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Restoring the bond strength of composite resin to bleached enamel with Epigallocatechin-3-gallate, Hypericin, and Resveratrol

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Abstract

Objective: This study investigated the effects of three herbal antioxidants on restoring the compromised bond strength of composite resin to bleached enamel.

Methods: In this in vitro study, 50 extracted human central incisors were divided into five groups. Group A (negative control) received no bleaching or antioxidant treatment. The teeth in groups B to E were bleached and subjected to the following surface treatment: Group B (positive control): no surface treatment; Group C: application of 5% Epigallocatechin-3-gallate (EGCG); Group D: application of 10% Hypericin; and Group E: application of 10% Resveratrol. The antioxidant solutions were applied for 10 minutes. All specimens were restored with a nanohybrid composite resin, and shear bond strength (SBS) was measured 24 hours after restoration using a universal testing machine. Data were analyzed using one-way ANOVA and Tukey's post hoc test with a significance set at $P<0.05$.

Results: ANOVA revealed a significant difference in SBS among the groups ($P=0.011$). The bleaching alone (Group B) significantly reduced SBS compared to other groups ($P< 0.05$). All three antioxidants (5% EGCG, 10% Hypericin, 10% Resveratrol) fully restored SBS to the level of the non-bleached control (Group A), with no significant differences between them ($P>0.05$). Among antioxidants, 5% EGCG showed the highest mean SBS (5.06 ± 1.57 MPa), followed closely by 10% Hypericin (4.92 ± 1.05 MPa) and 10% Resveratrol (4.79 ± 1.16 MPa).

Conclusions: All herbal antioxidants, including 5% EGCG, 10% Hypericin, and 10% Resveratrol, effectively improved the bond strength of composite resin to recently bleached enamel.

Keywords: Antioxidants, Bleaching agents, Dental bonding, Dental enamel, Polyphenols, Resveratrol

Introduction

Tooth bleaching has become a popular, non-invasive way to achieve a brighter, more attractive smile. After bleaching, additional restorative treatments such as composite resin bonding may be required to enhance the shape, color, or overall harmony of the teeth. A significant challenge in these procedures is the reduced bond strength between the composite resin and bleached enamel.

The reduced bond strength to bleached enamel is mainly attributed to the release of free radicals and anions during the bleaching process (1), which interfere with resin polymerization, leading to reduced bond strength between composite resin and enamel (2). Therefore, tooth restoration could not be performed immediately after bleaching. It is recommended that restorative treatments be delayed for at least two weeks; otherwise, an appropriate surface treatment should be applied to restore bond strength.

Several methods have been proposed to overcome the reduced bond strength to recently bleached enamel, including treating the enamel with ethanol or acetone-based solutions, removing the outermost enamel layer, applying antioxidants such as sodium ascorbate, and using bonding agents with organic solvents (3-5).

The use of antioxidants is a common method for restoring adhesion to recently bleached enamel. Antioxidants neutralize free radicals on the tooth surface, reducing oxidative damage caused by bleaching treatments. Consequently, the application of antioxidants enables immediate restoration after bleaching, eliminating the usual waiting period before bonding (6).

Both natural and synthetic antioxidants have been shown to effectively restore the compromised adhesion between composite resin and bleached enamel. Natural

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antioxidants are preferred over synthetic alternatives due to their biocompatibility, safety, and lower risk of adverse reactions. They effectively neutralize free radicals without introducing harmful chemicals, making them ideal for use on tooth surfaces after bleaching.

Green tea extract, particularly Epigallocatechin gallate (EGCG), is a natural antioxidant suitable for this purpose due to its minimal side effects, long-lasting effects, and relatively low cost. Al-Habsyi et al. (7) demonstrated that the antioxidant capacity of EGCG is 100 times greater than that of vitamin C and 25 times more potent than vitamin E.

Hypericin is a natural compound found in the flowering plant *Hypericum perforatum*, commonly known as St. John's Wort. It shows biological activity, antimicrobial, anti-inflammatory, and antioxidant effects. Its antioxidant properties help neutralize harmful free radicals in the body, enhancing oxidative stability in biological systems (8).

Resveratrol (3,5,4′-trihydroxystilbene) is a polyphenolic compound found in several plants, including peanuts, berries, and grapes. It is well-known for its potent antioxidant properties and is frequently studied for its potential health benefits, particularly its ability to reduce oxidative stress and inflammation. Recent studies suggest that Resveratrol may improve biocompatibility without causing a negative influence on µTBS of self-etch adhesives (9).

Although EGCG, Hypericin, and Resveratrol show antioxidant properties, there is limited comparative research on their effects on the shear bond strength (SBS) of composite resins to recently bleached enamel. The present study investigated the effect of EGCG, Hypericin, and Resveratrol on the SBS of a nanohybrid composite resin to bleached enamel.

Materials and methods

This in vitro study was approved by the ethics committee of CKS Teja Institute of Dental Sciences & Research, India (Approval No. CKS/ENDO/2025/01).

Sample size calculation

The sample size was calculated using G*Power software (version 3.1.9.2; Heinrich Heine University, Düsseldorf, Germany). Based on an effect size of $f = 0.55$ obtained from pilot data, with a significance level (α) of 0.05 and a statistical power ($1-\beta$) of 0.80, the minimum required sample size was determined to be 10 specimens per group. Considering five groups in this study, a total of 50 teeth were required.

Specimen preparation and grouping

Fifty non-carious human central incisors, extracted for periodontal reasons, were collected. Teeth were cleaned and scaled to remove calculus and debris, then stored in distilled water until use.

The specimens were embedded in acrylic resin blocks (15 × 15 mm), leaving the coronal portion above the cementoenamel junction exposed. The specimens were randomly assigned to five groups (n = 10). The teeth in Group A remained unbleached, whereas those in Groups B to E underwent in-office bleaching. Bleaching was performed with a 35% carbamide peroxide gel (Ultra White, Ammdent, Punjab, India) applied to the middle third of the labial enamel surface according to the manufacturer’s instructions. After bleaching, the specimens were thoroughly rinsed, gently wiped with a cotton ball, and dried.

The study groups were as follows:

Group A (Negative control): The teeth in this group received no bleaching or antioxidant treatment.

Group B (Positive control): The teeth were bleached, but no surface treatment was applied.

Group C: After bleaching, a freshly prepared 5% EGCG solution was applied to the surface with a microbrush and left for 10 minutes. The solution was made by dissolving EGCG powder (Vokin Biotech Pvt. Ltd., India) in absolute ethanol (Merck KGaA, Darmstadt, Germany). The specimens were then rinsed under running water and gently air-dried.

Group D: After bleaching, a 10% Hypericin solution was applied to the surface with a microbrush and left for 10 minutes. The solution was prepared by dissolving Hypericin powder (Sigma-Aldrich, MA, USA) in ethanol (Merck KGaA). The specimens were then rinsed under running water and gently air-dried.

Group E: After bleaching, a 10% Resveratrol solution was prepared by dissolving Resveratrol powder (DCM Technical Services, India) in ethyl alcohol (Merck KGaA). The solution was then applied to the surface with a microbrush and left for 10 minutes. The specimens were rinsed under running water and gently air-dried.

Tooth restoration

Specimens from all groups were etched with 37% phosphoric acid (Ivoclar Vivadent, Schaan, Liechtenstein) for 20 seconds, then rinsed and air-dried for 20 seconds. A layer of Tetric N-Bond Universal bonding agent (Ivoclar Vivadent) was applied to the etched surfaces and light-cured for 20 seconds, following the manufacturer's instructions. A stainless-steel mold measuring 5 mm × 4 mm was positioned on the labial enamel surface to define the bonding area.

Before composite resin placement, the mold was cleaned with 96% ethanol and dried. The inner surface of the stainless-steel mold was then coated with a polytetrafluoroethylene (PTFE)-based lubricant (PRESTO GmbH & Co. KG, Osnabrück, Germany) to prevent the composite resin from adhering to the mold. This preparation ensured smooth sample extraction without damage (10).

A nanohybrid composite resin (Tetric N-Ceram; Ivoclar Vivadent) was applied in 2 mm increments within the mold. Each increment was light-cured for 40 seconds using a high-intensity LED light-curing unit (≥1200 mW/cm^2; Bluephase N; Ivoclar Vivadent). The specimens were then stored in distilled water at room temperature for 24 hours before undergoing shear bond strength testing.

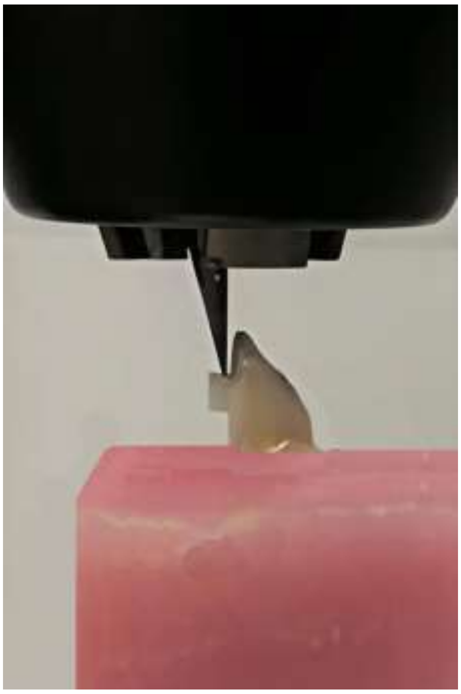

Figure 1. A specimen undergoing bond strength testing

Shear bond strength (SBS) measurement

Each mounted specimen was secured to a jig for bond strength testing (Figure 1). Shear bond strength was measured using a chisel-shaped crosshead in a universal testing machine (Instron 3369, Instron Corp., Norwood, MA, USA) at a crosshead speed of 1 mm/min. The failure force was divided by the cross-sectional area to calculate the bond strength in megapascals (MPa).

Statistical analysis

Data analysis was performed using IBM SPSS Statistics for Windows version 26.0 (IBM Corp., Armonk, NY, USA). One-way ANOVA was used for comparing bond strength among the study groups, followed by Tukey's post hoc test for pairwise comparisons. A significance level of $P<0.05$ was considered statistically significant.

Results

Table 1 presents the mean and standard deviation (SD) of SBS values in the study groups. One-way ANOVA indicated a significant difference in SBS among the groups ($P=0.011$; Table 1). Tukey's post hoc test showed that group B (positive control) had significantly lower bond strength compared to all other groups ($P<0.05$). No significant differences were found between the non-bleached control group (Group A) and any of the antioxidant-treated groups ($P>0.05$; Table 1).

Among the antioxidants, 5% EGCG (Group C: 5.06 ± 1.57 MPa) showed the highest mean bond strength, followed by 10% Hypericin (Group D: 4.92 ± 1.05 MPa) and 10% Resveratrol (Group E: 4.79 ± 1.16 MPa), with no statistically significant differences between them ($P>0.05$; Table 1). Figure 2 illustrates a comparison of SBS values across the study groups.

Discussion

This study investigated the effects of three herbal antioxidants, 5% Epigallocatechin-3-gallate (EGCG), 10% Hypericin, and 10% Resveratrol, on restoring the compromised shear bond strength of composite resin to bleached enamel. In this study, the lowest bond

Table 1. The mean and standard deviation (SD) of bond strength values (MPa) in the study groups

Group	Definition	Shear bond strength Mean ± SD*
A	Negative control	4.79 ± 1.46^a
B	Positive control	2.82 ± 1.23^b
C	5% EGCG	5.06 ± 1.57^a
D	10% Hypericin	4.92 ± 1.05^a
E	10% Resveratrol	4.79 ± 1.16^a
P-value*		0.011

*Different superscripted letters denote statistically significant differences between groups at P<0.05.

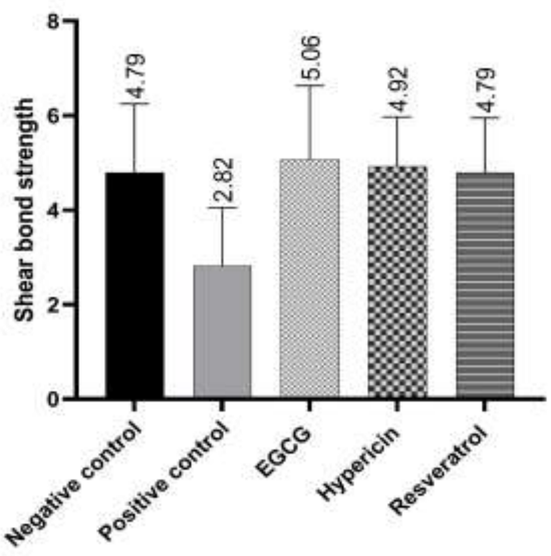

Figure 2. Comparison of shear bond strength values (MPa) in the study groups (Positive control: No bleaching or antioxidant treatment; Negative control: Bleaching without antioxidant application; EGCG: Epigallocatechin-3-gallate

strength (2.82 ± 1.23 MPa) was observed in bleached enamel without antioxidant treatment (Group B), implying the detrimental effects of bleaching on the adhesion of composite resins.

Several mechanisms have been proposed to explain the negative impact of bleaching on bond strength. The release of oxygen-free radicals (OFRs) and other reactive species, along with morphological and compositional changes in tooth structure, may contribute to decreased resin tag formation penetration as well as impaired polymerization of adhesive agents. These factors result in a reduction in bond strength between the composite resin and bleached enamel (11, 12). Typically, this reduction in bond strength persists for up to two weeks after bleaching, requiring a delay before restorative procedures can be conducted (13).

In the present study, the application of all herbal antioxidants, including 5% EGCG, 10% Hypericin, and 10% Resveratrol, significantly increased bond strength to bleached enamel, reaching values comparable to the mean value of the control group (4.79 ± 1.46). This finding is in agreement with several studies that demonstrated antioxidants can effectively neutralize free radicals, allowing for immediate restoration to bleached enamel (14-16).

Among the antioxidants tested, Group C (5% EGCG) showed the highest shear bond strength (5.06 ± 1.57 MPa). Epigallocatechin gallate (EGCG) is a primary polyphenol found in green tea, present in the highest concentrations compared to other polyphenols such as epigallocatechin (EGC), epicatechin gallate (ECG), and epicatechin (EC) (17-19). The antioxidant capacity of green tea's compounds is ranked as follows: EGCG > EGC > ECG > EG (17-19). The strong antioxidative properties of EGCG are attributed to its polyphenolic structure and chemical composition (13, 19, 20). The findings of the present study are consistent with Khamverdi et al. (13), who proposed that applying an EGCG solution, regardless of the concentration or application time, increased the bond strength of composite resin to bleached enamel. Other studies also demonstrated that a 10-minute application of green tea extract or 5% EGCG after bleaching improves the shear bond strength of composite resin restorations (7, 15).

In contrast to the findings of this study, Jain et al. (21) found that EGCG did not significantly improve shear bond strength after bleaching, possibly due to variations in study design, EGCG concentration, and application time. Ozakar-Ilday et al. (22) observed that sodium ascorbate, chitosan, catalase, and EGCG significantly enhanced bond strength after office bleaching but not following home bleaching. Office bleaching employs higher concentrations of bleaching agents over shorter periods, causing more immediate but often reversible enamel changes that antioxidants can quickly neutralize. In contrast, home bleaching involves lower concentrations of hydrogen peroxide over longer durations, leading to more profound and prolonged enamel alterations that delay bond strength recovery. This explains why antioxidants improve bond strength more effectively after office bleaching than home bleaching (23).

According to the findings of this study, the shear bond strength of specimens in Group D (10% Hypericin: 4.92 ± 1.05 MPa) was slightly lower than that in Group C (EGCG), although statistically comparable to other antioxidants and the unbleached control group. This finding implies that 10% Hypericin is effective in reversing the compromised bond strength to bleached enamel. Similarly, Yilmaz et al. (8) evaluated the effect of 10% Hypericum perforatum L. (HPL) extract on bond strength to bleached dentin and found that HPL was as effective as 10% sodium ascorbate in reversing the reduced dentin bond strength caused by bleaching.

The bond strength value of specimens in Group E (10% Resveratrol: 4.79 ± 1.16 MPa) was slightly lower than that in groups C (5% EGCG) and D (10% Hypericin). Resveratrol (3, 5, 4′-trans-trihydroxystilbene) is an active ingredient extracted from Polygonum cuspidatum, a plant belonging to the Polygonaceae family (24). It exhibits significant antioxidant properties due to the high concentration of hydroxyl groups in its phenolic structure (25). The present findings align with those of Cengiz-Yanardag and Karakaya (25), who showed that applying 1 micromolar Resveratrol for 10 minutes significantly improved the microshear bond strength (μSBS) of the adhesive to enamel bleached with 40% hydrogen peroxide. This improvement was attributed to Resveratrol's strong antioxidant properties, which effectively neutralize residual free radicals.

This study has some limitations. The study was conducted in vitro, and the results may not fully reflect clinical conditions, as factors such as saliva, masticatory forces, and temperature fluctuations can influence bond strength. Further in vivo research is needed to validate whether the present findings can be replicated in a real-world clinical setting.

Conclusions

Within the study's limitations, the lowest bond strength was observed in bleached enamel without antioxidant application. The use of all herbal antioxidants, including 5% EGCG, 10% Hypericin, and 10% Resveratrol, significantly restored the reduced bond strength to bleached enamel, bringing it to values comparable to unbleached enamel.

Conflict of interest

The authors declare that they have no conflict of interest.

Ethical considerations

This in vitro study was approved by the ethics committee of CKS Teja Institute of Dental Sciences & Research, India (Approval No. CKS/ENDO/2025/01).

Author Contributions

V.K.N., M.D.V., and C.S.K. developed the project, helped in data analysis, and edited the manuscript. S.S.K., K.S.C., and R.B. collected and analyzed the data and wrote the manuscript. All authors read and approved the final manuscript.

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