Influence of Application of Dimethyl Sulfoxide on the Bonding Properties to Eroded Dentin

Andres Felipe Millan Cardenas[a] / Lucila Cristina Rodrigues Araujo[b] / Anna Luiza Szesz[c] / Rudys Rodolfo de Jesus Tavarez[d] / Fabiana Suelen Figuerêdo de Siqueira[e] / Alessandra Reis[f] / Alessandro D. Loguercio[g]

Purpose: To evaluate the effect of dimethyl sulfoxide (DMSO) on the microtensile bond strength (μTBS), nanoleakage (NL), and degree of conversion (DC) of universal adhesives on eroded dentin.

Materials and Methods: One hundred thirty-four extracted (134) human third molars were selected for the study. After the dentin surface was exposed, 128 teeth were randomly assigned to 16 experimental groups as follows: dentin surfaces (sound dentin and eroded dentin), application of DMSO (without or with the application of DMSO), and adhesive strategies (etch-and-rinse [ER] and self-etch [SE]). The universal adhesive systems iBond Universal (IBU) and Scotchbond Universal (SBU) were applied and the teeth were restored using a resin composite. After 24 h in distilled water at 37°C, the samples were sectioned and evaluated for μTBS. Selected sticks from each tooth were used for evaluating NL and DC. The remaining six teeth were used to measure the thickness of the collagen layer of the artificially eroded dentin using scanning electron microscopy. Data on μTBS, NL, and DC (%) were analyzed using three-way ANOVA and Tukey's test ($\alpha = 0.05$).

Results: Significantly lower μTBS ($p = 0.0001$) and DC ($p = 0.01$) were observed for eroded dentin than for sound dentin. However, a significant increase in the μTBS ($p = 0.0007$) and DC ($p = 0.001$) was observed for both substrates when DMSO was applied. Moreover, the application of DMSO decreased the concentration of silver nitrate at the bottom of the hybrid layer for both sound and eroded dentin ($p = 0.002$). Eroded dentin showed enlarged tubules with the presence of a collapsed collagen fibril layer approximately 5.0 ± 2.0 mm of thickness.

Conclusion: The bonding performance of both tested universal adhesives improved on both sound and eroded dentin with DMSO pretreatment.

Keywords: dimethyl sulfoxide, adhesive, microtensile bond strength, tooth erosion.

J Adhes Dent 2021; 23: 589–598.
doi: 10.3290/j.jad.b2287671

Submitted for publication: 17.05.20; accepted for publication: 26.08.21

Dental erosion is a growing concern in dentistry world wide, owing to its increased prevalence.[23,40] Dental erosion is caused by repeated contact with acids that are not derived from bacteria,[24] resulting in the loss of minerals and exposure of dentin.[25] This increases tooth sensitivity and impairs esthetics, making restorative treatment necessary.[56]

However, it is expected that acid challenge also alters the chemical and structural composition of the dentinal surface, thus jeopardizing the restorative properties of the ma-

[a] *Professor, Department of Postgraduate Program in Dentistry, CEUMA University, São Luis, Maranhão, Brazil. Research idea, designed testing assembly, performed raman spectroscopy test, co-wrote paper and proofread the manuscript.*

[b] *PhD Student at Department of Postgraduate Program in Dentistry, CEUMA University, São Luis, Maranhão, Brazil. Designed testing assembly, performed bond strength experiments, co-wrote paper and proofread the manuscript.*

[c] *Postdoctoral Fellow at Department of Restorative Dentistry, State University of Ponta Grossa, Ponta Grossa, Paraná, Brazil; Professor, Department of Restorative Dentistry, State University of North of Paraná, Jacarezinho, Paraná, Brazil. Designed testing assembly, performed nanoleakage test, co-wrote paper and proofread the manuscript.*

[d] *Professor at Department of Postgraduate Program in Dentistry, CEUMA University, São Luis, Maranhão, Brazil. Co-wrote paper and proofread the manuscript.*

[e] *Professor at Department of Postgraduate Program in Dentistry, CEUMA University, São Luis, Maranhão, Brazil. Research idea, designed testing assembly, performed bond strength experiments, co-wrote paper and proofread the manuscript.*

[f] *Professor at Department of Restorative Dentistry, State University of Ponta Grossa, Ponta Grossa, Paraná, Brazil. Co-wrote paper, provided consulting for statistical analysis and contributed substantially to the discussion and proofread the manuscript.*

[g] *Professor at Department of Restorative Dentistry, State University of Ponta Grossa, Ponta Grossa, Paraná, Brazil. Provided consulting for statistical analysis and contributed substantially to the discussion and proofread the manuscript.*

Correspondence: *Professor Alessandro D. Loguercio, School of Dentistry, Ponta Grossa State University, Universidade Estadual de Ponta Grossa, Departamento daloguercio@hotmail.come Odontologia, Avenida Carlos Cavalcanti, 4748 Uvaranas, Ponta Grossa, Paraná, 84030-900, Brazil. Tel: +55-42-3220-3741; e-mail: aloguercio@hotmail.com*

Table 1 Adhesive (batch number), groups, composition, and application modes

Adhesive (batch number)	Composition	Application mode			
		Without application of DMSO		With application of DMSO	
		Etch-and-rinse	Self-etch	Etch-and-rinse	Self-etch
iBond Universal [IBU]; Heraeus-Kulzer (010024)	Acetone, UDMA, TEG-DMA, 4-methacryloxyethyltrimellitic anhydride, photoinitiator	1. Apply etchant for 15 s 2. Rinse for 10 s 3. Air dry for 3-5 s to keep the surface slightly moist 4. Apply adhesive as for self-etch mode	1. Keep dentin surface slightly moist 2. Apply the adhesive to the entire preparation with a microbrush and rub in for 20 s 3. Direct a gentle stream of air over the liquid for about 5 s until it no longer moves and the solvent is evaporated completely 4. Light cure for 10 s at 1200 mW/cm^2	1. Apply etchant for 15 s 2. Rinse for 10 s 3. Air dry for 5 s to keep the surface slightly moist 4. Active application of 50% DMSO for 60 s 5. Air stream for 5-10 s keeping the surface slightly moist 6. Apply adhesive as for self-etch mode without DMSO	1. Keep dentin surface slightly moist 2. Active application of 50% DMSO for 60 s 3. Apply air stream for 5-10 s, keeping the surface slightly moist 4. Apply adhesive as for self-etch mode without DMSO
Scotchbond Universal (SBU); 3M Oral Care (638367)	10-MDP phosphate monomer, dimethacrylate resins, bis-GMA, HEMA, methacrylate modifiedb polyalkenoic acid copolymer, camphor quinone, filler, ethanol, water, initiators, silane	1. Apply etchant for 15 s 2. Rinse for 10 s 3. Air dry for 3 to 5 s to keep the surface slightly moist 4. Apply adhesive as for self-etch mode.	1. Keep dentin surface slightly moist 2. Apply the adhesive to the entire preparation and leave undisturbed for 20 s 3. Direct a gentle stream of air over the liquid for approximately 5 s until it no longer moves and the solvent evaporates completely 4. Light cure for 10 s at 1200 mW/cm^2	1. Apply etchant for 15 s 2. Rinse for 10 s 3. Air dry for 5 s to keep the surface slightly moist 4. Active application of 50% DMSO for 60 s 5. Air stream for 5-10 s keep the surface slightly moist 6. Apply adhesive as for self-etch mode without DMSO	1. Keep dentin surface slightly moist 2. Active application of 50% DMSO for 60 s 3. Air stream for 5 to 10 s, keeping the surface slightly moist 4. Apply adhesive as for self-etch mode without DMSO

UDMA: urethane dimethacrylate; TEG-DMA, triethyleneglycol dimethacrylate; 10-MDP: 10-methacryloyloxydecyl dihydrogen phosphate; bis-GMA: bisphenol A diglycidyl methacrylate; HEMA: 2-hydroxyethyl methacrylate; DMSO: dimethyl sulfoxide.

terials deposited on the eroded substrates.[12,51] The continuous and progressive mineral loss creates a mesh of exposed collagen fibrils (organic matrix) on the surface of eroded dentin. These behave as physical barriers against adhesive infiltration.[37,49,71]

Additionally, the collagen fibrils that are not impregnated by the adhesives become susceptible to hydrolysis and create areas rich in water at the hybrid layer, which promotes interfacial defects[37,43,49] and limits the durability of the adhesive interface. In contrast, an increase in demineralization can also be accelerated by the presence of pepsins,[46] matrix metalloproteinases (MMPs), and cathepsins, which naturally occur in saliva and dentin,[68] thus further impairing the restorative procedure.

An interesting alternative for better infiltration of resinous monomers is to apply dimethyl sulfoxide (DMSO: $(CH_3)_2SO$) as an additional primer before applying the adhesive to demineralized dentin.[54,60] DMSO is a polar aprotic solvent that can improve bonding to sound dentin[53,60] by dissolving both its polar and non-polar components. Moreover, DMSO is completely miscible in adhesives.[15] Owing to its small size and amphiphilic nature, DMSO efficiently penetrates biological surfaces[27] and can dissociate highly crosslinked collagen into a sparse network of fibrils.[70] This is attributed to the fact that DMSO breaks the self-associative tendency of water,[63] consequently improving the wettability of demineralized dentin[28] and maintaining its bonding to sound dentin.[54,60]

Although several alternatives for improving adhesion to eroded dentin have been evaluated,[8,13,49,71] no previous study has attempted to improve monomer infiltration into eroded dentin using DMSO as an additional primer.

Therefore, this in vitro study evaluated the effect of DMSO on the microtensile bond strength (μTBS), nanoleakage (NL), and degree of conversion (DC) of universal adhesives applied onto artificially eroded dentin. The null hypotheses were that regardless of the adhesive, the application of DMSO to eroded dentin would not (1) affect bond strength, (2) improve the quality of the adhesive interface in relation to NL, and (3) affect the DC.

MATERIAL AND METHODS

Selection and Preparation of Teeth

One hundred thirty-four (134) caries-free, extracted human molars were used in this study. The study was approved by the Ethics Committee of the local university (2.851.586). The molars were disinfected in 0.1% thymol solution and stored in distilled water. These were used within six months of extraction.

The teeth were sectioned parallel to the occlusal surface using a low-speed diamond saw (IsoMet 1000, Buehler; Lake Bluff, IL, USA) under cooling water to expose the mid-coronal dentin. The flat dentin surfaces were polished using 600-grit silicon carbide (SiC) paper under running water for 60 s to create a standardized smear layer. Usually, occlusal dentin is used for microtensile bond strength testing, because it is easy to prepare and yields many resin-dentin bonded sticks per tooth.

Experimental Design

A total of 128 teeth were randomly assigned to 16 experimental conditions (n = 8) to measure the microtensile bond strength, analyse nanoleakage, and measure the degree of conversion inside the hybrid layer. Specifically, the conditions were 1. dentin surfaces (sound dentin and eroded dentin); 2. application of DMSO (with or without DMSO); 3. adhesive strategies (etch-and-rinse [ER] or self-etch [SE]). Two chemically different universal adhesives were evaluated. While iBond Universal (Heraeus Kulzer; Hanau, Germany) is an acetone-containing, HEMA-free universal adhesive, Scotchbond Universal (SBU; 3M Oral Care; St Paul, MN, USA, also known as Single Bond Universal in some countries) contains ethanol and HEMA. Both treatments were applied according to the manufacturers' instructions. The materials used, batch numbers, compositions, and application modes are listed in Table 1. The remaining six teeth were used to measure the collagen layer thickness of the artificially eroded dentin using scanning electron microscopy (SEM).

Sample Size Calculation

The sample size was determined by considering the μTBS of SBU on dentin. The mean and standard deviation of SBU reported in the literature were 49.8 ± 5.5 MPa.[4,30,51] To detect a difference of 8 MPa among the tested groups using a two-sided test and a significance level and power of 5 and 80%, respectively, the minimum sample size was eight teeth per group (www.sealedenvelope.com). A value of 8 MPa was used, based on a comprehensive revision of bond strength values of dentin published by De Munck et al.[7] In that study, the authors observed that, on average, a decrease of 8 MPa was obtained when the immediate bond strength was compared with the bond strength after a year of aging. These results indicated that 8 MPa can be considered a significant value in terms of in vitro bond strength evaluation.[7]

pH Cycling Model

Sixty-four molars were randomly selected to simulate erosive demineralization. Before erosive cycling, the lateral and root areas were covered with two layers of nail varnish to allow erosive demineralization only on the occlusal surface. The erosive cycle of de- and remineralization was accomplished by immersing each specimen in a cola soft drink with a pH of 2.6 (Coca-Cola; Rio de Janeiro, RJ, Brazil) four times for 90 s (10 ml per specimen) for five days.[26,51,68] After each demineralization cycle, the specimens were rinsed with deionized water for 10 s and immersed in remineralizing solution with a pH of 6.7 (4.08 mM H_3PO_4, 20.10 mM KCl, 11.90 mM Na_2CO_3, and 1.98 mM $CaCl_2$, 10 ml per specimen) for 60 min.[11,51] The cola-based soft drink and remineralization solution were replaced after each erosive challenge. The pH levels of all solutions were monitored periodically using a pH meter.

Restorative Procedures

All teeth were thoroughly rinsed with water, and the surrounding enamel was removed using a diamond bur in a high-speed handpiece (#2135, KG Sorensen; São Paulo, SP, Brazil) under water irrigation. All bonding procedures were performed by a single operator.

For groups in which DMSO was applied, the experimental primer containing DMSO was prepared according to the method described by Stape et al.[54] For this, 50 μl of DMSO (Sigma Aldrich; St Louis, MO, USA, pH 8.2) was mixed with 50% (v/v) water. The primers containing DMSO were changed daily.

For the ER and SE strategies, the DMSO primer was scrubbed on the surface for 60 s using a microbrush. To remove excess primer, an air stream was gently applied for 5 s using an air syringe at a distance of 10.0 cm.[52,54] Independent of DMSO application, the dentin surface was kept slightly moist before applying the adhesive. The adhesives were applied as shown in Table 1.

Further, composite resin (Opallis, FGM; Joinville, Brazil) was built up in 2-mm layer increments that were light cured for 40 s with an LED light-curing unit set at 1200 mW/cm^2 (Radii, SDI; Bayswater, Victoria, Australia). A radiometer (Demetron L.E.D. Radiometer, Kerr Sybron Dental Specialties; Middleton, WI, USA) was used to check the light intensity every 4 specimens.

After storage in distilled water at 37°C for 24 h, the restored teeth were sectioned longitudinally in the mesio-distal and buccal-lingual directions across the bonded interfaces using a slow-speed diamond saw (IsoMet, Buehler) to obtain 0.8-mm^2 resin-dentin bonded sticks. The sticks were measured using digital calipers (Digimatic Caliper, Mitutoyo; Tokyo, Japan) to calculate the cross-sectional bonding area. The number of resin-dentin bonded sticks suffering pre-test failure (PT) during specimen preparation was recorded for each tooth. The resin-dentin bonded sticks were assigned as follows: two resin-dentin bonded sticks per tooth from each experimental condition group were used to evaluate the in situ DC within the adhesive/hybrid layers; three resin-dentin bonded sticks per tooth were used to evaluate NL; and the remaining resin-dentin bonded sticks were tested to determine μTBS.

Microtensile Bond Strength (μTBS)

The resin-dentin bonded sticks were attached to a notched Geraldeli jig[33] using cyanoacrylate adhesive (Super Bonder Gel, Loctite; São Paulo, Brazil) and tested in tension (Instron; Enfield, CT, USA) at a crosshead speed of 1.0 mm/min until failure occurred. The μTBS (MPa) were calculated by dividing the load at failure by the cross-sectional bonding area.

The failure mode of the resin-dentin bonded sticks was classified as cohesive (C, failure exclusively within the den-

Table 2 Number of specimens (%) according to the fracture mode

	Sound dentin												Eroded dentin											
	Without DMSO						With DMSO						Without DMSO						With DMSO					
	ER			SE			ER			SE			ER			SE			ER			SE		
Adhesive	A/M	C	PT	A/M	C	PT	A/M	C	PT	A/M	C	PT	A/M	C	PT	A/M	C	PT	A/M	C	PT	A/M	C	PT
IBU	80 (99)	0 (0)	1 (1)	84 (99)	0 (0)	1 (1)	80 (98)	0 (0)	2 (2)	78 (98)	0 (0)	2 (2)	70 (95)	0 (0)	4 (5)	78 (93)	0 (0)	6 (7)	84 (98)	0 (0)	2 (2)	87 (99)	0 (0)	1 (1)
SBU	86 (99)	1 (1)	0 (0)	90 (100)	0 (0)	0 (0)	85 (97)	3 (3)	0 (0)	87 (98)	2 (2)	0 (0)	76 (94)	0 (0)	5 (6)	76 (96)	0 (0)	3 (4)	81 (96)	0 (0)	3 (4)	80 (96)	0 (0)	3 (4)

A/M: adhesive/mixed fracture mode; C: cohesive fracture mode; PT: pre-test failure; DMSO: dimethyl sulfoxide; ER: etch-and-rinse; IBU: iBond Universal; SBU: Scothbond Universal; SE: self-etch.

tin or resin composite) or adhesive/mixed (A/M, failure at the adhesive-dentin interface, or with partial cohesive failure of the neighboring substrates). Failure mode was determined using a stereomicroscope at 100X magnification (Olympus SZ40; Tokyo, Japan). Specimens that failed prematurely were included in the tooth mean for statistical analysis using an arbitrary value (50% of the lowest measured value).

Nanoleakage Analysis (NL)

Three resin-dentin bonded sticks for each tooth were placed in 50% (w/v) ammoniacal silver nitrate solution in the dark for 24 h and subsequently immersed in a photodeveloping solution for 8 h under fluorescent light.[57] The specimens were sequentially wet polished with 600-, 1000-, 1200-, 1500-, 2000-, and 2500-grit SiC papers and 1.0- and 0.25-µm diamond paste (Buehler) using a polishing cloth. The silver penetration of the specimens was observed using a field-emission scanning electron microscope (FE-SEM, VEGA3 TESCAN; Shimadzu, Tokyo, Japan) in backscatter mode after cleaning, drying, and sputter-coating with carbon-gold. The adhesive-dentin interfaces were analyzed using FE-SEM.

The amount of NL within the adhesive layer, hybrid layer, and resin tags of each stick was measured in three regions (5 µm x 5 µm) of the resin-dentin bonded sticks.[38] After obtaining the first image in the center of the stick, two new images were taken 0.3 mm to the right and 0.3 mm to the left of the center by a technician who was blinded to the experimental conditions under evaluation.[19] The relative percentage of NL was calculated based on the amount of Ag present within the adhesive and hybrid layers in each specimen using ImageJ software (NIH; Bethesda, MD, USA).[47]

Degree of Conversion Inside Hybrid Layer

Two resin-dentin bonded sticks from each tooth were randomly evaluated at 24 h. Then, the sticks were wet polished with 600-, 1000-, and 2000-grit SiC papers and ultrasonically cleaned for 2 min each time after switching from a coarser-grit to a finer-grit SiC paper, with 20 min of cleaning after the last step. Raman spectra were collected using a micro-Raman spectrometer (XploRA ONE Raman microscope, Horiba Scientific; Piscataway, NJ, USA) to investigate the DC inside the hybrid layer of the adhesive interfaces. The micro-Raman spectrometer was internally calibrated for zero and coefficient values using a silicon standard sample provided by the manufacturer. The Raman microscope was configured to operate with a 785-nm diode laser, 100X objective, 600 lines/mm grafting centered between 500 and 1800 cm^{-1} using 100 mW power, spatial resolution of approximately 3 µm, spectral resolution of 5 cm^{-1}, and accumulation time of 25 s with three co-additions. The spectra were obtained in the middle of the hybrid layer, an arbitrary area of the intertubular dentin, and between two dentin tubules at three different sites for each specimen. The spectra were post-processed using Opus Spectroscopy Software version 6.5 (Bruker; Illerica, MA, USA). Two-point baseline and a maximum band-height ratio protocol were used to measure the absorption intensity, as described by Hass et al[19] and Wang et al.[66] The average of the values was used for statistical analysis, and the spectra of the uncured adhesives were considered as references.

The ratio of the double-bond content of monomer to polymer in the adhesive was quantified by calculating the ratio of the aliphatic C=C (vinyl) absorption (1638 cm^{-1}) to the aromatic C=C absorption (1608 cm^{-1}) signals for both polymerized and unpolymerized samples (n = 5). The DC was calculated using the following formula: DC (%) = $(1–[R_{cured}/R_{uncured}]) \times 100$, where "R" is the ratio of aliphatic and aromatic peak intensities at 1638 and 1608 cm^{-1} in the cured and uncured adhesives, respectively.[19] In addition, more intense peaks were observed for all materials, and the corresponding chemical bonding was recorded.

Thickness of the Exposed Collagen Layer

To evaluate the thickness of the collagen layer that was exposed by pH cycling, six teeth were used. The teeth were sectioned parallel to the occlusal surface using a low-speed diamond saw (IsoMet 1000, Buehler) under water cooling to expose the mid-coronal dentin. Subsequently, each tooth was transversely sectioned in a buccal-to-lingual direction to obtain two halves per tooth (n = 12 specimens). The pre-cut grooves were made on the pulp side to permit segmentation.

Table 3 Means and standard deviations of dentin bond strengths (MPa), as well as statistical analysis for all experimental groups

	Sound dentin				Eroded dentin			
	Without DMSO		With DMSO		Without DMSO		With DMSO	
Adhesive	ER	SE	ER	SE	ER	SE	ER	SE
IBU	35.6 ± 2.5^{B}	35.6 ± 2.4^{B}	43.6 ± 2.7^{A}	41.2 ± 2.2^{A}	24.4 ± 3.4^{D}	22.3 ± 2.7^{D}	30.9 ± 2.4^{C}	30.8 ± 1.6^{C}
SBU	49.3 ± 3.2^{b}	48.8 ± 3.4^{b}	55.0 ± 2.8^{a}	55.6 ± 2.5^{a}	30.7 ± 2.5^{d}	27.5 ± 3.8^{d}	36.6 ± 2.9^{c}	36.9 ± 2.5^{c}

For each adhesive, different capital or lower letters indicate statistically significant differences between the groups (three-way ANOVA; Tukey's test, $p < 0.05$).
ER: etch-and-rinse; SE: self-etch; DMSO: dimethyl sulfoxide; IBU: iBond Universal; SBU: Scothbond Universal.

Table 4 Means and standard deviations of nanoleakage values (%), as well as statistical analysis for all experimental groups*

	Sound dentin				Eroded dentin			
	Without DMSO		With DMSO		Without DMSO		With DMSO	
Adhesive	ER	SE	ER	SE	ER	SE	ER	SE
IBU	12.3 ± 1.7^{B}	11.8 ± 2.2^{B}	7.0 ± 1.9^{A}	7.9 ± 1.4^{A}	24.2 ± 2.3^{D}	22.1 ± 1.8^{D}	18.4 ± 1.8^{C}	17.8 ± 1.4^{C}
SBU	8.8 ± 1.7^{b}	8.1 ± 1.2^{b}	5.2 ± 1.5^{a}	5.5 ± 1.4^{a}	16.0 ± 2.1^{d}	15.4 ± 1.5^{d}	12.6 ± 1.8^{c}	12.7 ± 1.8^{c}

*For each adhesive, different capital or lowercase letters indicate statistically significant differences between the groups (three-way ANOVA; Tukey's test, $p < 0.05$).
ER: etch-and-rinse; SE: self-etch; DMSO: dimethyl sulfoxide; IBU: iBond Universal; SBU: Scothbond Universal.

Table 5 Means and standard deviations of degree of conversion values (%), as well as statistical analysis for all experimental groups*

	Sound dentin				Eroded dentin			
	Without DMSO		With DMSO		Without DMSO		With DMSO	
Adhesive system	ER	SE	ER	SE	ER	SE	ER	SE
IBU	56.3 ± 2.2^{B}	57.9 ± 2.2^{B}	64.2 ± 2.0^{A}	65.3 ± 2.4^{A}	46.2 ± 1.4^{D}	47.9 ± 2.2^{D}	52.5 ± 2.1^{C}	53.3 ± 2.7^{B,C}
SBU	66.4 ± 1.4^{b}	65.8 ± 1.8^{b}	71. ± 1.5^{a}	70.9 ± 1.4^{a}	54.1 ± 2.2^{d}	55.2 ± 2.1^{d}	62.2 ± 1.7^{b,c}	60.8 ± 2.3^{c}

*For each adhesive, different capital or lower letters indicate statistically significant differences between the groups (three-way ANOVA; Tukey's test, $p < 0.05$). ER: etch-and-rinse; SE: self-etch; DMSO: dimethyl sulfoxide; IBU: iBond Universal; SBU: Scothbond Universal.

The specimens from each tooth were divided into sound and eroded dentins. To permit erosive demineralization to occur only on the occlusal surface, the lateral and pulpal areas were covered with two layers of nail varnish. Subsequently, the specimens were subjected to demineralization based on the pH-cycling model.

After demineralization, to keep the dentin wet, the surfaces were rinsed with tap water for 30 s and air dried for 5 s using an air syringe at a distance of 10.0 cm. The specimens were treated according to the method reported by Siqueira et al[49] and Kenshima et al,[22] and the entire surface was examined using SEM (MIRA3 LM, Tescan Orsay Holding; Warrendale, PA, USA). Three photomicrographs of the representative surface areas were taken at 5000X magnification and were used to measure the thickness of the exposed collagen layer.

Statistical Analysis

The Shapiro-Wilk test was used to assess data distribution normality. Barlett's test was performed to determine the validity of the assumption of equal variances. The mean μTBS and percentages of NL and DC in all bonded sticks from the same tooth were averaged for statistical analyses; therefore, the experimental unit in the study was the tooth. Specimens with PT were included in the tooth mean for statistical analysis as half of the minimum bond strength measured in the study (4.0 MPa).[38,39] The μTBS (MPa), NL (%), and DC (%) means for every test group were obtained from the average of the 8 teeth used per group. For each adhesive, μTBS (MPa), NL (%), and DC (%) data were subjected to three-way ANOVA (dentin surfaces vs application of DMSO vs adhesive strategies) and Tukey's test with a level of significance of 5%.

RESULTS

Microtensile Bond Strength (μTBS)

The mean cross-sectional area of the tested resin-dentin bonded sticks was 0.78 ± 0.5 mm^2. Approximately 20 to 25 resin-dentin bonded sticks were obtained per tooth, including PTs. The most common failure pattern was adhesive/mixed in all experimental groups (Table 2).

The μTBS cross-product interaction was insignificant (Table 3; p = 0.38). However, the primary factors, that is, dentin surface and application of DMSO, were significant (p = 0.0001 and p = 0007, respectively; Table 3), whereas the adhesive strategy showed no difference (p = 0.76). Considerably lower μTBS was observed for eroded dentin than for sound dentin (Table 3; p = 0.0001). However, a large increase in the μTBS was observed for both substrates when DMSO was applied (Table 3; p = 0.0007).

Nanoleakage Analysis

The cross-product interaction was insignificant (Table 4; p = 0.45). In contrast, the dentin surface and application of DMSO were significant (p = 0.008 and p = 0.002, respectively; Table 4), whereas the adhesive strategy showed no difference (p = 0.63). A significantly higher NL value was observed for the eroded dentin than for the sound dentin (Table 4; p = 0.008). The application of DMSO decreased the NL values for both sound and eroded dentin (Table 4; p = 0.002).

Degree of Conversion Inside the Hybrid Layer

In terms of DC, the cross-product interaction was insignificant (Table 5; p = 0.38). However, the primary factors, that is, dentin surface and application of DMSO, were significant (p = 0.01, and p = 0.01, respectively; Table 5), whereas the adhesive strategy showed no difference (p = 0.70). Considerably lower DC was observed for the eroded dentin compared than for the sound dentin (Table 5; p = 0.01). A significant increase in the DC was observed for both substrates when DMSO was applied (Table 5; p = 0.001).

Thickness of the Exposed Collagen Layer

Figure 2 showed representative SEM images of the sound (a) and eroded dentin (b). For sound dentin, a smear plug is apparent in the lumen of the tubule without any change in the tubular diameter (a). For eroded dentin, more enlarged tubules with a collapsed collagen fibril layer were observed. The thickness of a collapsed collagen fibril layer was approximately 5.0 ± 2.0 mm (b).

DISCUSSION

In this study, significantly lower bond strength was observed for eroded dentin than for sound dentin. It is known that erosion can result the dissolution of the mineral component, leaving a zone of dense, fibrous collagen network that has buffering properties.[3] The erosive challenge results in a higher degree of dentin demineralization and a deeper demineralized layer.

This erosion is caused by the presence of phosphoric acid in the cola-based soft drink.[48] This method has been frequently used to cause erosive demineralization in dentin prior to the application of adhesives.[1,8,51] Although other erosive solutions can be used to erode dentin,[48] the erosive effect induced by cola-based soft drinks is a more realistic erosive challenge, with fewer pre-test failures of resin-dentin bonded sticks[8] compared to citric acid.

The significant loss of minerals caused by cola-based soft drinks is accompanied by a loss in collagen periodicity,[9] thereby promoting a higher porosity of the collagen matrix, similar to that occurring in caries.[59] Moreover, the loss of collagen periodicity in eroded dentin could be attributed to increased enzymatic activity.[58] The spaces between the collagen fibrils are occupied by a large amount of water.

Silver nitrate infiltration of the bonding interface reflected the presence of water-rich zones, revealing inconsistent resin infiltration of the demineralized collagen in both substrates. However, this was significantly more evident in eroded dentin. All the features led to the formation of a structurally imperfect and highly porous hybrid layer,[67] resulting in areas of hydrophilic predominance and demineralized zones with collagen fibrils that were incompletely encapsulated by resin monomers.[43,44] This could contribute to the lower bond strength to eroded dentin.

Thus, we hypothesized that the large volume of water associated with deeply demineralized dentin in the resin-eroded dentin interface could contribute to lower μTBS and DC, as well as increased NL, regardless of the adhesive strategy.

In contrast, regardless of the adhesive, the application of an additional primer containing DMSO promoted an increase in the adhesive properties in both sound and eroded dentin substrates. DMSO has the ability to dissociate the highly cross-linked collagen into a sparse network of apparent fibrils[70] by suppressing the interpeptide hydrogen bonding.[21] Additionally, it can enhance the wetting of collagen by adhesives, allowing increased adhesive infiltration into the exposed collagen matrix.

Thus, improving the encasement of collagen within the polymerized matrix can permit hydrogen to bond with proteins,[69] thereby preventing the collapse of collagen fibrils, which destroys the interfibrillar spaces that serve as diffusion channels.[28] All these features permit the adhesive to cover and wet the entire surface, in addition to the deeper adhesive penetration. Hence, bond strengths increased significantly, so that the first null hypothesis could not be accepted. The second hypothesis was also rejected, as the application of an additional primer containing DMSO significantly reduced NL for both adhesives, reducing the extension of the porous, water-rich, and poorly impregnated resin layer below the hybrid layer.

This improvement can be attributed to several factors related to their biomodification properties. DMSO is a polar aprotic solvent that dissolves both polar and nonpolar compounds. It is a polyfunctional molecule with two hydrophobic CH_3 groups and a highly polar S=O group. The partial negative charge on the oxygen atom of the DMSO molecule favors the formation of two hydrogen bonds with water mol-

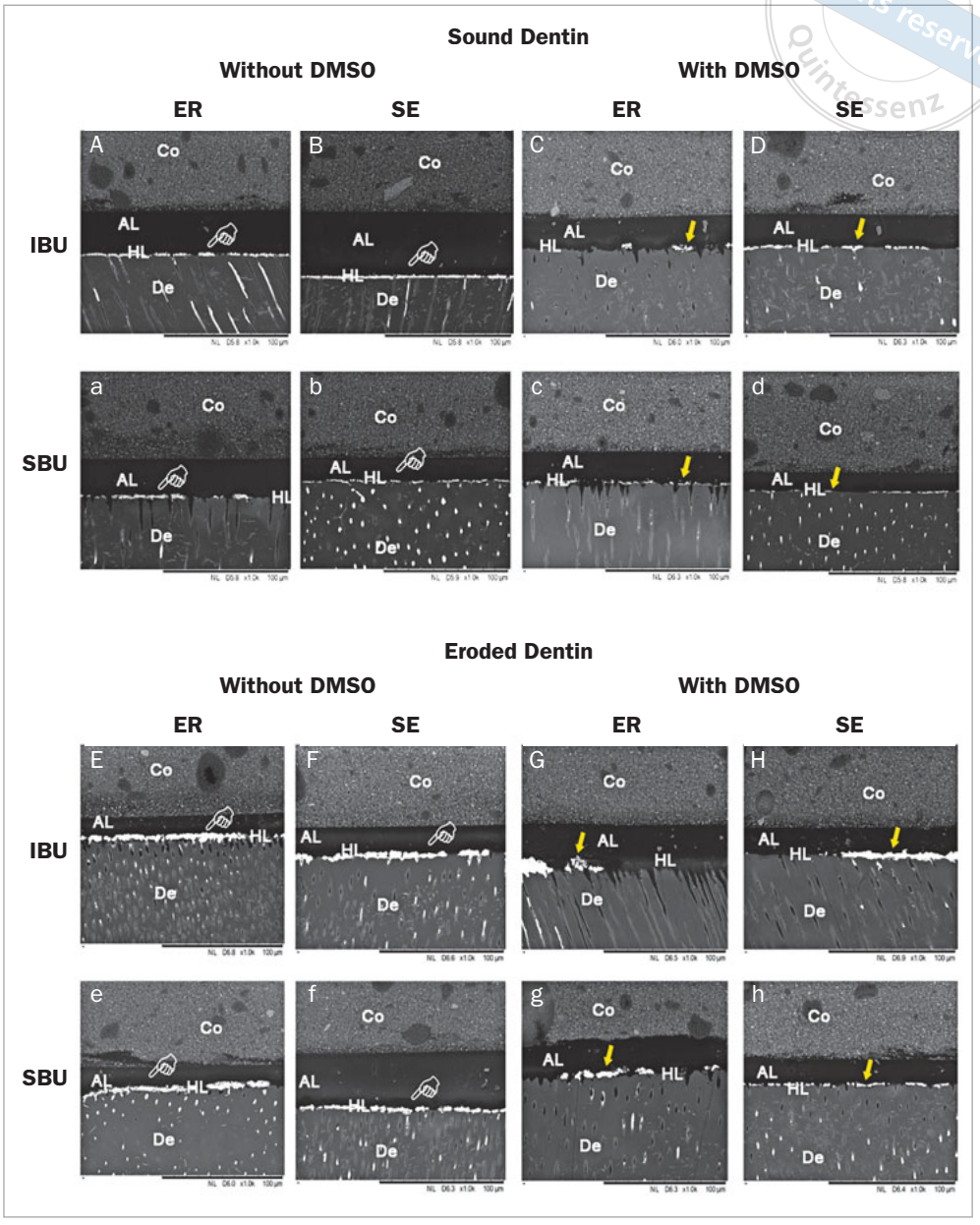

Fig 1 Representative backscattered SEM images for different experimental groups (1000X). Sound dentin exhibited some nanoleakage at the hybrid layer (white hands), regardless of the adhesive strategy used (A–D and a–d), as opposed to the eroded dentin (E–H and e–h). Groups treated with DMSO exhibited decreased silver nitrate uptake at the bottom of the hybrid layer in sound dentin (yellow arrows) (C, D, c, and d) and eroded dentin (yellow arrows) (G, H, g, and h), as opposed to the controls without DMSO (dimethyl sulfoxide).

ecules,[60] thereby reducing the self-associative tendency of water.[64]

A significant increase in DC was observed after applying DMSO onto eroded and sound dentin, leading us to reject the third null hypothesis as well. This may be explained by the fact that DMSO increased the infiltration of resinous monomers and decreased the self-associative tendency of water.[64] This could reduce the number of water molecules entrapped between the polymeric chains, increasing the degree of polymerization. In addition, DMSO decreased the termination rate of polymerization of methacrylate free radicals,[16] which can benefit the DC.

In this study, DMSO primer was applied for 60 s, which was the most frequently used duration in the literature.[53-55] However, some studies that used 30 s also had promising results.[41,60] To the best of our knowledge, no study has evaluated the application time of DMSO, and this requires further investigation.

Regarding the evaluation of the universal adhesives, although no direct comparison between the universal adhesives was performed, there was evidence of better performance of SBU compared to IBU in sound and eroded dentin. IBU is an acetone-containing, HEMA-free universal adhesive. Acetone is frequently used in adhesives owing to

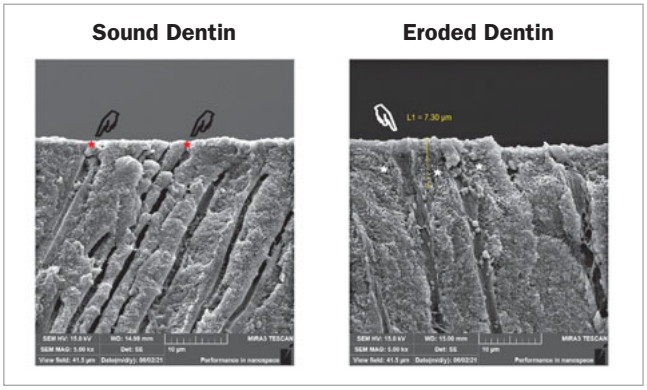

Fig 2 Representative SEM images of sound and eroded dentin. Sound dentin showed a smear layer, which covered the lumen (red asterisk) of the tubules (black hands). Eroded dentin showed a decalcified organic matrix with enlarged lumen tubules (white hands) and a collagen fibril layer that was exposed (white asterisk) at approximately 5.0 ± 2.0 mm of thickness.

its higher vapor pressure compared to that of ethanol and/or water.[31,32] However, the rapid evaporation of acetone increases the concentration of monomers in the adhesive,[29] thereby lowering the vapor pressure of the remaining residual solvents and making it more difficult to evaporate all the acetone added.[29,61]

According to Cho and Dickens,[5] a higher acetone content in the adhesives resulted in the formation of pores in the cured adhesive interface. This was more pronounced in HEMA-free adhesives, such as IBU. Thus, the rapid evaporation of acetone might not allow sufficient time for the monomers to adequately infiltrate the dentin and promote the formation of a uniform hybrid layer,[36,61] despite the DMSO pretreatment.

Although the exact composition of each universal adhesive evaluated was proprietary information, the results of this study demonstrated no significant difference between the ER and SE strategies regardless of the dentin substrate and adhesive used. The adhesives were consistent with the universal concept behind the adhesives.[6,12,50] Specifically, the adhesive properties tested were not compromised by the adhesive strategies applied to both substrates.

Universal adhesives were developed to simplify both the existing adhesive strategies (one-step SE and two-step ER adhesives), allowing the application of each strategy according to the clinician's preference or clinical demand.[34,65] However, it is worth mentioning that when applied as an ER strategy, it is necessary to use an additional step (etching with phosphoric acid), which contradicts the clinician's preference for simplification.

Additionally, the ER strategy requires pre-etching with phosphoric acid to demineralize the 6- to 9-mm surface of intertubular dentin and create porosities within the underlying collagen-fiber matrix, which is associated with erosive demineralization. This phenomenon can cause deeper demineralization of intertubular and peritubular dentin,[18,35] which challenges complete infiltration by the universal adhesive used in ER strategy.

Nevertheless, there is no consensus in the literature about which mode of application (ER or SE strategies) is the most suitable when using universal adhesives in eroded dentin. In different in vitro studies, no significant differences were found when the two strategies were compared.[2,12,14,51] Siqueira et al[51] evaluated several commercially available universal adhesives using SE and ER strategies in eroded dentin. Their results showed that the performance of the universal adhesives did not depend on the adhesive strategy used. Therefore, SE mode may be preferable, as it is more user-friendly (shorter application time, fewer steps) and less technique sensitive (no wet bonding, simple drying) than the ER strategy.[62] Nevertheless, further long-term studies are necessary to confirm these results.

Some important factors related to the biological properties and application time of DMSO should be mentioned. The cytotoxicity of DMSO is not the main biological concern, because DMSO is classified as a class-3 solvent, that is, it is the least toxic and poses the lowest risk to human health.[10] Indeed, Hebling et al[20] showed that DMSO did not evoke any substantial cytotoxic effects in direct contact with pulp tissue. However, there is a possibility that DMSO can carry cytotoxic and adhesive-monomer components toward the pulp; this should be taken into account, especially in deep cavities, as DMSO can increase the cytotoxicity of the adhesive based on the monomers present in the adhesive.[42]

One of the limitations of this study was the use of artificial saliva instead of natural saliva. Although natural saliva is problematic in controlling cross infection (when not stored in disposable containers) and may undergo compositional changes, resulting in decreased buffer capacity[17] and quick decomposition.[45] Only natural saliva exhibits salivary proteins that bind to calcium. However, although artificial saliva contains electrolytes similar to that of natural saliva, it lacks salivary proteins, promoting a high degree of supersaturation that is prone to precipitate excessive amounts of calcium phosphate.[48] Therefore, future studies need to be conducted to evaluate how the type of saliva can influence results such as those observed in this study.

Finally, although this study demonstrated the beneficial effect of DMSO pretreatment in improving the bonding properties of eroded dentin, it only evaluated the immediate bonding properties. Therefore, further in vitro and in vivo studies are necessary to determine whether DMSO can result in good performance after long-term storage.

CONCLUSION

This study provided evidence that eroded dentin is a more difficult substrate for bonding than sound dentin when using universal adhesives. However, a potential alternative to improve the bonding performance of universal adhesives is pretreatment with DMSO.

ACKNOWLEDGMENTS

This study was performed by Anna Szesz as partial fulfillment of her fellow degree at the State University of Ponta Grossa (UEPG), Ponta Grossa, PR, Brazil. This study was partially supported by the State Foundation of Support to Research, Scientific and Technological Development of Maranhao (FAPEMA) from the State Government of Maranhao Brazil, under grants 002/2019, the National Council for Scientific and Technological Development (CNPq) under grants 303332/2017-4 and 308286/2019-7, and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) - Finance Code 001. The authors are grateful for the technical support of the interdisciplinary laboratory CLABMU/UEPG.

REFERENCES

1. Assuncao CM, Dos Santos NM, Essvein TE, Silva MGR, Erhardt MCG, Rodrigues JA. Microshear bond strength of adhesive systems on eroded primary enamel and dentin. Pediatr Dent 2020;42:47–52.
2. Augusto MG, Torres C, Pucci CR, Schlueter N, Borges AB. Bond Stability of a universal adhesive system to eroded/abraded dentin after deproteinization. Oper Dent 2018;43:291–300.
3. Breschi L, Gobbi P, Mazzotti G, Falconi M, Ellis TH, Stangel I. High resolution SEM evaluation of dentin etched with maleic and citric acid. Dent Mater 2002;18:26–35.
4. Chen C, Niu LN, Xie H, Zhang ZY, Zhou LQ, Jiao K, Chen JH, Pashley DH, Tay FR. Bonding of universal adhesives to dentine–Old wine in new bottles? J Dent 2015;43:525–536.
5. Cho BH, Dickens SH. Effects of the acetone content of single solution dentin bonding agents on the adhesive layer thickness and the microtensile bond strength. Dent Mater 2004;20:107–115.
6. Cruz JB, Bonini G, Lenzi TL, Imparato JC, Raggio DP. Bonding stability of adhesive systems to eroded dentin. Braz Oral Res 2015;29.
7. De Munck J, Mine A, Poitevin A, Van Ende A, Cardoso MV, Van Landuyt KL, Peumans M, Van Meerbeek B. Meta-analytical review of parameters involved in dentin bonding. J Dent Res 2012;91:351–357.
8. de Siqueira FSF, Hilgemberg B, Araujo LCR, Hass V, Bandeca MC, Gomes JC, Reis A, Loguercio AD, Cardenas AFM. Improving bonding to eroded dentin by using collagen cross-linking agents: 2 years of water storage. Clin Oral Investig 2020;24:809–822.
9. Deyhle H, Bunk O, Muller B. Nanostructure of healthy and caries-affected human teeth. Nanomedicine 2011;7:694–701.
10. European Medicines Agency. ICH guideline Q3C (R6) on impurities: guideline for residual solvents – Step 5. 2019;2021.
11. Flury S, Koch T, Peutzfeldt A, Lussi A, Ganss C. The effect of a tin-containing fluoride mouth rinse on the bond between resin composite and erosively demineralised dentin. Clin Oral Investig 2013;17:217–225.
12. Forgerini TV, Ribeiro JF, Rocha RO, Soares FZ, Lenzi TL. Role of etching mode on bonding longevity of a universal adhesive to eroded dentin. J Adhes Dent 2017;19:69–75.
13. Francisconi-dos-Rios LF, Casas-Apayco LC, Calabria MP, Francisconi PA, Borges AF, Wang L. Role of chlorhexidine in bond strength to artificially eroded dentin over time. J Adhes Dent 2015;17:133–139.
14. Frattes FC, Augusto MG, Torres CRG, Pucci CR, Borges AB. Bond strength to eroded enamel and dentin using a universal adhesive system. J Adhes Dent 2017;19:121–127.
15. Geurtsen W, Lehmann F, Spahl W, Leyhausen G. Cytotoxicity of 35 dental resin composite monomers/additives in permanent 3T3 and three human primary fibroblast cultures. J Biomed Mater Res 1998;41:474–480.
16. Gupta SN, Nandi US. Role of dimethyl sulfoxide as a solvent for vinyl polymerization. J Polymer Sci Part A-1 Polymer Chem 1970;8:1493–1501.
17. Hall AF, Buchanan CA, Millett DT, Creanor SL, Strang R, Foye RH. The effect of saliva on enamel and dentine erosion. J Dent 1999;27:333–339.
18. Hashimoto M, Ohno H, Endo K, Kaga M, Sano H, Oguchi H. The effect of hybrid layer thickness on bond strength: demineralized dentin zone of the hybrid layer. Dent Mater 2000;16:406–411.
19. Hass V, Dobrovolski M, Zander-Grande C, Martins GC, Gordillo LA, Rodrigues Accorinte Mde L, et al. Correlation between degree of conversion, resin-dentin bond strength and nanoleakage of simplified etch-and-rinse adhesives. Dent Mater 2013;29:921–928.
20. Hebling J, Bianchi L, Basso FG, Scheffel DL, Soares DG, Carrilho MR, Pashley DH, Tjäderhane L, de Souza Costa CA. Cytotoxicity of dimethyl sulfoxide (DMSO) in direct contact with odontoblast-like cells. Dent Mater 2015;31:399–405.
21. Hirshburg J, Choi B, Nelson JS, Yeh AT. Collagen solubility correlates with skin optical clearing. J Biomed Opt 2006;11:040501.
22. Kenshima S, Reis A, Uceda-Gomez N, Tancredo Lde L, Filho LE, Nogueira FN, Loguercio AD Effect of smear layer thickness and pH of self-etching adhesive systems on the bond strength and gap formation to dentin. J Adhes Dent 2005;7:117–126.
23. Kreulen CM, Van't Spijker A, Rodriguez JM, Bronkhorst EM, Creugers NH, Bartlett DW. Systematic review of the prevalence of tooth wear in children and adolescents. Caries Res 2010;44:151–159.
24. Larsen MJ. Chemical events during tooth dissolution. J Dent Res 1990; 69(Spec No):575–580; discussion 634–576.
25. Lussi A, Schlueter N, Rakhmatullina E, Ganss C. Dental erosion – an overview with emphasis on chemical and histopathological aspects. Caries Res 2011;45(suppl 1):2–12.
26. Magalhaes AC, Levy FM, Souza BM, Cardoso CA, Cassiano LP, Pessan JP, Buzalaf MAR. Inhibition of tooth erosion by milk containing different fluoride concentrations: an in vitro study. J Dent 2014;42:498–502.
27. Marren K. Dimethyl sulfoxide: an effective penetration enhancer for topical administration of NSAIDs. Phys Sportsmed 2011;39:75–82.
28. Mehtala P, Pashley DH, Tjaderhane L. Effect of dimethyl sulfoxide on dentin collagen. Dent Mater 2017;33:915–922.
29. Miettinen VM, Vallittu PK, Docent DT. Water sorption and solubility of glass fiber-reinforced denture polymethyl methacrylate resin. J Prosthet Dent 1997;77:531–534.
30. Munoz MA, Luque I, Hass V, Reis A, Loguercio AD, Bombarda NH. Immediate bonding properties of universal adhesives to dentine. J Dent 2013; 41:404–411.
31. Nunes TG, Garcia FC, Osorio R, Carvalho R, Toledano M. Polymerization efficacy of simplified adhesive systems studied by NMR and MRI techniques. Dent Mater 2006;22:963–972.
32. Pashley EL, Zhang Y, Lockwood PE, Rueggeberg FA, Pashley DH. Effects of HEMA on water evaporation from water-HEMA mixtures. Dent Mater 1998;14:6–10.
33. Perdigao J, Geraldeli S, Carmo AR, Dutra HR. In vivo influence of residual moisture on microtensile bond strengths of one-bottle adhesives. J Esthet Restor Dent 2002;14:31–38.
34. Perdigao J, Kose C, Mena-Serrano AP, De Paula EA, Tay LY, Reis A, Loguercio AD. A new universal simplified adhesive: 18-month clinical evaluation. Oper Dent 2014;39:113–127.
35. Perdigão J, Lopes M. The effect of etching time on dentin demineralization. Quintessence International 2001;32:19–26.
36. Platt JA, Almeida J, Gonzalez-Cabezas C, Rhodes B, Moore BK. The effect of double adhesive application on the shear bond strength to dentin of compomers using three one-bottle adhesive systems. Oper Dent 2001; 26:313–317.
37. Prati C, Montebugnoli L, Suppa P, Valdre G, Mongiorgi R. Permeability and morphology of dentin after erosion induced by acidic drinks. J Periodontol 2003;74:428–436.
38. Reis A, Grande RH, Oliveira GM, Lopes GC, Loguercio AD. A 2-year evaluation of moisture on microtensile bond strength and nanoleakage. Dent Mater 2007;23:862–870.
39. Reis A, Loguercio AD, Azevedo CL, de Carvalho RM, da Julio Singer M, Grande RH. Moisture spectrum of demineralized dentin for adhesive systems with different solvent bases. J Adhes Dent 2003;5:183–192.
40. Salas MM, Nascimento GG, Huysmans MC, Demarco FF. Estimated prevalence of erosive tooth wear in permanent teeth of children and adolescents: an epidemiological systematic review and meta-regression analysis. J Dent 2015;43:42–50.

41. Salim Al-Ani AA, Mutluay M, Stape THS, Tjäderhane L, Tezvergil-Mutluay A. Effect of various dimethyl sulfoxide concentrations on the durability of dentin bonding and hybrid layer quality. Dent Mater J 2018;37:501–505.
42. Salim Al-Ani AAS, Salim IA, Seseogullari-Dirihan R, Mutluay M, Tjäderhane L, Tezvergil-Mutluay A. Incorporation of dimethyl sulfoxide into experimental hydrophilic and hydrophobic adhesive resins: evaluation of cytotoxic activities. Eur J Oral Sci 2021;129:e12756.
43. Sano H, Shono T, Takatsu T, Hosoda H. Microporous dentin zone beneath resin-impregnated layer. Oper Dent 1994;19:59–64.
44. Sano H, Takatsu T, Ciucchi B, Horner JA, Matthews WG, Pashley DH. Nanoleakage: leakage within the hybrid layer. Oper Dent 1995;20:18–25.
45. Schipper RG, Silletti E, Vingerhoeds MH. Saliva as research material: biochemical, physicochemical and practical aspects. Arch Oral Biol 2007;52: 1114–1135.
46. Schlueter N, Hara A, Shellis RP, Ganss C. Methods for the measurement and characterization of erosion in enamel and dentine. Caries Res 2011; 45 Suppl 1:13–23.
47. Schneider CA, Rasband WS, Eliceiri KW. NIH Image to ImageJ: 25 years of image analysis. Nat Methods 2012;9:671–675.
48. Shellis RP, Ganss C, Ren Y, Zero DT, Lussi A. Methodology and models in erosion research: discussion and conclusions. Caries Res 2011;45 (suppl 1):69–77.
49. Siqueira F, Cardenas A, Gomes GM, Chibinski AC, Gomes O, Bandeca MC, Loguercio AD, Gomes JC.. Three-year effects of deproteinization on the in vitro durability of resin/dentin-eroded interfaces. Oper Dent 2018;43:60–70.
50. Siqueira F, Cardenas AM, Gutierrez MF, Malaquias P, Hass V, Reis A, Loguercio AD, Perdigão J. Laboratory performance of universal adhesive systems for luting CAD/CAM restorative materials. J Adhes Dent 2016; 18:331–340.
51. Siqueira FSF, Cardenas AM, Ocampo JB, Hass V, Bandeca MC, Gomes JC, Reis A, Loguercio AD. Bonding performance of universal adhesives to eroded dentin. J Adhes Dent 2018;20:121–132.
52. Stape TH, Tezvergil-Mutluay A, Mutluay MM, Martins LR, do Prado RL, Pizi EC, Tjäderhane L. Influence of dimethyl sulfoxide used as a solvent on the physical properties and long-term dentin bonding of hydrophilic resins. J Mech Behav Biomed Mater 2016;64:220–228.
53. Stape TH, Tjäderhane L, Marques MR, Aguiar FH, Martins LR. Effect of dimethyl sulfoxide wet-bonding technique on hybrid layer quality and dentin bond strength. Dent Mater 2015;31:676–683.
54. Stape TH, Tjäderhane L, Tezvergil-Mutluay A, Yanikian CR, Szesz AL, Loguercio AD, Martins LRM. Dentin bond optimization using the dimethyl sulfoxide-wet bonding strategy: A 2-year in vitro study. Dent Mater 2016;32: 1472–1481.
55. Stape THS, Mutluay MM, Tjäderhane L, Uurasjarvi E, Koistinen A, Tezvergil-Mutluay A. The pursuit of resin-dentin bond durability: Simultaneous enhancement of collagen structure and polymer network formation in hybrid layers. Dent Mater 2021.
56. Taji S, Seow WK. A literature review of dental erosion in children. Aust Dent J 2010;55:358–367; quiz 475.
57. Tay FR, Pashley DH, Suh BI, Carvalho RM, Itthagarun A. Single-step adhesives are permeable membranes. J Dent 2002;30:371–382.
58. Tjäderhane L, Buzalaf MA, Carrilho M, Chaussain C. Matrix metalloproteinases and other matrix proteinases in relation to cariology: the era of 'dentin degradomics'. Caries Res 2015;49:193–208.
59. Tjäderhane L, Larjava H, Sorsa T, Uitto VJ, Larmas M, Salo T. The activation and function of host matrix metalloproteinases in dentin matrix breakdown in caries lesions. J Dent Res 1998;77:1622–1629.
60. Tjäderhane L, Mehtala P, Scaffa P, Vidal C, Paakkonen V, Breschi L, Hebling J, Tay FR, Nascimento FD, Pashley DH, Carrilho MR. The effect of dimethyl sulfoxide (DMSO) on dentin bonding and nanoleakage of etch-and-rinse adhesives. Dent Mater 2013;29:1055–1062.
61. Van Landuyt KL, Snauwaert J, Peumans M, De Munck J, Lambrechts P, Van Meerbeek B. The role of HEMA in one-step self-etch adhesives. Dent Mater 2008;24:1412–1419.
62. Van Meerbeek B, Yoshihara K, Yoshida Y, Mine A, De Munck J, Van Landuyt KL. State of the art of self-etch adhesives. Dent Mater 2011; 27:17–28.
63. Vishnyakov A, Laaksonen A, Widmalm G. Molecular dynamics simulations of alpha-D-Manp-(1–>3)-beta-D-Glcp-OMe in methanol and in dimethyl sulfoxide solutions. J Mol Graph Model 2001;19:338–342, 396–337.
64. Vishnyakov A, Lyubartsev AP, Laaksonen A. Molecular dynamics simulations of dimethyl sulfoxide and dimethyl sulfoxide–water mixture. J Phys Chem A 2001;105:1702–1710.
65. Wagner A, Wendler M, Petschelt A, Belli R, Lohbauer U. Bonding performance of universal adhesives in different etching modes. J Dent 2014; 42:800–807.
66. Wang R, Li Y, Hass V, Peng Z, Wang Y. Methacrylate-functionalized proanthocyanidins as novel polymerizable collagen cross-linkers – Part 2: Effects on polymerization, microhardness and leaching of adhesives. Dent Mater 2021;37:1193–1201.
67. Wang Y, Spencer P. Effect of acid etching time and technique on interfacial characteristics of the adhesive-dentin bond using differential staining. Eur J Oral Sci 2004;112:293–299.
68. Zarella BL, Cardoso CA, Pela VT, Kato MT, Tjäderhane L, Buzalaf MA. The role of matrix metalloproteinases and cysteine-cathepsins on the progression of dentine erosion. Arch Oral Biol 2015;60:1340–1345.
69. Zheng Y-J, Ornstein RL. A molecular dynamics and quantum mechanics analysis of the effect of DMSO on enzyme structure and dynamics: subtilisin. J Am Chem Soc 1996;118:4175–4180.
70. Zimmerley M, McClure RA, Choi B, Potma EO. Following dimethyl sulfoxide skin optical clearing dynamics with quantitative nonlinear multimodal microscopy. Appl Opt 2009;48:D79–87.
71. Zimmerli B, De Munck J, Lussi A, Lambrechts P, Van Meerbeek B. Long-term bonding to eroded dentin requires superficial bur preparation. Clin Oral Investig 2012;16:1451–1461.

Clinical relevance: Pretreatment with DMSO is a potential alternative to improve the bonding performance of universal adhesives in sound and eroded dentins.