticed in samples after µTBS testing (Figure 4).

Discussion

The present study tested the hypothesis that CAD disinfected with CHX would show similar SBS as compared to curcumin and riboflavin activated by PDT, Er, Cr: YSGG, Nd: YAG, and Diode Laser. It was further postulated that µTBS would be significantly lower in CAD surface compared to SD with different adhesive systems. The question related to SBS of CAD sterilized with different disinfecting agents and restored with composite resin was partially accepted as Er,

Table II. µTBS (mean ± SD) of CAD and SD surface after using two different adhesive systems.

Adhesive	Composition	Dentin	N	µTBS (mean ± SD)	*p*-value!
Scotch bond	HEMA, BISGMA, Maleic acid, polyalkenoate copolymer	CAD SD	10 10	18.48 ± 1.22^{A} 19.55 ± 1.69^{A}	< 0.05
All-Bond 2	UDMA, HEMA, BISGMA, BPDM, NTG-GMA	CAD SD	10 10	12.22 ± 0.64^{B} 20.74 ± 1.22^{A}	

CAD: Caries affected dentin; SD Sound dentin. Dissimilar capital letters within the same column represent a statistically significant difference [Tukey Honestly Significant Difference (HSD)]. !Showing significant differences among study groups (ANOVA).

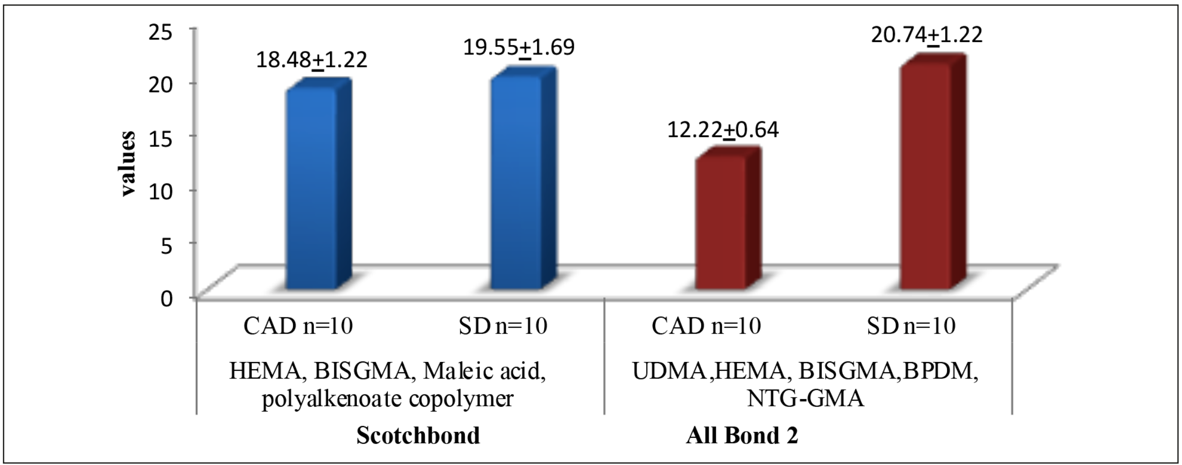

Figure 2. μTBS (mean ± SD) of CAD and SD surface after using two different adhesive systems

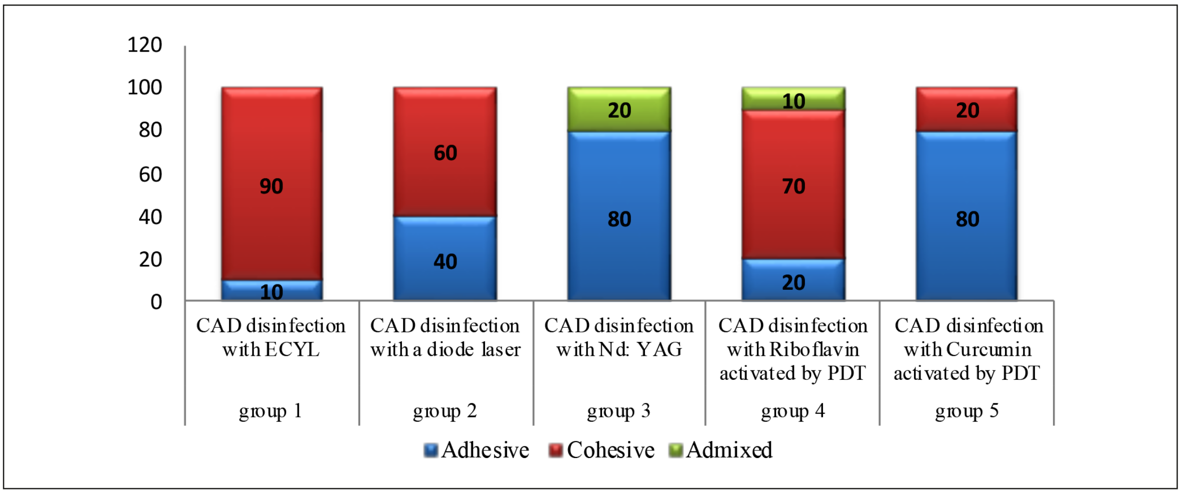

Figure 3. Percentage of failure mode after SBS testing.

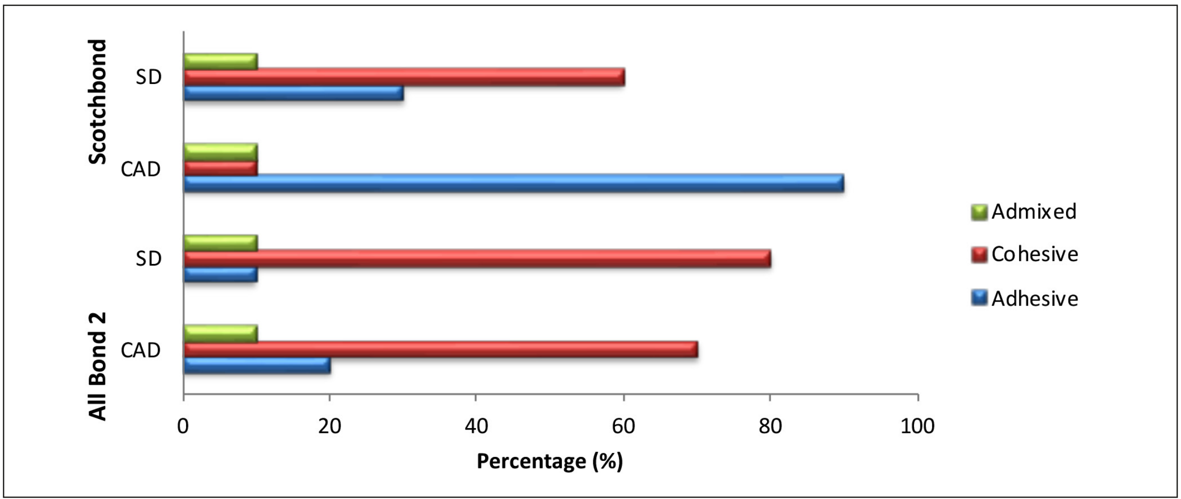

Figure 4. Percentages of failure mode after μTBS testing.

Cr: YSGG, Diode Laser, and riboflavin demonstrated bond values similar to CAD disinfected with CHX. Similarly, the assumption that µTBS will be lower in CAD compared to SD on the application of different adhesive systems also showed partial acceptance as the use of scotch bond adhesive demonstrated comparable µTBS to both CAD and SD. The current study utilized a universal testing machine (UTM) to assess SBS and µTBS. The procedure used for testing, homogenously distributes stress through the dentin structure, indicating a low failure rate[12]. The method is cost-effective and convenient and provides depth profiling and screening of the adhesive system. Additionally, this technique can reciprocate oral conditions with better sensitivity[11,24]. Overall, using a universal testing machine for µTBS and SBS testing provided highly accurate measurements of bond strength, smaller sample sizes, a better understanding of bond failure, and higher resolution[10].

CHX is a commonly used antibacterial cavity disinfectant, and it has been extensively researched for its efficacy in reducing oral bacteria and preventing infection[25]. CHX is effective against both Gram-positive and Gram-negative bacteria with a prolonged antimicrobial effect[26]. Therefore, CHX is considered to be the gold standard against various cavity disinfectants. Moreover, it has been found to improve the bond strength of restorative material by reducing the surface tension of dentin and allowing better penetration of adhesive materials[27]. Additionally, it has been shown to have anti-collagenolytic properties, i.e., it inhibits the activity of matrix metalloproteinases (MMPs) that can break down the collagen matrix of dentin compromising bond strength[28]. In the present study, ECYL, Diode Laser, and riboflavin demonstrated bond values similar to CHX. Concerning ECYL, when used at low power and frequency, the laser energy interacts with water molecules present in the dentin surface and causes the water to evaporate. This leads to dehydration and shrinkage of the collagen fibers[29]. The resulting collagen shrinkage can result in a reduction in the surface area of the dentin, allowing better penetration and adhesion[30]. Additionally, the dehydration process can also result in a change in the surface characteristics of the dentin from being hydrophilic to hydrophobic, which can further improve the adhesion of restorative materials[31].

Similarly, Diode Laser improves bond values to CAD surface according to two established principles. Primarily, Diode causes micro-cracking and roughening of the surface[32]. This roughening can create a more suitable surface for bonding with restorative materials. Secondly, it increases the surface energy of dentin, which can improve the wettability and adhesion of restorative materials[33].

The outcomes of the present study are in line with already reported works by Aljamhan et al[30] and Arslan et al[34], which reported that riboflavin activated by PDT acts as a cross-linking agent on dentin structure. In addition to its cross-linking properties, Riboflavin has also been found to inhibit the activity of the MMP-9 enzyme. The MMP-9 enzyme is known to degrade the extracellular matrix within the dentin, which can weaken the bond between the adhesive and the tooth surface[35]. By inhibiting the activity of MMP-9, riboflavin can stabilize the hybrid layer and prevent matrix destruction, which improves the resin infiltration to the reinforced matrix[36,37]. The findings of the present study are also in concurrence with the work reported by Cova et al[36], Fawzy et al[38], and Daood et al[39].

CAD surface disinfected with curcumin activated by PDT and Nd: YAG laser showed bond integrity significantly lower than CAD surface disinfected with ECYL, riboflavin, and Diode Laser. The reason for this reduction in bond strength is not entirely clear, but it may be related to the oxidative stress caused by the reactive oxygen species (ROS) generated during PDT[40]. These ROS can damage the organic components of the tooth structure and the adhesive interface and may precipitate between the CAD surface and restorative material, deteriorating bond integrity[41]. Similarly, Nd: YAG laser, which operates at a wavelength of 1,064 nm, is not well absorbed by dentin and has a poor water affinity, reflecting poor bond values[42]. When interpreting µTBS values obtained in this study, it was found that 3M™ Adper™ Scotchbond™ multi-purpose adhesive demonstrated better tensile strength to both CAD surface and SD. The probable reason for this outcome is the polyalkenoate acid copolymer and monomers which establish the chemical bond with dentin irrespective of the type of adhesive. These findings are in support of already reported works by Nakajima et al[43,44] who reported that the use of Scotchbond Multipurpose Adhesive on SD and CAD forms long, well-formed resin tags with lateral extensions and thin, good-quality hybrid layer, promoting better adhesive penetration and resulting in favorable µTBS.

Debonded samples after SBS showed that CAD surface treated with ECYL and Diode Laser resulted in cohesive failures due to thermomechanical ablation. Among photosensitizers, CAD disinfected with riboflavin and CAD disinfected using CHX also exhibited cohesive failures. This failure type is usually related to high SBS. Similarly, when evaluating debonded surfaces after µTBS, cohesive failure was dominant among SD and CAD after the application of different adhesives. Multiple external factors can also contribute to failure type, including tubular occlusion due to remineralization, anatomy of dentinal tubules, the binding capacity of the adhesive, and microporosities within the material.

A laboratory-based study[45] found that the concentration, form, and activation of photosensitizers may affect certain outcomes, but additional research is needed to evaluate the impact of different photosensitizers at various concentrations on the mechanical properties of caries-affected dentin, including flexural strength, tensile strength, and microhardness. To confirm clinical effectiveness, it would be necessary to artificially grow different bacterial strains on caries-affected dentin surfaces and assess the effects of different photosensitizers on them. Furthermore, it would be beneficial to evaluate the use of different types of adhesives, such as self-etch and total-etch and rinse, on caries-affected and sound dentin, and to perform scanning electron microscopy to support the findings of the current study.

Conclusions

CAD disinfection with Er, Cr: YSGG, Diode Laser, and riboflavin activated by photodynamic therapy have the potential to be used as an alternative to CHX for acceptable shear bond strength. The use of Adper™ Scotchbond™ multi-purpose adhesive on CAD and SD did not significantly compromise µTBS.

Conflict of Interest

The Authors declare that they have no conflict of interests.

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Ethics Approval

The Ethical Committee of King Saud University approved the study under IRB#FC/4748-23.

Authors' Contribution

All authors have read and agreed to the published version of the manuscript.

Informed Consent

Not applicable.

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