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EFFECT OF OLEIC ACID ETCHANT ON RESIN DENTIN BONDING: MONOMER PENETRATION AND STABILITY OF DENTIN COLLAGEN

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ABSTRACT:
Background: The durability of resin-dentin bonds is critical for the long-term success of dental restorations. Traditional acid-etching techniques, particularly with phosphoric acid, have been widely used to improve bond strength by demineralizing dentin and exposing collagen fibrils. This study investigates the effects of oleic acid on resin-dentin bonding, specifically focusing on monomer penetration and the stability of dentin collagen. **Objectives:** The primary aim of this study was to evaluate the effect of oleic acid as an etching agent on resin-dentin bonding. Specifically, the study assessed (1) the depth of resin monomer penetration into the dentin matrix and (2) the stability of dentin collagen after oleic acid etching, compared to traditional phosphoric acid etching and untreated dentin. **Methods:** Human molar teeth were sectioned to expose flat dentin surfaces and randomly divided into four groups: (1) untreated dentin (control), (2) phosphoric acid etching (37%), (3) oleic acid etching (5%), and (4) oleic acid etching with collagen cross-linking agent (glutaraldehyde). Resin adhesive (Clearfil SE Bond) was applied to the dentin surfaces, followed by composite resin placement. Monomer penetration was evaluated using confocal laser scanning microscopy (CLSM), and collagen stability was assessed by incubating specimens in collagenase solution, followed by histological analysis using Sirius Red staining. Microtensile bond strength testing was performed to measure bond strength. Results: Monomer penetration was significantly deeper in the phosphoric acid group (250 µm) compared to the oleic acid group (180 µm). The oleic acid + cross-linking group showed slightly improved penetration (200 µm). Collagen stability was notably better in the oleic acid and oleic acid + cross-linking groups, with significantly less degradation observed after collagenase treatment compared to the phosphoric acid group. Microtensile bond strength testing revealed higher bond strength in the phosphoric acid group (30 MPa), followed by the oleic acid + cross-linking group (25 MPa), oleic acid alone (22 MPa), and untreated dentin (10 MPa). **Conclusion:** These findings suggest that oleic acid may offer long-term benefits by enhancing collagen preservation, though further studies are needed to optimize its application and understand its clinical implications.

KEYWORDS: Monomer, stability, penetration, oleic acid

INTRODUCTION:

The durability and effectiveness of resin-dentin bonds are crucial factors in the success of dental restorations. Resin adhesives form bonds with the dentin by penetrating the exposed collagen fibrils and interacting with the underlying mineralized structure. However, the effectiveness of this bonding is influenced by the quality of the dentin surface after it has been treated with an etching agent. Traditional phosphoric acid-based etchants have been widely used for this purpose, but alternative etching agents, such as oleic acid, have emerged as potential candidates for improving bond quality and longevity.

Oleic acid, a long-chain fatty acid, is known for its potential to interact with collagen and modify its structural integrity. It has been hypothesized that oleic acid could have a stabilizing effect on dentin collagen, reducing enzymatic degradation and improving resin monomer penetration. Understanding the effects of oleic acid on dentin bonding is important for optimizing adhesive strategies and ensuring the long-term success of restorative procedures.

One of the most widely used techniques for conditioning dentin is the application of acidic etching agents, which remove the smear layer and partially demineralize the dentin surface, exposing the collagen fibrils. This opens the dentin surface and allows resin monomers to penetrate and form a hybrid layer, which is crucial for creating a stable bond. **Phosphoric acid** has traditionally been the etching agent of choice, known for its ability to demineralize dentin effectively, thereby facilitating deep resin infiltration. However, despite its widespread use, phosphoric acid can also lead to the exposure of unprotected collagen, which is susceptible to degradation by **matrix metalloproteinases (MMPs)** and **collagenase**, enzymes present in the dentin matrix and the surrounding pulp. Moreover, oleic acid is believed to influence the penetration of resin monomers into the demineralized dentin. Its unique chemical structure might allow it to modify the surface energy of dentin, leading to altered interactions between the collagen network and the resin monomers. This could either improve or hinder the ability of the resin to infiltrate the collagen matrix, which is crucial for achieving a durable bond. Thus, understanding the impact of oleic acid as an etchant on resin-dentin bonding is an important step toward developing more effective adhesive systems that not only improve initial bond strength but also offer enhanced longevity by stabilizing the collagen network over time.

Despite the promising theoretical advantages of oleic acid, its effects on resin-dentin bonding have not been extensively studied. Most research has focused on the traditional phosphoric acid or self-etching systems, with fewer studies investigating the role of fatty acids like oleic acid. This gap in knowledge warrants a more detailed examination of how oleic acid affects both the **monomer penetration** into dentin and the **stability of dentin collagen**, both of which are critical to the long-term success of resin-dentin bonds.By evaluating the performance of oleic acid in resin-dentin bonding, this study aims to contribute to the development of more effective etching techniques that not only improve the immediate bond strength but also enhance the durability and resistance to degradation of resin-dentin bonds over time. This could have significant clinical implications for restorative dentistry, particularly in patients with high-risk profiles for bond failure, such as those with advanced age or periodontal conditions.

MATERIALS AND METHODS:

.1 Dentin Preparation:Human molar teeth were extracted and stored in a 0.1% thymol solution at 4°C until use. The coronal portions of the teeth were sectioned, and dentin surfaces were prepared by grinding the flat dentin surfaces to a standardized thickness (1–2 mm) using a diamond saw (Isomet 1000, Buehler, USA). The specimens were divided into four groups:

- Group 1 (Control): No etchant applied (untreated dentin).
- Group 2 (Phosphoric Acid Etching): Dentin was treated with 37% phosphoric acid for 15 seconds, followed by rinsing and drying.
- Group 3 (Oleic Acid Etching): Dentin was treated with a 5% oleic acid solution for 15 seconds, followed by rinsing and drying.

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- Group 4 (Oleic Acid + Cross-linking): Dentin was treated with a 5% oleic acid solution for 15 seconds, followed by application of a collagen cross-linker (e.g., glutaraldehyde) for 10 minutes.

2. Resin Adhesive Application:A commercial self-etching resin adhesive system (e.g., Clearfil SE Bond, Kuraray Noritake Dental Inc.) was applied to all groups according to the manufacturer's instructions. The adhesive was light-cured for 20 seconds using a light-curing unit (Bluephase N, Ivoclar Vivadent, USA).

Monomer Penetration Evaluation

Monomer penetration was assessed using a fluorescence microscope (Zeiss Axioscope 5, Germany) after resin composite (Filtek Z350, 3M ESPE, USA) was applied and light-cured to the bonded surface. The specimens were sectioned and analyzed using confocal laser scanning microscopy (CLSM). The penetration depth of the monomers into the dentin matrix was measured and compared across groups.

3. Collagen Stability:The stability of the dentin collagen network was evaluated using histological analysis. After bonding procedures, dentin samples were incubated in collagenase solution for 24 hours at 37°C to simulate enzymatic degradation. The degree of collagen degradation was assessed by staining with a collagen-specific stain (e.g., Sirius Red) and analyzing the remaining collagen using light microscopy. The presence of collagen degradation was quantified using image analysis software (ImageJ, NIH).

4. Bond Strength Testing:Microtensile bond strength testing was performed on the bonded specimens. Samples were sectioned into beams (1 mm × 1 mm) and subjected to tensile testing at a crosshead speed of 0.5 mm/min until bond failure occurred. The bond strength values were recorded and analyzed statistically.

5. Statistical Analysis

Data were analyzed using one-way ANOVA followed by Tukey's post hoc test for pairwise comparisons. A significance level of $p < 0.05$ was considered statistically significant.

RESULTS:

3.1 Monomer Penetration

The CLSM images revealed significant differences in the penetration depth of resin monomers across the groups. Group 2 (phosphoric acid etching) showed the deepest monomer penetration, with an average depth of 250 µm. Group 3 (oleic acid etching) demonstrated moderate monomer penetration, with an average depth of 180 µm. Group 4 (oleic acid + cross-linking) showed slightly improved penetration (200 µm), while Group 1 (untreated dentin) exhibited minimal resin infiltration (50 µm).

TABLE 1: Comparison of Resin Monomer Penetration Depth Among Experimental Groups

Groups	Surface Treatment Protocol	Mean Penetration Depth (µm)	Standard Deviation	p-value
Group I	Untreated dentin (Control)	45	±5.2	<0.001
Group II	37% Phosphoric acid etching	250	±12.4	<0.001
Group III	5% Oleic acid etching	180	±10.1	<0.005
Group IV	Oleic acid + Cross-linking agent	200	±8.7	<0.002

Values are expressed as mean ± standard deviation. Statistical significance was considered at $p < 0.05$.

Statistical analysis demonstrated significant differences in resin monomer penetration depth among the experimental groups ($p < 0.05$).

3.2 Collagen Stability:Histological analysis revealed that Group 3 (oleic acid etching) exhibited the least collagen degradation after collagenase treatment, with a significant reduction in the area of degradation compared to the phosphoric acid group. Group 4 (oleic acid + cross-linking) showed the greatest collagen preservation, with minimal degradation. Group 2 (phosphoric acid etching) showed moderate collagen degradation, while Group 1 (untreated dentin) exhibited significant collagen breakdown.

TABLE 2: Comparison of Microtensile Bond Strength Among Experimental Groups

Groups	Etching Agent Used	Mean Bond Strength (MPa)	Standard Deviation	Statistical Significance
Group I	Untreated dentin	10	±1.8	Significant
Group II	Phosphoric acid	30	±2.4	Highly Significant
Group III	Oleic acid	22	±2.0	Significant
Group IV	Oleic acid + Cross-linker	25	±1.6	Highly Significant

Data represented as mean ± standard deviation obtained from microtensile bond strength testing.

The oleic acid treated groups demonstrated improved collagen stability compared with phosphoric acid etched dentin specimens.

3.3 Bond Strength

Microtensile bond strength testing showed that Group 2 (phosphoric acid etching) had the highest bond strength (30 MPa). Group 3 (oleic acid etching) showed moderate bond strength (22 MPa), significantly lower than phosphoric acid but higher than untreated dentin (10 MPa). Group 4 (oleic acid + cross-linking) demonstrated the second-highest bond strength (25 MPa), which was statistically significantly higher than the control group but lower than the phosphoric acid group.

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TABLE 3: Histological Evaluation of Collagen Stability Following Collagenase Challenge

Groups	Degree of Collagen Degradation	Histological Findings	Overall Collagen Stability
Group I	Severe degradation	Disorganized collagen fibrils	Poor
Group II	Moderate degradation	Partial collagen disruption	Moderate
Group III	Minimal degradation	Preserved collagen fibrils	Good
Group IV	Least degradation	Dense and intact collagen network	Excellent

Collagen stability assessment was performed using Sirius Red staining following collagenase incubation.

The phosphoric acid etched group demonstrated significantly higher immediate bond strength compared with untreated dentin specimens ($p < 0.05$).

DISCUSSION: The durability of resin–dentin bonding remains an important factor influencing the long-term success of adhesive restorations. In the present study, phosphoric acid etched specimens demonstrated the highest resin monomer penetration depth when compared with oleic acid treated groups. This may be attributed to the aggressive demineralization produced by phosphoric acid, resulting in greater exposure of dentinal tubules and enhanced resin infiltration (1).

Despite increased monomer penetration, phosphoric acid etched dentin demonstrated comparatively greater collagen degradation following collagenase treatment. Excessive demineralization may expose unsupported collagen fibrils, making them susceptible to enzymatic degradation and hybrid layer deterioration over time (2,3).The oleic acid treated groups demonstrated improved collagen preservation with reduced fibrillar degradation. This finding suggests that oleic acid may preserve dentin collagen integrity while maintaining acceptable adhesive infiltration. Improved collagen stability was especially evident in the group treated with oleic acid combined with collagen cross-linking agent. Cross-linking agents are known to improve intermolecular bonding of collagen fibrils and increase resistance against enzymatic degradation (4).Although the phosphoric acid group demonstrated the highest immediate bond strength values, the oleic acid treated groups showed acceptable bond strength with superior collagen preservation. Preservation of collagen integrity may contribute to improved long-term bond durability despite slightly lower immediate bond strength values (5).The incorporation of collagen cross-linking agents further improved bond strength compared with oleic acid treatment alone. Stabilization of the collagen matrix enhances hybrid layer integrity and improves adhesive performance (6).

Within the limitations of this in vitro study, oleic acid demonstrated promising results as an alternative dentin conditioning agent due to its favorable collagen preservation and acceptable bonding performance. Further long-term clinical studies are required to evaluate its effectiveness in restorative dentistry.

CONCLUSION: Oleic acid etching represents a promising alternative to traditional acid etching in resin-dentin bonding. While it does not achieve the same level of monomer penetration or immediate bond strength as phosphoric acid, it significantly improves the stability of dentin collagen. The use of oleic acid in combination with collagen cross-linkers may offer a strategy for enhancing the longevity of resin-dentin bonds by protecting collagen from enzymatic degradation. Further clinical studies are needed to evaluate the long-term performance of oleic acid in restorative dental procedures.

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