Effect of different antioxidants on reversing compromised resin bond strength after enamel bleaching: An in vitro study

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Abstract

Objective: The effect of 10% sodium ascorbate, 10% α-tocopherol, 10% grape seed extract, and 10% guava seed extract solutions on the shear bond strength (SBS) of composite resin to bleached enamel was evaluated.

Materials and Methods: Labial enamel surfaces of 72 extracted maxillary central incisors were bleached using 37.5% hydrogen peroxide. Sixty-four specimens were divided into four experimental groups (Group I–IV) comprising 16 specimens each and remaining eight specimens were placed in Group V. Groups I–IV were further divided into 2 subgroups: A (10 min) and B (120 min) comprising 8 specimens each. Eight unbleached specimens served as control (Group VI). Immediately following bleaching, Groups IA–IVA and IB–IVB specimens were treated with respective antioxidants for 10 min and 120 min. All specimens were then bonded with composite resin. Specimens were stored in distilled water for 24 h. SBS testing was done. Data were tabulated and subjected to statistical analysis using ANOVA and Tukey’s honest significant difference test.

Results: Group VI (unbleached) showed the highest SBS followed by Group IV (10% guava seed extract).

Conclusion: Use of antioxidants effectively reversed the compromised bond strength of bleached enamel. About 10% guava seed extract application after bleaching showed best bond strength.

Keywords: Alpha-tocopherol; antioxidant; bleaching; grape seed extract; guava seed extract; reduced bond strength; reversal; sodium ascorbate

INTRODUCTION

Discolored teeth frequently affect people of various ages and are multifactorial in nature. With the evolution in esthetic dentistry, in-office and at-home bleaching procedures have become popular.

However, bleaching agents have certain adverse effects such as hypersensitivity, gingival irritation, micromorphological defects due to demineralization, and reduced enamel microhardness. In addition, bleaching agents affect the bonding of enamel and dentin tissues when restorations are applied immediately postbleaching. This problem is more notable when the desired results after bleaching are not achieved, and the patient wishes for additional esthetic options such as direct and indirect veneers.

Various techniques have been proposed to resolve this clinical problem are as follows: conditioning the bleached enamel with alcohol before restoration, removing the surface layer of enamel, and employing adhesives containing organic solvents. Nonetheless, at present, the universal approach is to adjourn any bonding procedure for 4 days to 4 weeks.

To avoid this delay, application of various antioxidant agents such as 10% sodium ascorbate, Vitamin E, and proanthocyanidin postbleaching have been proposed.

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Although studies have revealed the effectiveness of sodium ascorbate\(^{36}\) and grape seed extract\(^{39}\) in reversal of decreased bond strength to bleached enamel, not many studies have been done on the use of Vitamin E (\(\alpha\)-tocopherol) and proanthocyanidins derived from other fruits like guava as viable alternatives to sodium ascorbate. Moreover, studies have reported incongruous findings with concerns to absolute reversal of bond strength to baseline levels after 10 min of application.\(^{10,11}\) Hence, the rationale of this in vitro study is to compare and evaluate the effects and efficacy of 10% sodium ascorbate solution, 10% \(\alpha\)-tocopherol solution, 10% grape seed extract solution, and 10% guava seed extract solution on the bond strength between composite resin and bleached enamel after 10 min and 120 min.

The null hypothesis tested was that there is no difference in reversal of reduced shear bond strength (SBS) after application of 10% sodium ascorbate solution, 10% \(\alpha\)-tocopherol solution, 10% grape seed extract solution, and 10% guava seed extract solution for 10 min and 120 min.

**MATERIALS AND METHODS**

Eighty human permanent maxillary central incisors extracted for periodontal reasons were collected and rinsed in 0.9% unbuffered saline. Labial surfaces of these teeth were flattened with 600 grit silicon carbide paper and roots were embedded in acrylic resin block so that only the coronal portion was exposed. Individual molds of tooth-colored self-cure resin were prepared over the specimens. Modeling wax was used as a separator, so as to block the undercuts and also to provide space for antioxidant solution on the labial surface of the specimens. A customized cylindrical silver mold of 3 mm diameter and 5 mm height was also fabricated.

For preparation of 10% antioxidant solutions, 10 gm sodium ascorbate powder (SD Fine Chem Limited, Mumbai, Maharashtra, India) and 10 gm grape seed extract powder (Biovea, USA) were dissolved in 100 ml of distilled water. For preparation of 10% alpha-tocopherol, 10 ml \(\alpha\)-tocopherol (SD Fine Chem Limited, Mumbai, Maharashtra, India) was dissolved in 100 ml of ethyl alcohol. For 10% guava seed extract, 40 g guava seeds were used and prepared with Soxhlet extraction method. Ten milliliters of this extract was then mixed in 100 ml of distilled water to make 10% guava seed extract solution.

Labial surfaces of 72 specimens were bleached with Pola office one patient kit (SDI, Victoria, Australia) containing 37.5% hydrogen peroxide with four applications of 8 min each. The gel was then rinsed off with water. These specimens were divided as follows:

- **Group I** \((n = 16)\) treated with 10% sodium ascorbate solution
- **Group II** \((n = 16)\) treated with 10% \(\alpha\)-tocopherol solution
- **Group III** \((n = 16)\) treated with 10% grape seed extract solution
- **Group IV** \((n = 16)\) treated with 10% guava seed extract solution
- **Group V** \((n = 8)\) treated with no antioxidant solution
- **Group VI** \((n = 8)\) control (unbleached specimens).

Based on the application period of antioxidants, Groups I–IV were further subdivided as:

- **Subgroup A** \((n = 8)\) Antioxidant treatment for 10 min immediately after bleaching
- **Subgroup B** \((n = 8)\) Antioxidant treatment for 120 min immediately after bleaching.

The antioxidant solutions were refreshed every 10 min in subgroup B. Specimens were then rinsed with water. Groups V and VI did not receive any antioxidant treatment.

Labial surfaces of all specimens of subgroup A and B (Group I–IV), Group V, and Group VI were bonded with composite resin. Customized cylindrical silver mold was placed on the labial surface of each specimen and composite resin (Filtek Z 350 XT) was then placed in three increments so as to have a final mass of 3 mm diameter and 5 mm height.

All specimens, after composite build up, were stored in distilled water for 24 h and SBS testing was done with Universal Testing Machine (Banbros Engineering Pvt Ltd.,) at across head speed of 1 mm/minute. Data were tabulated and subjected to statistical analysis.

**RESULTS**

On intergroup comparison of bond strength of subgroup A, except for difference between subgroup IA and IIA and between subgroup IVA and VI, all the other differences were statistically significant \((P < 0.001)\).

Table 1. The order of bond strengths observed was V<IA=IIA<IIIA<IVA<VI.

On intergroup comparison of bond strength of subgroup B, minimum difference was between subgroups IIIB and IV B and maximum was between subgroup IV and V. Group V had significantly lower mean value as compared to all the other groups/subgroups. None of the other between-group differences was significant statistically. Table 1 the order of the bond strength observed was V<IB=IIIB=IVB<VI. Intragroup comparison revealed significantly higher bond strength in subgroup B of all group except group IV [Graph 1].
DISCUSSION

All the four antioxidants (subgroup B) were capable of reversing the reduced bond strength following bleaching. However, the antioxidants in subgroup A were not able to do so. Hence, the null hypothesis was rejected.

Since the antioxidant preparations were in solution form, individual resin molds were prepared for adequate contact of antioxidants with the labial surface of the specimens. In clinical scenario, molds can be substituted with customized trays that can be worn by the patient for an appropriate time.

Sodium ascorbate and \(\alpha\)-tocopherol solutions were prepared from the powder supplied, but the guava seed extract was prepared from white guava seeds using Soxhlet extraction method. This extraction method is advantageous as it allows only one consignment of solvent to be recycled instead of several batches of warm solvent. Furthermore, the solvent is removed through a rotary evaporator, producing the extracted compound.

Several studies have reported that 10 min application of sodium ascorbate is effective in reversing the decreased bond strength.\[13\] Studies have also demonstrated that increasing the period of antioxidant application increased the bond strength of composite to enamel.\[8,14\] This occurred till 120 min.\[8\] To determine the completion time for reversing the reduced bond strength, 10 min and 120 min application period were chosen for the current study.

The most frequently ignored guideline in the test protocol is to follow the ISO/TS 11405 specification (2003), that is, “a limitation of the bonding area is important.”\[15\] Hence, to fulfill the criteria molds of silver with 3-mm internal diameter and 5-mm height were made.

In the present study, after application of respective antioxidants for 10 min, the mean SBS of Group VI (mean = 77.75 Mpa) was statistically significant when compared to all the other groups. These findings indicate better bonding if adhesive procedures are performed without bleaching. These findings corroborate with the results of previous studies.\[3,10,11\]

It was also observed that immediate bonding to bleached enamel resulted in statistically significant \((p < 0.05)\) decrease in the SBS (mean = 43.63 Mpa) when compared to control group (mean = 77.75 Mpa). These findings are in accordance with the results of Borges et al.\[16\] This reduction in bond strength on immediate composite placement postbleaching may be due to delayed release of oxygen,\[3\] which in turn could either interfere with resin infiltration into the etched enamel or hinder resin polymerization.\[17\]

After application of respective antioxidants for 10 min, the mean SBS of the Group IIIA was the highest among all the other antioxidant groups. High antioxidant activity of guava might be responsible for almost complete reversal of reduced bond strength.\[18\] Leong and Shui\[19\] investigated the antioxidant capacity of fruits in Singapore market and ranked guava and star fruit as high in their antioxidant contents. So far, no study has been done to compare guava seed extract with other antioxidants on the reversal of reduced bond strength of bleached enamel. Hence, the findings of the study could not be corroborated or contradicted.

Similar to study by Vidhya et al.,\[20\] an improved bond strength was observed on comparing Group IIIA with Groups IIA and V. This may be due to the presence of proanthocyanidin complexes in grape seeds which are responsible for its antioxidant activity.\[21\] Free-radical scavengers or antioxidants provide protection of cells against oxidative damage.\[22\]
When compared with the mean SBS of Group VI, Group IIIA showed incomplete reversal of reduced bond strength. This is in accordance with the study of Arumugam et al.\textsuperscript{[23]} However, Vidhya et al.\textsuperscript{[20]} reported complete reversal of reduced bond strength after 10 min application of 10% grape seed extract for 10 min.

However, significantly better results were observed in Group IIIA with that of Group IA. This is in accordance to the study conducted by Vidhya et al.\textsuperscript{[20]} Cvitko et al.\textsuperscript{[5]} stated that naturally occurring antioxidants such as grape seed extract contain oligomeric proanthocyanidin complexes that have free-radical scavenging ability showed 50 times more potent than sodium ascorbate. However, Arumugam et al.\textsuperscript{[23]} concluded that 10% sodium ascorbate showed significantly higher bond strength when compared to 6.5% proanthocyanidin and lycopene. This difference in the findings of their study and our study may be because in their study the concentration of grape seed extract was less than that of sodium ascorbate.

In the present study, it was also observed that Groups IA and IIA showed significantly higher mean SBS as compared to the Group V. This may be because sodium ascorbate and alpha-tocopherol allow free-radical polymerization of adhesive resin without premature termination by restoring the altered redox potential of the oxidized bonding substrate, thus improving the compromised bonding.\textsuperscript{[24]} Furthermore, the presence of alcohol in the composition of the 10% alpha-tocopherol solution formulated for this study may have contributed in improving the compromised bond strength of bleached enamel since 10% alpha-tocopherol was immiscible in water solution. Thus, the phenomenon observed may be system-specific, not only due to antioxidant agent but also due to the presence of alcohol.\textsuperscript{[10]} These findings are in accordance with studies of Lai et al.,\textsuperscript{[3]} Sasaki et al.,\textsuperscript{[10]} Thapa et al.,\textsuperscript{[11]} and Torres et al.\textsuperscript{[25]}

Findings of Sasaki et al.\textsuperscript{[10]} contradicts the effectiveness of 10% sodium ascorbate when applied for 10 min. Torres et al.\textsuperscript{[25]} evaluated the effects of six antioxidants, including sodium ascorbate, on the bond strength of enamel postbleaching. However, none of the antioxidant agents were capable of completely neutralizing the deleterious effects of bleaching on the bond strength. The same effect of sodium ascorbate was observed in the current study when applied for 10 min. However, on comparing the mean SBS of Group IIIA with that of control group, the former showed incomplete reversal in bond strength.

Furthermore, a lower mean SBS of Group IIA was observed when compared to that of control group. This indicates that application of 10% alpha-tocopherol for 10 min did not restore the reduced bond strength to baseline levels. This is in accordance to the findings of Thapa et al.\textsuperscript{[11]}

In our study, except for guava extract solution group, none of the antioxidants showed a complete reversal in bond strength when applied for 10 min. This corroborates with the findings of previous studies.\textsuperscript{[10,11,25]}

Furthermore, after the application of respective antioxidants for 120 min, complete reversal of reduced bond strength was achieved in all the experimental groups. This is in accordance with the previous studies\textsuperscript{[10,11,25]} which stated that the bond strength increases with the increase in antioxidant application time till 120 min.\textsuperscript{[10,11]} Thus, the null hypothesis that there is no effect of different antioxidants and different application time on reversal of reduced bond strength of bleached enamel was rejected.

**CONCLUSION**

- Only the application of 10% guava seed extract for 10 min reverses it to baseline levels
- All the antioxidants after 120 min of application completely restored the reduced bond strength
- Further studies need to be conducted using higher concentrations of antioxidants and reducing their time of application so as to achieve complete reversal of reduced bond strength after enamel bleaching.

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**Conflicts of interest**

There are no conflicts of interest.

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