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An Evaluation of the Bond Strength of Universal Adhesives to Amalgam-Contaminated Dentin in Resin Composite Restorations

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ABSTRACT

Background and Objective: The substitution of amalgam restorations with composite resin has consistently been a matter of clinical concern, primarily due to the potential for dentin contamination by residual amalgam particles and the subsequent adverse effects on adhesive bond strength. This study aimed to evaluate the microshear bond strength of a universal adhesive to amalgam-contaminated dentin in composite resin restorations.

Methods: In this in vitro study, a total of 32 sound premolar teeth were randomly assigned to 8 experimental groups, with 4 teeth per group and 2 bonded specimens prepared from each tooth. The groups were defined as follows: G1 and G2 served as negative controls (healthy, non-contaminated dentin) using self-etch and etch-and-rinse adhesive application modes, respectively. G3 and G4 received contaminated dentin treated with self-etch and etch-and-rinse modes, respectively. G5 and G6 received contaminated dentin followed by application of 2% chlorhexidine prior to adhesive application in self-etch and etch-and-rinse modes, respectively. G7 and G8 received contaminated dentin followed by removal of a 0.5 mm layer of superficial dentin, again using self-etch and etch-and-rinse modes, respectively. After the occlusal enamel was removed, all groups except the control groups received amalgam restorations. The specimens were then subjected to thermocycling (10,000 cycles, temperatures ranging from 5°C to 55°C). Subsequently, the amalgam was removed, and a universal adhesive was applied to the dentin surfaces, followed by fabrication of composite resin cylinders. Microshear bond strength (µSBS) was measured at a crosshead speed of 0.5 mm/min after additional thermocycling, and failure patterns were examined using a stereomicroscope.

Findings: The highest mean bond strength values were observed in the control groups: G1 (self-etch) yielded 17.80±3.78 MPa, and G2 (etch-and-rinse) yielded 18.88±3.44 MPa. In contrast, the lowest values were recorded in the dentin removal groups: G7 (self-etch) at 12.89±3.45 MPa and G8 (etch-and-rinse) at 13.84±4.35 MPa. Statistical analysis revealed that the bond strengths of G1 and G2 were significantly superior to those of G4 (contaminated dentin, etch-and-rinse), G7, and G8. However, groups pretreated with 2% chlorhexidine (G5 and G6) did not demonstrate statistically significant differences compared to controls. Furthermore, no significant association was found between bond strength values and failure types, nor between failure patterns and the etching mode employed.

Conclusion: The findings of this study revealed that the universal adhesive system, when used in self-etch mode, produced bond strength results on amalgam-contaminated dentin that were similar to those obtained on healthy, non-contaminated dentin. Furthermore, the application of 2% chlorhexidine helped preserve bond strength values. Conversely, the removal of excessive dentin (0.5 mm) is not recommended as it led to reduced bond strength.

Keywords: *Universal Adhesive, Dentin Adhesion, Microtensile Bond Strength, Chlorhexidine*.

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Introduction

In the field of restorative dentistry, dental amalgam has long been established as one of the most conventional and commonly employed materials for the restoration of decayed or damaged teeth (1, 2). However, growing concerns related to environmental pollution from mercury content as well as aesthetic limitations have driven the progressive shift toward newer restorative materials, particularly resin composites, which are increasingly being utilized as substitutes for amalgam (3-5). One of the principal challenges encountered when using these composite materials lies in the uncertainty surrounding their ability to achieve a reliable and durable bond to dentin surfaces that have been contaminated by prior amalgam restorations (6). Nonetheless, resin composites possess various advantages, including superior aesthetic qualities, favorable mechanical properties, and effective adhesion to the remaining tooth structure in posterior regions (7, 8).

Conversely, it is well-established that the long-term clinical success of resin composite restorations is essentially dependent upon the achievement of a reliable and durable adhesive bond to the underlying tooth structure (9). A growing body of scientific literature indicates that the advent of universal adhesive systems has considerably streamlined this process by introducing simpler application protocols while simultaneously delivering improved bonding performance across various clinical scenarios (10, 11). Utilizing 10-MDP (10-methacryloyloxydecyl dihydrogen phosphate) chemistry, these adhesives form durable bonds with a variety of substrates, including dentin (under differing moisture conditions), enamel, ceramics, and metallic surfaces (10, 11). Additional reported advantages of universal adhesives include single-step application, enhanced marginal integrity of restorations, and favorable compatibility with indirect restorative procedures (12).

One of the principal challenges encountered during the replacement of existing amalgam restorations is the progressive contamination of the underlying dentin by metallic corrosion products, including tin, zinc, and copper, which accumulate over the functional lifespan of the amalgam (13). A substantial body of evidence has demonstrated that these residual metallic particles can adversely compromise the bonding performance of subsequently applied adhesive systems (13-15). Specifically, Harnirattisai et al. reported a statistically significant reduction in microshear bond strength on discolored dentin found beneath amalgam restorations when compared to healthy, non-contaminated dentin. This reduction was observed with both etch-and-rinse and self-etch adhesive strategies (14).

However, the existing body of research regarding adhesion to amalgam-contaminated dentin has yielded contradictory results. For instance, the study conducted by Harnirattisai et al. documented a considerable reduction in bond strength ranging from 20% to 30% on contaminated dentin surfaces (14). In contrast, the investigation by Scholtanus reported no significant difference in bond strength values when comparing tin/zinc-contaminated dentin to healthy dentin, provided that multi-step etch-and-rinse adhesive systems were employed (13).

Despite the growing body of evidence regarding adhesive bonding to various substrates, the application of universal adhesives specifically to amalgam-contaminated dentin has received comparatively little attention in the literature. Recent findings have demonstrated that contamination with aluminum chloride significantly reduces the bonding efficacy of self-etch adhesive systems, whereas etch-and-rinse systems remain unaffected by such contamination (16). Furthermore, it has been shown that six-month contamination with amalgam leads to a reduction in bond strength, and that the removal of the contaminated dentin layer paradoxically exacerbates this detrimental effect rather than mitigating it (15). Pretreatment with chlorhexidine, while not resulting in a statistically significant difference compared to control groups, has nonetheless shown a consistent trend toward improved bonding performance on contaminated dentin surfaces (17). A substantial number of studies have demonstrated that dentin contamination with amalgam-derived materials may compromise the bond strength of subsequent restorative materials. In light of these findings, the utilization of universal adhesive systems in conjunction with chlorhexidine pretreatment has been proposed as a viable strategy to enhance adhesive bonding under contaminated conditions. Conversely, the removal of even a minor amount of dentin during the course of restorative procedures may inadvertently exert a negative influence on the resulting bond strength. Therefore, the primary objective of the present study was to investigate the shear bond strength of a universal adhesive applied to amalgam-contaminated dentin, along with a comprehensive assessment of the effects of partial dentin removal and chlorhexidine pretreatment on bonding performance.

Methods

This in vitro study was conducted following formal approval by the Ethics Committee of Guilan University of Medical Sciences (ethics code: IR.GUMS.REC.1402.182). The required sample size was determined based on the results of a preliminary pilot study, utilizing PASS statistical software (version 11; NCSS LLC, USA) for power analysis. A total of 32 intact, non-carious human premolar teeth were employed in this study. These teeth had been extracted for orthodontic or periodontal indications and were obtained from dental clinics.

The experimental design consisted of eight distinct groups, with four teeth assigned to each group. From each tooth, two separate bonded specimens were prepared, yielding a total of eight specimens per group. Specimens were randomly allocated to the groups. The groups were defined as follows: G1 and G2 (control groups): Healthy, non-contaminated dentin; adhesive applied in self-etch mode (G1) and etch-and-rinse mode (G2). G3 and G4: Amalgam-contaminated dentin; self-etch mode (G3) and etch-and-rinse mode (G4). G5 and G6: Amalgam-contaminated dentin followed by pretreatment with 2% chlorhexidine; self-etch mode (G5) and etch-and-rinse mode (G6). G7 and G8: Amalgam-contaminated dentin followed by removal of a 0.5 mm layer of superficial dentin; self-etch mode (G7) and etch-and-rinse mode (G8). Premolar teeth eligible for inclusion were those possessing intact crowns, free from dental caries, cracks, or fractures, with no evidence of previous restorations or any structural defects. Teeth exhibiting extensive carious lesions, visible cracks or craze lines, or prior restorative treatments were excluded from the study. The independent variable was the etching mode (self-etch versus etch-and-rinse), while the dependent variable was the shear bond strength (Table 1).

Table 1. The materials employed in this study and their respective chemical compositions

Material	Brand	Composition
Universal All-Bond	Bisco, Schaumburg, IL, USA	BisGMA, 2-Hydroxyethyl Methacrylate, 10-Methacryloyloxydecyl Dihydrogen Phosphate, Ethyl 4-dimethylaminobenzoate
Cavity Cleanser	Bisco, Itasca, IL, USA	2% chlorhexidine digluconate
Composite Resin Opalis, Shade A2	FGM dental products, Joinville, Brazil	Bis-GMA, Bis-EMA, UDMA, TEGDMA, barium-aluminum, silanized silicate and nanoparticles of silicon dioxide, camphor quinone, accelerators, stabilizers and pigments

Specimen Preparation: The collected premolar teeth underwent a disinfection protocol by immersion in a 0.025% thymol solution maintained at a temperature of 4°C for a maximum duration of 14 days. Following disinfection, the teeth were embedded and fixed in self-curing acrylic resin (Acropars, Marlic Co., Iran) to facilitate handling during subsequent procedures. In order to expose the mid-coronal dentin surface, the occlusal enamel was carefully removed using a rotary instrument under continuous water cooling to prevent overheating and potential damage to the underlying dentin. Complete removal of the enamel layer was confirmed by examination under a stereomicroscope at 50× magnification (Motic B1, China). For the experimental groups G3 through G8, standardized cylindrical cavities measuring 1.5 mm in both depth and diameter were prepared on the mesial and distal aspects of each tooth using a carbide bur (No. 008/835, TeesKavan, Iran). These cavities were designed to provide adequate mechanical retention for the subsequent amalgam restoration. To facilitate the stabilization and condensation of amalgam, matrix bands with a height of 1.5 mm were placed circumferentially around the occlusal surface of each tooth. Amalgam was then condensed into the prepared cavities as well as over the occlusal surface to achieve a uniform height of 1.5 mm (Figure 1). After allowing the amalgam to set for 60 minutes, the specimens were stored in distilled water at 25°C for a period of 24 hours prior to further processing.

Figure 1. Amalgam placed on the occlusal surface and in the mesial and distal cavities

Specimens were subjected to thermocycling for 10,000 cycles between 5°C and 55°C with a dwell time of 30 seconds (Delta DBBP-2 device, South Korea). Following thermocycling, amalgam restorations were mechanically removed using a non-sharp instrument.

Following amalgam removal, a universal adhesive system (All-Bond Universal®, BISCO, USA) was applied to all specimens strictly in accordance with the manufacturer's recommendations. For the etch-and-rinse protocol, the dentin surface was first etched with 37% phosphoric acid for 15 seconds, followed by thorough rinsing with water and gentle drying using a sterile cotton pellet. For the self-etch protocol, the universal adhesive was applied to the dentin surface with an active rubbing motion for 20 seconds, after which a gentle stream of compressed air was directed onto the surface to facilitate solvent evaporation. In the chlorhexidine pretreatment groups (G5 and G6), a 2% chlorhexidine solution was applied to the contaminated dentin surface for 60 seconds prior to the adhesive application; the surface was then dried with a gentle air stream.

To standardize the bonding area, Tygon® tubes measuring 1 mm in internal diameter and 2 mm in height (Small Parts Inc., USA) were carefully positioned on the buccal and lingual one-third regions of each tooth. These tubes were then bonded to the underlying dentin surface according to the specific adhesive protocol assigned to each experimental group. A resin composite material was subsequently placed incrementally into the tubes, and each incremental layer was light-cured for 20 seconds at an irradiance of 1200 mW/cm² using an LED curing light (Bluedent LED, Bulgaria) (Figure 2).

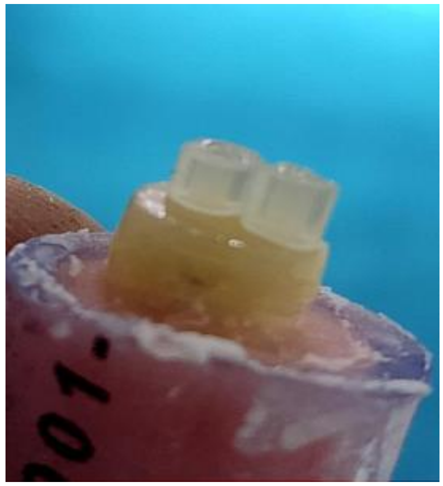

Figure 2. Composite resin cylinders constructed by means of Tygon® tubes (measuring 1 mm in internal diameter and 2 mm in height) positioned on the buccal and lingual tooth surfaces

To ensure consistent and standardized light exposure across all specimens, the output intensity of the LED light-curing device was calibrated immediately prior to each curing cycle using a calibrated LED radiometer (Woodpecker Medical, China). Following the completion of the curing procedure, the Tygon® tubes were gently removed from each specimen to avoid any mechanical disruption of the bonded interface. The specimens were then immersed in distilled water maintained at a temperature of 25°C for a period of 24 hours to permit post-curing relaxation. After this storage interval, the specimens were subjected to a second thermocycling regimen consisting of 10,000 cycles, employing identical parameters to those used in the initial thermocycling protocol (5-55°C, 30-second dwell time).

Microshear Bond Strength Testing Protocol: The microshear bond strength (μSBS) of the composite-dentin interface was assessed using a universal testing machine (STM-20, Santam, Iran) equipped with a load cell of appropriate capacity. The test was conducted at a crosshead speed of 0.5 mm/min using the wire-loop method, which involves a thin wire loop positioned around the base of the composite cylinder to apply a uniform shearing force (Figure 3). Bond strength results (calculated by dividing the maximum force at failure by the cross-sectional area of the specimen) were expressed in megapascals (MPa).

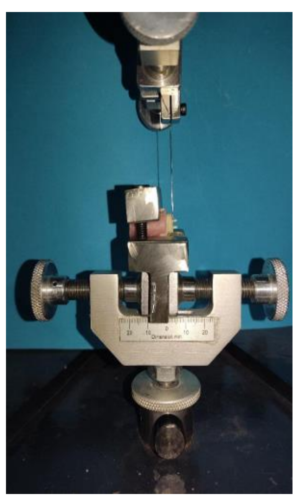

Figure 3. Microshear bond strength testing via the wire-loop technique

Failure Analysis: Failure patterns were examined using a stereomicroscope (2.5× magnification, Motic DM-143-FBGG, China) and classified into three categories based on the location of failure: adhesive failure (when more than 80% of the fractured surface occurred within the adhesive zone or at the adhesive-composite interface), cohesive failure (when more than 80% of the fractured surface occurred within the composite resin), and mixed failure (when both adhesive and cohesive failures were observed) (18).

Statistical analysis was performed using SPSS software version 28 (IBM Corporation, USA). Two-way analysis of variance (Two-way ANOVA) was used to evaluate the interaction effects of factors on bond strength, and pairwise comparisons were conducted using Tukey's post hoc test. The correlation between bond strength and failure pattern type was assessed using Spearman's correlation coefficient, and the distribution of failure patterns between etching methods was analyzed using the Mann-Whitney U test. A p-value ≤ 0.05 was considered statistically significant.

Results

Two-way analysis of variance revealed a statistically significant difference between the groups. According to the results, the control groups (G1 and G2) exhibited the highest mean bond strength values (17.80±3.78 MPa and 18.88±3.44 MPa, respectively). In contrast, the dentin removal groups (G7 and G8) showed the lowest values (12.89±3.45 MPa and 13.84±4.35 MPa, respectively) (p=0.048) (Table 2).

Subsequent pairwise comparisons using Tukey's post hoc test revealed specific significant differences. G1 demonstrated significantly higher bond strength than G7 (mean difference of approximately 4.91 MPa, p=0.001). Furthermore, G2 exhibited significantly higher bond strength compared to both G4 (contaminated dentin without pretreatment, etch-and-rinse) with a mean difference of 4.26 MPa (p=0.04) and G8 with a mean difference of 5.04 MPa (p=0.01). All other pairwise comparisons failed to reach statistical significance. Notably, the chlorhexidine-pretreated groups (G5 and G6) showed intermediate bond strength values that were not statistically distinguishable from either the control groups or the other amalgam-contaminated groups, suggesting a potential protective but not fully restorative effect.

Table 2. Bond strength of universal adhesive in each group and comparison with the control group

Group	Min-Max	Mean±SD	Comparison with control (mean difference)
Self-Etch			
G1 (Control)	12.21-22.84	17.80±3.78	-
G3 (Without preparation)	8.48-20.30	14.05±4.46	3.75
G5 (Chlorhexidine)	8.44-20.57	15.50±4.42	2.30
G7 (Dentin removal)	8.60-16.59	12.89±3.45	4.91
Etch-and-Rinse			
G2 (Control)	14.02-22.91	18.88±3.44	-
G4 (Without preparation)	9.27-21.17	14.62±3.84	4.26
G6 (Chlorhexidine)	9.27-22.91	16.05±4.36	2.83
G8 (Dentin removal)	9.12-20.63	13.84±4.35	5.04

Spearman's correlation analysis revealed no statistically significant correlation between bond strength values and failure types (adhesive failure: p=0.425, rs=0.330; mixed failure: p=0.425, rs=0.330). The Mann-Whitney U test similarly demonstrated no statistically significant difference in the distribution of failure patterns between the self-etch and etch-and-rinse etching methods (Table 3). Adhesive failures were the most prevalent, accounting for 67.2% of all specimens, suggesting that the adhesive-dentin interface represents the primary locus of bond failure (Figure 4).

Table 3. Relationship between etching methods and failure types

Method	Sample	Failure Type	p-value*
Self-Etch	23	Adhesive	0.317
Etch-and-Rinse	20	Adhesive	0.317
Self-Etch	9	Mixed	0.317
Etch-and-Rinse	12	Mixed	0.317

*Mann-Whitney U

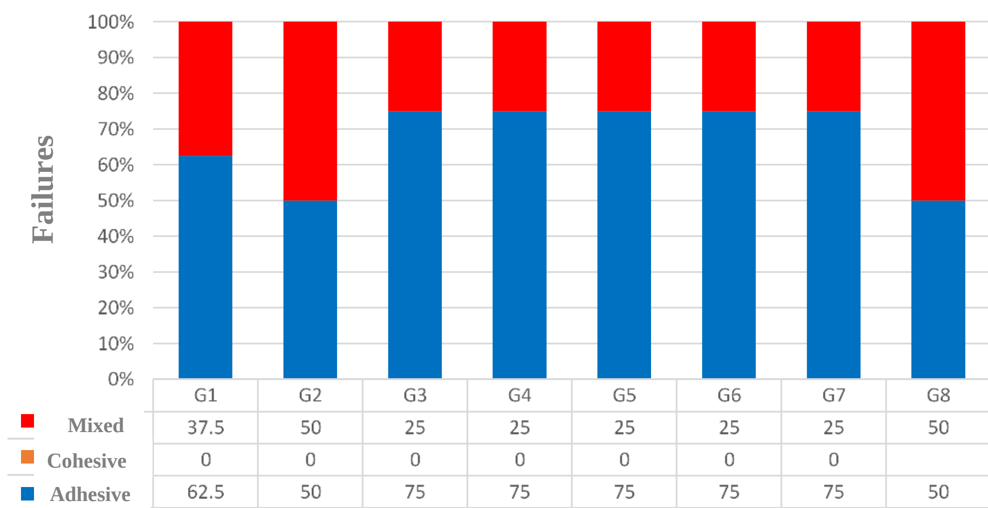

Figure 4. Types of failure in each group

Discussion

The principal finding of the present study was that the application of a universal adhesive in self-etch mode to amalgam-contaminated dentin yielded bond strength values comparable to those obtained on healthy dentin. Conversely, the etch-and-rinse protocol (performed without any adjunctive pretreatment) resulted in a statistically significant reduction in bond strength. This difference can be explained by the functional chemistry inherent to universal adhesive systems. The 10-MDP functional monomers contained within these adhesives possess the ability to chemically interact with metallic substrates, forming insoluble calcium-MDP salts while concurrently performing surface etching through their acidic functional groups (19-21). In contrast, etching with 37% phosphoric acid in the etch-and-rinse protocol removes calcium ions from the dentin surface, leaving behind a loosely arranged network of hydrated collagen fibers. The resulting lack of available calcium for the formation of stable, insoluble calcium-MDP salts compromises the long-term integrity of the adhesive interface, particularly following aging (22). Consequently, the self-etch application mode appears to be the preferred strategy when utilizing universal adhesives on amalgam-contaminated dentin.

A substantial and statistically significant reduction in bond strength was observed when the etch-and-rinse protocol was applied to dentin surfaces that had been contaminated by prior amalgam restorations. This finding is in close agreement with the results reported by Harnirattisai et al. (14), as well as by Alshehri et al. (17), both of whom documented diminished bonding performance on amalgam-contaminated dentin regardless of the specific adhesive technique utilized. Furthermore, Ghavamnasiri et al. (15) similarly reported a significant decrease in bond strength values following a six-month period of artificial aging, reinforcing the notion that contamination negatively impacts long-term adhesive stability. The underlying mechanism for this compromised performance appears to be multifactorial. Primarily, corrosion products released from the amalgam-particularly zinc ions-are thought to interfere with the infiltration of resin monomers into the demineralized dentin matrix and also disrupt the subsequent polymerization process (15, 16). Furthermore, silver sulfide has been implicated in binding to collagen fibrils, thereby contributing to the long-term instability of the hybrid layer (19).

Chlorhexidine pretreatment applied in the etch-and-rinse groups successfully restored bond strength values to levels comparable to those observed in the control group, thereby demonstrating its clinical utility when used alongside universal adhesive systems. Similarly, in the self-etch groups, chlorhexidine application preserved bond strength at levels equivalent to those achieved on healthy dentin. These findings are in agreement with the observations of Alshehri et al. (17), who reported the effectiveness of self-etch mode, and with the study by Rayar et al. (23), which documented improved bonding following chlorhexidine pretreatment. Chlorhexidine functions as a matrix metalloproteinase (MMP) inhibitor; by inhibiting enzymatic degradation and reducing nanoleakage, it contributes to the preservation of hybrid layer integrity over time (24, 25).

The removal of a 0.5 mm layer of superficial dentin resulted in a statistically significant reduction in bond strength, irrespective of whether the etch-and-rinse or self-etch adhesive protocol was employed. These findings are in accordance with previous reports by Alshehri et al. (17), Ghavamnasiri et al. (15), and Almozher et al. (26). This observed decrease in bond strength can be attributed to several alterations in the dentin substrate following superficial dentin removal, including increased dentinal tubule density, elevated water content, a reduction in the amount of intertubular dentin, and the persistent presence of residual amalgam corrosion products within the remaining dentin (27, 28). Consequently, the routine removal of dentin during the replacement of amalgam restorations is not recommended.

Adhesive failures were predominant across all experimental groups, accounting for 67.2% of all observed failure patterns. Notably, no cohesive failures within the composite resin were identified in any specimen. This failure pattern distribution is consistent with the findings reported by Luque-Martinez et al. (29), Muñoz et al. (30), Morsy et al. (31), and Daneshkazemi et al. (32), all of whom similarly identified the adhesive-dentin interface as the principal locus of bond failure. In contrast, Firat et al. (33) reported different findings, describing a higher prevalence of cohesive and mixed failure patterns. This discrepancy is likely attributable to methodological differences between studies, particularly variations in loading methods or specimen dimensions.

Statistical analysis revealed no significant correlation between failure type and bond strength values. Furthermore, no significant association was detected between the etching protocol employed (self-etch versus etch-and-rinse) and the distribution of failure patterns. While Morsy et al. (31) and Sabatini (34) have reported an association between mixed failure patterns and higher bond strength values, the findings of the present study are consistent with those of Daneshkazemi et al. (32), demonstrating that universal adhesive systems maintain a consistent failure pattern irrespective of the application technique utilized (35).

The present study is subject to several limitations. First, the in vitro nature of the investigation meant that the laboratory conditions did not completely simulate the artificial aging process or the complex intraoral environment, particularly with respect to the absence of artificial saliva. Second, inherent limitations associated with the microshear bond strength (μSBS) testing methodology should be acknowledged. These include the necessity of performing adhesive bonding outside the Tygon tube, which may introduce variability in the results, as well as the potential for pre-test stress to be applied during removal of the tubes prior to the definitive bond strength test. Finally, to further validate the findings of this study, long-term clinical studies and investigations incorporating other universal adhesive systems are strongly recommended.

The findings of this investigation indicate that the application of a universal adhesive system in self-etch mode to amalgam-contaminated dentin produces shear bond strength values comparable to those obtained on healthy dentin. Chlorhexidine pretreatment effectively preserves bond strength, whereas the removal of 0.5 mm of dentin leads to a significant reduction in bond strength.

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