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COMPARATIVE EVALUATION OF IMMEDIATE BOND STRENGTH TO BLEACHED ENAMEL FOLLOWING APPLICATION OF VARIOUS ANTIOXIDANT SOLUTIONS

Alisha Dhingra[1], Ashu K. Gupta[2], Anshu Minocha[3], Nayantara Sen[4]
[1]PG Student, Department of Conservative Dentistry & Endodontics, H.P. Govt. Dental College & Hospital, Himachal Pradesh, India
[2]Professor & Head, Department of Conservative Dentistry & Endodontics, H.P. Govt. Dental College & Hospital, Himachal Pradesh, India
[3]Assistant Professor, Department of Conservative Dentistry & Endodontics, H.P. Govt. Dental College & Hospital, Himachal Pradesh, India
[4]PG Student, Department of Conservative Dentistry & Endodontics, H.P. Govt. Dental College & Hospital, Himachal Pradesh, India

ABSTRACT

Aim: The aim of this study was to compare the reversal of shear bond strength of composite to bleached enamel immediately after bleaching followed by application of various antioxidant solutions. **Material and Methods:** Seventy central incisors were divided into seven groups. Groups I and II served as unbleached and bleached controls respectively. Groups III, IV, V, VI and VII served as the experimental groups and were subjected to 37.5% hydrogen peroxide bleaching followed by 10 min application of 10% sodium ascorbate, 25% alpha-tocopherol, 6.5% grape seed extract, 5% lycopene and 5% green tea extract respectively. Following composite bonding, shear bond strength was determined and the results were analyzed using ANOVA followed by Post Hoc Multiple Comparisons test. **Results:**The bond strength values for Group I (positive control) were maximum and significantly different than all the other groups except Grape seed extract group (Group V). When compared to Group II (bleached control), all the groups showed significantly higher bond strength. Significant difference in the bond strength values were seen between Group III (10% sodium ascorbate) and Group V. Also values for Group V were significantly different from Group VI (5% lycopene). All the other values showed insignificantly different results. **Conclusion:** All the antioxidant solutions improved the shear bond strength values after bleaching but only Grape seed extract application reversed the values to the non bleached levels. Lycopene was least effective. Other antioxidants showed comparable results.

Keywords : Bleaching, Antioxidant, Sodium ascorbate, alpha-Tocopherol, Grape seed extract, Lycopene, Green tea extract.

INTRODUCTION:

In a highly appearance-driven society, tooth whitening techniques continue to become ever more prolific and diverse.[1] Previous studies have shown that hydrogen peroxide and carbamide peroxide used as bleaching agents affect the bond strength of composites to acid etched enamel when bonding is performed immediately after the bleaching treatment. Some authors assert that the oxygen remains in the dental structure after bleaching and can interfere with the polymerization of adhesive monomers.[2] A delay in bonding of 1 to 3 weeks following the bleaching procedure is recommended for enamel to return to conditions that lead to normal bond strengths.[3,4] Nevertheless, it was observed that the use of anti-oxidant agents before the bonding process can reverse the compromised bonding to bleached enamel.[3-8] This is clinically relevant as in many cases patients want restorations as soon as the bleaching is done because of time constraints or high aesthetic demands.

Several studies have tested the efficacy of natural antioxidants like lycopene, alpha tocopherol, grape seed extract and green tea extract in comparison with sodium ascorbate but comparison amongst these antioxidants has not been performed. Therefore, in the

Corresponding Author:
Alisha Dhingra
E-mail:
dhingraalisha15@gmail.com
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current study we propose to evaluate and compare the shear bond strength to bleached enamel following the application of 10% sodium ascorbate, 25% alpha-tocopherol , 6.5% grape seed extract , 5% lycopene and 5% green tea extract as antioxidant solution immediately after bleaching for reversal of compromised bonding .

Sodium ascorbate has been traditionally used for reversing the compromised bond strength but a number of natural antioxidants are available today which can be used for this purpose.

Ascorbic acid and its sodium salts are potent antioxidants capable to donate two high-energy electrons to scavenge the free radicals.[7-8]

Alpha-tocopherol allows free-radical polymerization of the adhesive resin to proceed without premature termination by restoring the altered redox potential of the oxidized bonding substrate and improves resin bonding.[9-10] Another natural antioxidant, lycopene, a carotenoid found in tomato extract, is known to have free radical scavenging ability.[11]Proanthocyanidins are high molecular weight polymers that comprise the monomeric flavan 3-ol catechin and epicatechin.[12]Green tea catechins have been shown to possess potent antioxidant activity several times higher than vitamin C and vitamin E. The strong antioxidant properties of green tea have been attributed to catechins of epigallocatechin gallate (EGCG) and epigallocatechin (ECG).[13]

MATERIAL AND METHODS:

Seventy intact human maxillary central incisors extracted for periodontal reasons were used in the study. Exclusion criteria consisted of teeth with caries, cracks and non carious lesions. The roots were separated at the level of cemento-enamel junction using a water cooled diamond disc and were later sealed with light body elastomeric material to avoid penetration of the acrylic resin or bleaching solution into the pulp chamber. Specimen were then mounted in self cure resin with their buccal surfaces upward at the level of acrylic resin which were kept in distilled water until the resin was completely cured to avoid thermal effects generated by resin curing process. The buccal surfaces of teeth were flattened using 600 grit silicon carbide paper.

GROUPS (n=10)	STUDY PROCEDURE
Group I (Positive Control)	Positive Control i.e. composite bonding without 37.5% hydrogen peroxide bleaching.
Group II (Negative Control)	Negative Control i.e. composite bonding followed only by 37.5% hydrogen peroxide bleaching.
Group III (10% Sodium Ascorbate)	37.5% hydrogen peroxide bleaching+10% Sodium Ascorbate application+ composite bonding
Group IV (25% alpha-tocopherol)	37.5% hydrogen peroxide bleaching+25% alpha-tocopherol application+composite bonding
Group V (6.5% proanthocyanidin)	37.5% hydrogen peroxide bleaching+6.5% proanthocyanidin application+ composite bonding
Group VI (5% lycopene)	37.5% hydrogen peroxide bleaching+5% lycopene application+composite bonding)
Group VII (5% Green Tea Extract)	37.5% hydrogen peroxide bleaching+5% Green Tea Extract application+composite bonding

Table 1

Study Design - Refer to Table 1.

37.5% Hydrogen peroxide bleaching agent was applied according to the manufacturer's instructions for 24 minutes i.e. three applications of 8 minutes each. Ten grams of sodium ascorbate, 6.5 grams of grape seed extract, 5 grams of lycopene and 5 grams of green tea extract in powder form were dissolved in 100 ml distilled water to obtain 10%, 6.5%, 5% and 5% solution respectively. Twenty five grams of α-tocopherol powder was dissolved in 100ml of ethanol to obtain 25% of alpha-tocopherol solution. The solutions were applied using a calibrated syringe at the rate of 1ml/min for 10 minutes. The applied solutions were continuously agitated using a sterile brush and then air dried. The buccal surface of the samples was then etched using 37% phosphoric acid gel and bonding agent was applied and light cured as per manufacturer's instructions. A polypropylene casing of dimensions 3mm x 4mm was assembled and the resin composite was added in increments 1.5 - 2 mm thick and cured as per manufacturer's directions. Following complete curing polypropylene casing were cut. Bonded specimens were stored in deionized water for 24 h at 37°C before shear bond strength testing to artificially age the composite resins.[14] For measuring the shear bond strength, the samples were placed in a universal testing machine with a blade-shaped tip at the crosshead speed of 0.5 mm/min . The load at which failure occurred was recorded by software. The shear bond strength values of the samples were calculated and expressed in S.I. units Mega Pascal (MPa).

STATISTICALANALYSIS

Normality of quantitative data were checked by measures of Kolmogorov Smirnov tests of normality. As data for Shear bond strength in MPa was normally distributed data means of seven groups were compared using One-Way ANOVA followed by Post Hoc Multiple Comparisons test. All statistical tests were two-sided and performed at a significance level of α=.05. Analysis was conducted using IBM SPSS STATISTICS (version 22.0).

Group	Mean Shear Bond Strength Value ± Standard Deviation
I. No Bleaching	19.30±1.81 *II,III,IV,VI,VII
II. No Antioxidant	7.83±2.21 *I, III, IV, V, VI, VII
III. 10% Sodium Ascorbate	13.4±2.15 *I, II, V
IV. 25% Alpha Tocopherol	14.9 ± 2.58 *I, II, VI
V. 6.5% Grape Seed Extract	17.01 ± 2.32 * II, III, VI
VI. 5% Lycopene	11.79 ± 3.03 * I, II, V
VII. 5% Green Tea Extract	13.72 ± 4.16 * I, II

Table 2

*Means statistically significant differences.

RESULTS: Refer to Table 2

Mean bond strength values of all the groups are as follows:

Group I No Bleaching - 19.3 ± 1.81 MPa

Group II No Antioxidant - 7.83 ± 2.21 MPa

Group III 10% Sodium Ascorbate - 13.4 ± 2.15 MPa

Group IV 25% alpha Tocopherol - 14.9 ± 2.58 MPa

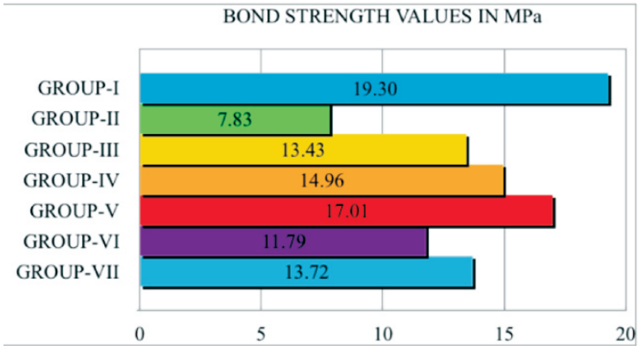
Group V 6.5% Grape seed extract- 17.1 ± 2.31MPa

Group VI 5% Lycopene - 11.79 ± 2.85 MPa

Group VII 5% Green tea extract - 13.7 ± 3.02 MPa

DISCUSSION:

The reduction in bond strength of adhesive restorations to tooth structure after dental bleaching has been investigated widely in the literature. Some authors reported that a high concentration of oxygen remains among the enamel prisms and in the dentin which prevents complete polymerization.[4]Others mentioned that vital bleaching alters the protein and mineral content of the superficial layers of enamel, leads to formation of incomplete resin tags and displayed a porous and granular view with a 'bubbly' appearancewhich may be responsible for reduced bond strength.[15]According to McGuckin et al, the void areas present at the interface decrease in bonds formed at longer time intervals after bleaching cessation, indicating a gradual elimination of hydrogen peroxide and its byproducts with consequently increase of the bond strength.[3]Hence, delay in bonding by 1-3 weeks following the bleaching procedure was recommended. But this rendered immediate reestablishment of further esthetic procedures impossible.[5,16] According to Lai et al., the antioxidant should be applied for atleast one



third of the bleaching time on the enamel surface so antioxidants were applied for 10 minutes.[8]

Sodium ascorbate probably restores the altered redox potential of the oxidized bonding substrate and allows free radical polymerization of the adhesive to proceed without premature termination, thus reversing the compromised bonding.[17] In the present in vitro study, the use of 10% sodium ascorbate solution (Group III - 13.43 MPa) as an antioxidant increased the shear bond strength when compared with Group II (7.83 MPa). But it did not increase the bond strength values to the level of unbleached enamel (Group I - 19.3 MPa). There have been studies where sodium ascorbate when used as an antioxidant completely reversed the bond strength values similar to that of unbleached enamel. But in these studies 10% carbamide peroxide was used as a bleaching agent. If we increase the hydrogen peroxide content of the bleaching agent to 37.5% as in our study, then complete reversal of bond strength values to that of unbleached enamel was not seen. This can be explained by Friere et al who indicated a direct correlation between the mass of hydrogen peroxide and that of the antioxidant agent required.[18] The result obtained in the present study is in concurrence with that of Torres et al who used 35% hydrogen peroxide.[19]

According to the material safety analyses, the level of health hazard for sodium ascorbate is higher than that of natural antioxidants. Moreover, sodium ascorbate has been found to be mutagenic for mammalian somatic cells.[20] Therefore, we should evaluate the effectiveness of natural antioxidants as substitute for their antioxidant action in reversing compromised bond strength.

Vitamin E functions as a chain-breaking antioxidant that prevents propagation of free radical reactions.[21] Furthermore, vitamin E is more oxidizing and stable than ascorbate because of its hydrophobicity.[22] However, the presence of alcohol in the vitamin E composition formulated for this study may have contributed to the good response in terms of antioxidant activity, as vitamin E is not usually miscible in water solutions.[6] The results obtained in this study are in accordance with those obtained by Whang et al, Sasaki et al and Asha Thapa et al as alpha tocopherol reversed the bond strength (Group IV - 14.96 MPa).[21,9,10]

Standardized Grape Seed Extracts (GSE) contain 74 to 78% oligomeric proanthocyanidins and less than approximately 6% of free flavanol monomers on a dry weight basis (Burdock, 2005).The antioxidant properties of GSE are primarily due to flavonoids that can perform scavenging action on free radicals [superoxide, hydroxyl, and 1,1-diphenyl-2-picrylhydrazyl (DPPH)], metal chelating properties and their effects on cell signaling pathways and gene expression. The antioxidant potential of Grape Seed Extract is twenty and fifty fold greater than those of vitamins E and C respectively.[13] Group V treated with grape seed extract yielded the highest bond strength of 17.01 MPa after antioxidant application . This result is similar to that obtained by Vidhya et al, et al, Subramonian, et al. and Abraham et al.[12,20,23] However, it was contrasting to study done by Arumugam et al., who attributed that large molecular size of oligomeric proanthocyanidin complexes decrease its diffusion and hence its antioxidant action.[11] The best results of grape seed extract in this study can be explained because of its multiple electron donation sites. They applied Lipinski's rule to justify their findings but it is used to asses the bioavailability of orally administered drugs in systemic circulation which is not the case as the antioxidants are applied topically over the bleached enamel surface. Hence, Lipinski's rule is not directly applicable in the current study.

Lycopene, a carotenoid compound, is a natural pigment synthesized by plants. It is a tetraterpene assembled from eight isoprene units composed entirely of carbon and hydrogen, containing 11 conjugated and 2 non-conjugated carbon - carbon double bonds (c = c).The application of 5% lycopene in the discussed study resulted in reversal of compromised bond strength owing to its antioxidant properties but it gave the lowest bond strength values amongst the antioxidant group. Similar result was indicated by Arumugum et al.[11] This can be explained by the low water solubility of lycopene, thereby limiting its diffusion across enamel surface to trap free radicals.

Polyphenolic compounds (mainly flavanoids) present in Green Tea Extract have demonstrated potential antioxidant properties due to their redox potential; that enable them to act in various forms such as hydrogen donors, reducing agents, nascent oxygen quenchers, and chelating metal ions in numerous food applications. Epigallocatechin gallate (EGCG) is the most active and abundant catechin in green tea. It is soluble in water and is a safe material.[13] Application of 5% Green Tea Extracts improved the bond strength values of bleached enamel which is in agreement with the results of studies done by Sharafeddin et al. and Khamverdi et al.[24,25]

The results of this study showed a high statistical significant value, it could reproduce clinical significance as well. Further clinical trials are needed to confirm these findings.

CONCLUSION

The best antioxidant activity of grape seed can be attributed to the specificity of OPCs for hy droxyl free radicals, the presence of multiple donor sites on OPCs that trap superoxide radicals and the esterification of (-) epicatechin by gallic acid in OPCs, which enhances the free radical scavenging ability. The lowest activity amongst all the antioxidantsshown by lycopene is probably due it lower water solubility despite its high antioxidant activity. The bond strength values obtained by alpha tocopherol group , lycopene group and green tea extract group were insignificantly different from each other indicating these agents have comparable action in reversing the compromised bond strength to bleached enamel.

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