Journal of
Kerman University of Medical Sciences

https://jkmu.kmu.ac.ir
10.34172/jkmu.3941
JKMU. 2025;32:3941

Original Article

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Microtensile Bond Strength Between Composite Resin and Discolored Dentin After Amalgam Replacement Using Different Universal Adhesives

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Abstract
Background: It is common to replace amalgam with composite resins for different reasons. Changes in the dentin substrate after amalgam restorations might affect the bond strength between composite resin and dentin. This study evaluated the microtensile bond strength of composite resin to discolored dentin following amalgam replacement with universal adhesives.
Methods: In this *in vitro* study, thirty-two sound human premolar teeth were collected. After preparing classic class I amalgam cavities measuring 2 mm in depth, 3 mm mesiodistally, and 2.4 mm buccolingually, half of the samples were randomly selected and restored with high-copper amalgam and underwent a thermocycling procedure (5000 cycles). They were stored at 37 ºC and 100% humidity for six months to form amalgam corrosion products. After removing the amalgam, each half was divided into four subgroups based on adhesive type (G-Premio Bond/All-Bond Universal) and bonding technique (self-etch/etch-and-rinse). Then, all samples were restored with composite resin. The microtensile bond strength was calculated, and data analysis was conducted using SPSS 25 and *t*-tests.
Results: The bond strength values in the amalgam-affected subgroups were significantly ($P<0.05$) lower than normal dentin. In all groups, the bond strength of the All-Bond Universal was higher than that of the G-Premio Bond. However, the bond strength in the etch-and-rinse technique in normal dentin was significantly ($P<0.05$) higher than the self-etch technique. The bond strength in the amalgam-affected dentin subgroups with the All-Bond Universal was comparable to the bond strength of this adhesive to normal dentin with the self-etch technique.
Conclusion: All-Bond Universal is suggested for both self-etch and etch-and-rinse techniques in amalgam replacement with composite resin. However, the self-etch technique is recommended if the G-Premio Bond is used.
Keywords: Dentin, Composite resins, Corrosion, Dental amalgam, Light-cured dental bonding

Citation: Shadman N, Rostami S, Eskandarizadeh A, Elmamooz N, Pooyan M, Azizi Shoul M. Microtensile bond strength between composite resin and discolored dentin after amalgam replacement using different universal adhesives. *Journal of Kerman University of Medical Sciences*. 2025;32:3941. doi:10.34172/jkmu.3941

Received: April 27, 2024, **Accepted**: August 17, 2025, **ePublished**: August 27, 2025

Introduction

Dental amalgam has been used to restore posterior teeth, even in extensive cavities, since the 19th century (1,2). In some cases, it is necessary to replace amalgam restorations because of recurrent caries, fractures, or esthetic reasons, and composite resins would be a good choice (3,4). The microstructure of deep dentin might be different from the normal dentin because of demineralization, remineralization, sclerosis, or precipitation of amalgam corrosion products into the dentin substrate (5,6). Employing caries-disclosing dye has demonstrated that calcium ions (Ca) have been substituted by tin (Sn) and zinc (Zn) ions in the dentin located beneath amalgam restorations (7). Consequently, achieving a robust bond between composite resin and dentin presents significant challenges when replacing amalgam restorations. Numerous investigations analyzing the implications of amalgam corrosion byproducts have indicated that these byproducts exert no discernible influence on the bond strength of composite resins to the underlying dentin. Conversely, several other studies have indicated that these byproducts negatively impact the bond strength of composite resin (8-11). Advances in the composition of dentin adhesives have overcome some challenges and improved the predictability of bonding to dentin substrate. The use of universal adhesives in dentistry is increasing due to their ease of use, functional monomer content, the feasibility of creating a chemical bond to the tooth structure, and the possibility of application in various techniques, including self-etch, etch-and-rinse, and selective etch (12,13).

Ghavamnasiri et al reported a higher bond strength for etch-and-rinse adhesives than the self-etch type when using composite resin on discolored dentin after amalgam removal (14). Several studies have recommended the cavity wall extension and complete elimination of discoloration until normal dentin is achieved (14, 15). Abdelnabi et al suggested a slight increase of cavity depth up to 0.5 mm to decrease the detrimental effects of amalgam corrosion products on the composite resin bond strength (15).

Among different tests related to dental materials and the bond strength of composite to tooth structure, tensile bond strength tests yield more reliable results due to the more homogeneous distribution of stresses at the bonded interface. However, the difficulty of preparing the samples for tensile bond strength tests results in more commonly using shear bond strength tests (16).

Universal bonding agents can form chemical bonds with metallic ions with functional monomers, such as 10-MDP (12,13). Since the discolored dentin under the amalgam restorations contains metallic elements, using these adhesives will likely enhance the bond strength of composite resins to this dentin substrate. There are a few studies about the bond strength of universal adhesives to discolored dentin after removing amalgam. Therefore, this study was designed to determine the microtensile bond strength of composite resins to dentin following amalgam replacement after universal adhesives application (G-Premio Bond and All-Bond Universal) and different adhesion strategies (self-etch or etch-and-rinse).

The null hypothesis was that the microtensile bond strength of composite resin to normal dentin and discolored dentin beneath amalgam restorations is the same when different universal adhesives and bonding strategies are applied.

Methods

Thirty-two sound human premolar teeth extracted for other reasons were selected. After debridement and removing tissue remnants, the teeth were stored in 5.25% NaOCl (sodium hypochlorite) (NikDarman, Tehran, Iran) for 5 minutes for disinfection. A two-piece stainless steel cylindrical mold with a diameter and height of 2 cm was used for mounting the samples using self-curing acrylic resin (Acropars 200, Tehran, Iran). The anatomic crown of each tooth remained out of the resin.

Class I amalgam cavities with 2 mm depth, mesiodistal width of 3 mm, and buccolingual width of 2 mm were prepared in all teeth using a 1.2 mm diamond cylindrical bur (Teezkavan, Tehran, Iran) with a high-speed handpiece under air-water spray. After preparing the cavities, all the samples were rinsed using air-water spray and dried with a gentle air stream. The teeth were incubated at 37 °C and 100% humidity during the study period in a normal saline solution (Behdad Co, Tehran, Iran). The solution was refreshed weekly.

Half of the samples ($n = 16$) were selected randomly and restored with high-copper amalgam (Gs-80 SDI Limited, Australia), and the other half of the samples were stored until composite filling. Amalgam samples were incubated at 100% humidity and 37 ºC for six months, followed by a 5000-round thermocycling procedure at 5–55 ºC (dwell time of 20 seconds and transfer time of 5 seconds) (Vafaei Industrial Co., Tehran, Iran), which is equal to 6 months of clinical service, to form amalgam corrosion products (17).

A #010 diamond fissure bur (Teezkavan, Tehran, Iran) was used with a high-speed handpiece and air-water spray to carefully remove the main part of the amalgam restoration from the cavities. The last amalgam layer was removed with a blunt dental explorer to prevent damage to the tooth structure and avoid removing the amalgam corrosion products on the surface.

All the steps were carried out by the same operator. At this stage, 16 teeth with normal dentin and 16 teeth with dentin affected by amalgam were randomly allocated to 8 subgroups ($n = 4$) as follows: (Figure 1)

CAE subgroup (control subgroup): After dentin etching with phosphoric acid for 15 seconds and rinsing the excess water, it was removed by blot-drying the surface, keeping the visible moisture of the preparation surface. Then, two layers of All-Bond Universal adhesive were applied. The surface was scrubbed with a micro brush for 10–15 seconds, air-dried for 10 seconds, then light-cured for 10 seconds using an LED light-curing unit at 1100 mW/cm2 light intensity. The light intensity was checked using a radiometer (Kerr Corp., Demi Ultra, USA).

AAE subgroup: A procedure similar to the CAE group was carried out on the dentin affected by amalgam.

CAS subgroup (control subgroup): Two coats of All-Bond Universal adhesive were applied to normal dentin surfaces and light-cured, same as the CAE group.

AAS subgroup: A procedure similar to the CAS group was carried out on the dentin affected by amalgam.

CGE subgroup (control subgroup): After dentin etching with phosphoric acid for 10–15 seconds, rinsing for 5 seconds, and air drying, G-Premio Bond adhesive was applied to the tooth structure using a micro brush. It was left for 10 seconds after application, dried for 5 seconds using high air pressure, and light-cured for 10 seconds.

AGE subgroup: A procedure similar to the CGE group was carried out on the dentin affected by amalgam.

CGS subgroup (control subgroup): G-Premio Bond adhesive was applied on the tooth structure using a micro brush, left for 10 seconds after application, dried for 5 seconds using air pressure, and light-cured for 10 seconds.

AGS subgroup: A procedure similar to the CGS group was carried out on the dentin affected by amalgam.

Table 1 shows the chemical composition of the adhesives used in the study (Table 1).

All cavities were restored with Vit-l-essence composite with A5 shade to facilitate the identification of the composite resin-tooth interface using the incremental technique in two 1-mm-thick layers. Each layer was light-cured occlusally using an LED light-curing unit for 20 seconds at a light intensity of 1100 mW/cm^2. The samples were stored in a moist environment at 37 °C for 24 hours, followed by the microtensile bond strength test preparation. The samples were prepared in a rod shape with a cross-section of 1 × 1 mm and a height of 4 mm (2 mm of dentin and 2 mm of composite) using an Isomet cutting instrument (Buchler Isomet Spectrographic Led, Low-Speed Saw, USA). Each tooth provided a mean of three rods (an average of 12 rods in each group).

The rods were fixed in a microtensile test unit (MicroTensile Tester, Bisco, USA) with cyanoacrylate to determine the microtensile bond strength. The dentin-composite resin interface was placed in the middle, and after calibration, a tensile force was applied to both sides of the rods at the speed of 1 mm/min until fracture happened.

Statistical Analysis

Data analysis was conducted by SPSS 25. The Kolmogorov-Smirnov test was used to check the normality distribution. Data distribution followed the normal curve ($P > 0.05$). In addition, Levene's test was used to analyze the homogeneity of bond strength variances in different groups. Due to sufficient evidence to claim that the variances are not equal, corrected freedom degrees were used. The three-way analysis of variance was applied to check the significance of the interaction between subgroups, and Tukey's post hoc test was used to compare the subgroups pairwise. Fisher's exact test was used to compare the frequency of favorable and unfavorable fractures across groups, with statistical significance set at $P < 0.05$.

Results

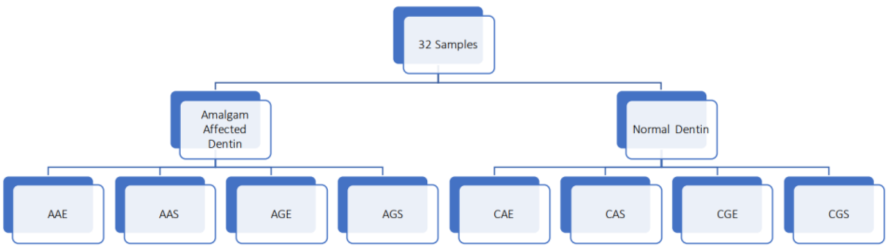
Table 2 shows the means and standard deviations of microtensile bond strengths in the study subgroups. The highest (33.14 MPa) and lowest (11.11 MPa) bond strengths were seen in the CAE and CGS groups, respectively (Table 2).

The findings derived from the three-way analysis of variance indicated that the main effects of Restoration type, adhesive, and bonding strategy were statistically significant ($P < 0.001$). The microtensile bond strength exhibited a markedly higher value in the control group compared to the Amalgam group. At the same time, All Bond Universal demonstrated a significantly superior bond strength compared to the G-Premio Bond, and the Etch-and-rinse technique showed a significantly elevated bond strength compared to Self-etch ($P < 0.001$).

No significant interaction was observed between the restoration type and the bonding agent ($P = 0.557$), nor was there a notable interaction between the bonding agent

Table 1. The chemical compositions of the material used in the study

Material	Manufacturer	Composition	pH
G-Premio Bond	GC, Tokyo, Japan	10-MDP (5–10%), methacrylate (10–20%), 4-MET, photoinitiator (1–5%), butylated hydroxytoluene (<1%), water (24%), acetone (25–50%), Silica	1.5
All-Bond Universal	Bisco, Schaumburg, USA	phosphoric acid ester monomer, 10-MDP, Bis GMA, ethanol, HEMA, photoinitiator	3.2
Vit-l-escence composite (micro hybrid)	Ultradent Products Inc., South Jordan, UT, USA	BisGMA & diluents, barium aluminoborosilicate (0.4–0.6 µm) including silica filler particles 0.04 to 0.1 µm and other fillers (average size 0.7 µm),75Wt%, 52Vol%, an amine co-initiator, camphorquinone, and a proprietary initiator	Not determined
Amalgam (Gs-80 Admixed)	SDI Limited Dental Alloy Products, Australia	40% silver, 31.3% tin, 28.7% copper, mercury ratio 1:0.9 (47.4% Hg)	Not available
Etchant (Ultra-Etch™ & Opal™ Etch)	Ultradent Products Inc., South Jordan, UT, USA	35% phosphoric acid, polyethylene glycol, aluminum oxide, dark blue pigment, cobalt zinc aluminate blue spinel, cobalt titanate green spinel	<1



A: amalgam (affected dentin) C: control (normal dentin)
A: All-Bond Universal G: G-Premio Bond
E: etch-and-rinse S: self-etch

Figure 1. Study group allocation

and the bonding technique ($P=0.543$).

The interaction between the restoration type and the bonding technique was significant ($P<0.001$). Within the control group, the microtensile bond strength achieved via the Etch-and-rinse method was significantly greater than that obtained by Self-etch ($P<0.001$). Conversely, in the Amalgam group, no statistically significant difference based on the bonding technique was identified ($P<0.001$) (Table 3).

The interaction among restoration type, bonding agent, and bonding technique was statistically significant ($P=0.007$). Given this, a Tukey's post hoc test was conducted for pairwise comparisons among the subgroups (Table 4).

The results showed that the microtensile bond strength of composite to discolored dentin was significantly lower than that of normal dentin. Compared to the G-Premio Bond adhesive, the bond strength values of the All-Bond Universal adhesive in the etch-and-rinse techniques to discolored dentin and the self-etch technique to normal dentin were significantly higher. The bond strength values of both adhesives to normal dentin in the etch-and-rinse technique were significantly higher than in the self-etch technique. The bond strengths with the etch-and-rinse and self-etch techniques to discolored dentin were not significantly different.

Table 2. The means and standard deviations of microtensile bond strength values in the samples in each subgroup in MPa

Restoration type	Bonding agent	Bonding strategy	*N*	M ± SD
Amalgam	G-Premio Bond	Etch-and-rinse	10	12.82 ± 5.01
		Self-etch	10	15.69 ± 3.72
	All Bond Universal	Etch-and-rinse	10	22.91 ± 7.73
		Self-etch	10	20.48 ± 5.45
Control	G-Premio Bond	Etch-and-rinse	10	28.39 ± 6.68
		Self-etch	10	11.11 ± 3.01
	All Bond Universal	Etch-and-rinse	10	33.14 ± 6.44
		Self-etch	10	24.11 ± 3.47

Discussion

The results of our study show that using different types of universal adhesives and different adhesion strategies can affect the bond strength of composite to discolored dentin following the removal of amalgam restorations. The microtensile bond strength values of composite to discolored dentin were significantly lower than normal. Compared to G-Premio Bond adhesive, All-Bond Universal adhesive had significantly higher microtensile bond strength than discolored dentin in the etch-and-rinse technique and normal dentin in the self-etch technique. Thus, the null hypothesis was rejected to some extent.

In some cases, defective amalgam restorations can be repaired. This is a better and less invasive choice than replacing the whole restoration as it reduces the risk of pulpal injury and preserves the tooth structure (18).

Table 3. Three-way analysis of variance to compare microtensile bond strength values (MPa) based on restoration type, bonding agent, and bonding technique

Source	*F (pdf)*	*P*	*η2*
Restoration type	$F\ (1,72)=26.21^{**}$	< 0.001	0.267
Bonding agent	$F\ (1,72)=45.19^{**}$	< 0.001	0.386
Bonding technique	$F\ (1,72)=28.36^{**}$	< 0.001	0.283
Restoration type * Bonding agent	$F\ (1,72)=0.35$	0.557	0.005
Restoration type * Bonding technique	$F\ (1,72)=30.36^{**}$	< 0.001	0.297
Bonding agent * Bonding technique	$F\ (1,72)=0.37$	0.543	0.005
Restoration type * Bonding agent * Bonding technique	$F\ (1,72)=7.79^{**}$	0.007	0.098

Dependent variable: μTBS(Mpa)
**$p<0.01$

Table 4. The results of Tukey's post hoc test to compare two by two microtensile values of samples in each subgroup in terms of MPa

The compared groups		Mean difference (I-J)	*Std. error*	*P*
Control / G-Premio / etch-and-rinse (CGE)	Amalgam / G-Premio / etch-and-rinse (AGT)	15.57	2.43	< 0.001
Control / G-Premio / self-etch (CGS)	Amalgam / G-Premio / self-etch (AGS)	-4.57	2.43	0.565
Control / All Bond / etch-and-rinse (CAE)	Amalgam / All Bond / etch-and-rinse (ABT)	10.23**	2.43	0.002
Control / All Bond / self-etch (CAS)	Amalgam / All Bond / self-etch (ABS)	3.6	2.43	0.807
			2.43	
Amalgam / All Bond / etch-and-rinse (AAE)	Amalgam / G-Premio / etch-and-rinse (AGT)	10.09**	2.43	0.002
Amalgam / All Bond / self-etch (AAS)	Amalgam / G-Premio / self-etch (AGS)	4.80	2.43	0.505
Control / All Bond / etch-and-rinse (CAE)	Control / G-Premio / etch-and-rinse (CGT)	4.47	2.43	0.519
Control / All Bond / self-etch (CAS)	Control / G-Premio / self-etch (CGS)	13.00**	2.43	< 0.001
			2.43	
Amalgam / G-Premio / self-etch (AGS)	Amalgam / G-Premio / etch-and-rinse (AGT)	2.88	2.43	0.935
Control / G-Premio / self-etch (CGS)	Control / G-Premio / etch-and-rinse (CGT)	17.28**	2.43	< 0.001
Amalgam / All Bond / self-etch (AAS)	Amalgam / All Bond / etch-and-rinse (ABT)	-2.42	2.43	0.973
Control / All Bond / self-etch (CAS)	Control / All Bond / etch-and-rinse (CBT)	-9.02**	2.43	0.009

However, in some cases, for reasons such as esthetics, the amalgam restoration should be replaced entirely with a tooth-colored material, such as composite resin (3,4). Therefore, using different universal adhesives, this *in vitro* study was conducted to determine the microtensile bond strength of composite resin to amalgam-affected dentin.

The most common theory about the decreasing the bond strength of amalgam-affected dentin is the effect of amalgam corrosion products on dentinal proteins (19). Mandava et al reported that amalgam corrosion products precipitate pulpal proteins in the tubular fluid and reduce dentin permeability, which can be a reason for decreased bond strength (19). In addition, metallic compounds resulting from amalgam corrosion products can bind to collagen fibril surfaces and affect resin monomer polymerization (20).

It has been reported that Cu, Sn, and Zn particles can initiate polymerization in UDM(urethane-dimethacrylate)-based monomers in the absence of tertiary amine at room temperature because a small amount of released ionic metals can act as a reducing agent for redox polymerization (20). The initiation mechanism might depend on the number of metallic particles because it has been reported that a high amount of Zn ions tends to delay the setting reactions of resin monomers (21).

On the other hand, these heavy metals decrease the acidic solubility of the smear layer. Also, phosphoric acid will have less effect on dentin containing these ions than on normal dentin. Therefore, the etchability of heavy metal-containing dentin decreases compared to the normal smear layer. Therefore, after removing the old amalgam restoration, the corrosion products will still exist, reducing the bond strength (19).

Mandava et al reported that the bond strength of cavities restored with amalgam was significantly lower compared to freshly prepared cavities. The microtensile bond strength test results with the etch-and-rinse technique to both dentin substrates were higher than the self-etch technique (19), which agrees with the results of the control group in this study.

Abdelnabi et al reported that amalgam corrosion products interfere with the bonding process; therefore, removing 0.5 mm of discolored dentin diminished these products and led to better bonding in self-etch and etch-and-rinse systems (15). Also, Ghavamnasiri et al suggested extending the cavities about 0.5 mm after removing the amalgam (14). This study detected similar microleakage after removing the discolored dentin under amalgam restorations and in samples with fresh composite resin restorations (14). Similar microtensile test results were reported by Moosavi et al. Their assay showed a significantly lower microtensile bond strength value in groups with amalgam-affected dentin than in normal dentin under composite restorations (22).

According to Fusayama's study, the hardness of dentin beneath old amalgam restorations ranged from 20.13 to 86.4 KHN. The 20.13 KHN value is at the hardness level of outer carious dentin, and the 84.6 KHN value is similar to the hardness of normal dentin. Therefore, the dentin affected by amalgam might be slightly softer than normal (23). However, in Harninattisai et al's study, the hardness of the affected dentin was not significantly different from normal dentin (9).

Redwan et al did not report a significant difference in the amount of microleakage between composite resin restorations placed on normal dentin and amalgam-affected dentin when the restorations were replaced. EDX analysis in the mentioned study did not detect metallic ions in the dentin beneath the amalgam; only calcium and phosphorus ions were detected in the samples. Although the metallic ions did not penetrate the discolored dentin compared to the fresh dentin, it showed higher microleakage, which was insignificant (11).

Alshehri et al showed that dentin contamination with amalgam has no significant effect on composite microleakage after amalgam replacement with composite (20).

In another study designed by Sachin Sunny (2019), there was no difference in microtensile bond strength and microleakage between normal and amalgam-affected dentin (21).

According to our study's results, the difference can be related to the study groups' allocation, different tests and analyses, and also different materials such as bonding agents.

In the current investigation, the microtensile bond strength of the specimens subjected to amalgam restoration with discolored dentin was inferior compared to the restorations utilizing normal dentin. Notably, the All-Bond Universal bonding agent, employed in etch-and-rinse and self-etch methodologies, produced total bond strength values analogous to those observed with normal dentin, specifically utilizing the self-etch technique. Consequently, this study revealed distinct functional disparities between the G-Premio Bond and All-Bond Universal bonding agents concerning the microtensile bond strength of the specimens. The maximum microtensile bond strength was recorded with the All-Bond Universal and G-Premio Bond adhesives applied to normal dentin through the etch-and-rinse technique. Nevertheless, the All-Bond Universal adhesive did not exhibit superiority over the G-Premio Bond within the etch-and-rinse modality. Conversely, in the context of the self-etch mode, the All-Bond Universal adhesive demonstrated a heightened bond strength on normal dentin.

Generally, universal adhesives differ in their components, such as cross-linking monomers, functional monomers, inhibitors, activators, solvents, the type and volume of fillers, etc., and also in their manufacturing techniques. Limited data are available on the shrinkage and hardness of these adhesives, which affects their physical and mechanical properties. Other factors affecting the bond strength test results are the application technique, differences between different operators, and insufficient time for drying the primer (24).

Additionally, there are particular differences between G-Premio Bond and All-Bond Universal bonding agents. G-Premio bond has 4-MET and 10-MDP as functional monomers and is a HEMA-free bonding agent. In contrast, 10-MDP is the only functional monomer in All-Bond Universal, and HEMA is a major component. Another difference is in acidic pH, which is 1.5 in the G-Premio bond and 3.2 in All-Bond Universal. Acidic pH plays an important role in adhesion by solving the mineral part of dentin to expose the collagenic scaffold. This acidity will be mostly buffered by dentin. However, in low-pH situations, it can put the calcium out of reach of functional monomers such as 10-MDP, leading to lower chemical adhesion to dentin (25).

Comba et al's study showed a higher microtensile bond strength in the etch-and-rinse than the self-etch mode with the All-Bond Universal adhesive to normal dentin, which is consistent with the present study (26).

It is worth mentioning that in *in vitro* conditions, many factors differ from clinical conditions, and it is impossible to imitate clinical conditions in the laboratory (27) completely. In this regard, the following factors can be mentioned: the impossibility of complete isolation in clinical conditions, changes in the viscosity of adhesives in clinical conditions due to their continuous use (28), and the extent of access of the tip of the light-curing system for efficient application (29).

The present study's samples underwent thermal cycles to simulate clinical conditions. Different aging processes have been used in previous studies, including immersing the samples in citric acid (30), normal saline solution (31), water storage at 37 °C for different times, and thermal cycles (32,33). Thermal cycles are hydraulic and thermal degradation (34) that simulate thermal disintegration through sudden temperature changes (35). It has been demonstrated that aging through 5000-round thermal cycles has more significant effects than immersing the samples in 3.5% citric acid for one week or immersion in boiling water for 8 hours (36).

When amalgam-filled teeth are used, the parameters affecting the test results increase. For example, the amalgam type used, the penetration depth of amalgam corrosion products, ensuring complete removal of caries, and the effect of caries on the dentin substrate are variables whose effects can be controlled if the conditions for producing corrosion products are matched in all the samples.

The limitations in this study include using limited material brands to be evaluated in laboratory circumstances. It would be helpful to design future clinical studies using other brands or to use different in-vitro study circumstances to mimic the oral environment, such as temperature alteration and chewing forces.

Conclusion

In amalgam replacement cases with composite resin, the use of All-Bond Universal in both etch-and-rinse and self-etch techniques is suggested. However, the self-etch technique is recommended if G-Premio Bond is used.

Acknowledgments

The authors thank Dr. Jahangir Mohammadi for the statistical analysis of the study.

Authors' Contribution

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Competing Interests

The authors declare that they do not have any conflict of interest.

Ethical Approval

The Research Ethics Committee of Kerman University of Medical Science, with the ethical code IR.KMU.REC.1399.349, approved the study.

Funding

This work was supported by Kerman University of Medical Sciences. The grant number is 98001124.

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