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Research Article

EFFECT OF DIFFERENT ANTI OXIDANTS ON SHEAR BOND STRENGTH BETWEEN NANO HYBRID COMPOSITE AND BLEACHED ENAMEL-AN INVITRO STUDY

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A B S T R A C T

Objective: The effect of 10% sodium ascorbate, 10% pine bark extract, and 10%Aloe vera on the shear bond strength of composite resin to bleached enamel was evaluated.

Materials and Methods: Fifty recently extracted human single rooted teeth were divided into six groups of 10 teeth each. In Group I (positive control), the composite bonding was done immediately after bleaching. Except Group V (negative control), the labial enamel surface of all specimens in the other groups were bleached with 35% hydrogen peroxide followed by composite restoration. Groups II, III, and IV specimens were treated with antioxidants 10% sodium ascorbate, 10% pine bark extract, 10%Aloe vera respectively, for 10 min and bonded with composite resin. All specimens were stored in deionized water for 24 h at 37°C before shear bond strength testing by using universal testing machine.The data obtained were tabulated and statistically analyzed using analysis of variance One way ANOVA test and post hoc Tukey test.

Results: The unbleached teeth showed the highest shear bond strength followed by the bleached teeth treated with the antioxidant 10% Aloe vera.

Conclusion: Within the limitations of this study, it was observed that the use of antioxidants effectively reversed the compromised bond strength of bleached enamel. Among the antioxidants, 10% Aloe vera application after bleaching showed better bond strength.

INTRODUCTION

A smile has been said to be among man's most important interactive communication skills. A beautiful smile is an added asset for beautiful face. The public longing for white teeth has driven dentists to strive to meet their patients' expectations of dental esthetics.[1]Esthetics by definition is "The science of beauty, the particular detail of an animate or inanimate object that makes it appealing to the eye"[2]

The appearance of the dentition is of concern to a large number of people seeking dental treatment and the colour of the teeth is of particular cosmetic importance. There has been a recent increase in interest in the treatment of tooth staining and discolouration as shown by the large number of tooth whitening agents appearing on the market.[3]

Among the several aesthetic dental treatments, bleaching is a non-invasive, relatively simple procedure to be performed. Increase in the demand for aesthetic dentistry has resulted in widespread practice of vital bleaching 4. Vital tooth bleaching is considered as a safe, popular, conservative and well accepted treatment option for discoloured teeth.[5]

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Bleaching agents in varying concentrations (Carbamide peroxide 35% to 37% or Hydrogen peroxide 30% to 40%) have been used to achieve rapid aesthetic results .[5,6] When applied on tooth surface, hydrogen peroxide undergoes ionic dissociation and gives rise to the formation of free radicals such as hydroxyl radical, per-hydroxyl, nascent oxygen, and superoxide anions, which are the most potent free radicals. These are extremely reactive and therefore react with the electron-rich regions of pigment within the tooth leading to dissociation of the larger pigmented molecules into smaller and less pigmented molecules .[5,6]

Studies have shown that the shear bond strength of composite resin bonded to the tooth surface immediately after bleaching was significantly lower than that unbleached tooth surface due to the presence of residual oxygen layer. Removal of this residual oxygen layer was found to increase the shear bond strength of composite resin to bleached enamel. The general approach of overcoming this post bleaching compromised bond strength was to delay the bonding procedure by a period varying from 24 h to 3 weeks.[7,8] This period is important to eliminate the residual oxygen from dental structure and re establish the enamel bond strength.[9]

Although the waiting period has advantages, however, not always this waiting time is possible, Thus, the discussion of

other methods for reversing the reduced bond strength to enamel after bleaching also becomes important. Several methods have been proposed to reverse the compromised bond strength following bleaching such as subjecting the bleached enamel to alcohol treatment before the restoration, removing the outermost layer of enamel, and the use of organic solvent containing adhesives and use of antioxidants.[10]

Antioxidants like 10% sodium ascorbate have also been used to reverse the reduced bond strength of bleached enamel.[11, 12, 13] Application of antioxidants was reported to be a reversal in the decline of Shear Bond Strength of composite resin to bleached enamel due to its protective role against free radical reactions. [14] Sodium ascorbate, grape seed extract, green tea, pomegranate peel extract, and aloevera are some antioxidants. [15 ,16] The interest in natural antioxidants of plant origin has greatly increased in recent years. Oligomeric proanthocyanidin complexes present in natural antioxidants like grape seed extract and pine bark extract have free radical scavenging activity. [13]

The aim of this *in vitro* study was to evaluate and compare the effects of different antioxidants on the bond strength of composite resins to bleached enamel. The hypothesis of this study was that the use of an antioxidant reversed the reduced bond strength of composite resins to bleached enamel.

Preparation of Specimens

Inclusion Criteria*:* Freshly extracted human maxillary central incisor teeth, extracted for periodontal reasons.

Exclusion Criteria*:* Teeth with severe attrition, erosion, decay, fractures, cracks, dried teeth with developmental defects were excluded.

50 Non carious extracted human single rooted teeth collected from Department of Oral and Maxillo facial Surgery, CKS Theja Institute of Dental Sciences & Research were used in the study.

They had intact labial enamel and no caries. After extraction the residue on teeth was removed and washed away with tap water. Teeth were stored in sterile normal saline solution until required. The roots were removed from crowns at Cemento Enamel Junction using straight slow speed hand piece and slow speed diamond disc under copious water spray. The teeth were mounted in an aluminium holder filled with cold cure acrylic resin with labial surface facing upwards.

The labial surfaces of teeth were at same level of embedding medium to form a flat surface. Then central portion of embedded tooth was ground flat with 600 grit silicon carbide paper.

Preparation of Solutions

Four Solutions were Prepared for this study

1. 10g of Sodium Ascorbate powder, (SD Fine Chem Limited) was weighed on weighing balance and the powder was dissolved in 100ml of distilled water and then placed on stirrer to make it homogenous to form 10% Sodium Ascorbate solution.
2. 10g of Pine bark extract (Zenith Nutrition) , was weighed on a weighing balance , and the powder was dissolved in 100ml of distilled water to make 10% Pine bark extact solution, and then placed on stirrer to make it homogenous to form 10% Pine bark extact solution.
3. 100 ml Aloe vera leaf solution(Health Viva) was taken then placed on stirrer to make it homogenous to form Aloe vera leaf solution.

Grouping of Specimens

Group 1(n=10): (Positive control) 10 Specimens were subjected to Bleaching, NO Anti oxidant is applied after bleaching, only composite restoration done immediately.

Group 2(n=10): 10 specimens after application of bleaching agent, were treated with 10% Sodium ascorbate solution for 10min, followed by composite restoration immediately.

Group 3(n=10): 10 specimens after application of bleaching agent , were treated with 10% Pine bark extract solution , for 10 min , followed by composite restoration immediately.
Group 4(n=10): 10 specimens after application of bleaching agent, were treated with 10%

Aloe vera solution for 10 min, followed by composite restoration done immediately.

Group 5 (n=10) (Negative control) 10 specimens were not subjected to bleaching, nor anti oxidant application (NO bleaching done, NO Anti oxidant application) only composite restoration done .

Bleaching Procedure

Exept in Group 5, remaining all the experimental groups (Group 1, 2, 3, 4) were subjected to 35% Hydrogen peroxide (SDI Pola office) on the flattened enamel surface for 10min as per manufacturer instructions. After completion of bleaching procedure, specimens were thoroughly rinsed with distilled water and air dried. Group 1 specimens received composite restoration immediately after bleaching without anti oxidant treatment. Group 2, 3, 4 specimens received sodium Ascorbate, pine bark extract, Aloe vera as respective anti - oxidant treatment immediately after bleaching.

Anti Oxidant Treatment

After the bleaching treatment, all the specimens of Group 2,3, 4 are subjected to anti oxidant treatment. Antioxidant solutions were applied under continuous agitation using an applicator on the enamel surfaces of the embedded teeth following bleaching process In Group 2 , specimens were subjected to 10% Sodium ascorbate solution treatment for 10min. and then rinsed with distilled water. Similarly Group 3 , specimens were subjected to 10% pine bark extract solution treatment for 10min. and then rinsed with distilled water, Group 4 , specimens were subjected to 10% Aloe vera solution treatment for 10min. and then rinsed with distilled water After anti-oxidant treatment, enamel surface was thoroughly rinsed with distilled water for 30 sec and subjected to bonding procedures.

Bonding Procedures

After application of antioxidant solutions, acid etching procedure was carried out for all specimens according to manufacturer's instruction by using 35% phosphoric acid (N ETCH, IVOCLAR VIVADENT) for 15s then specimens were washed and air dried for 5sec.A total adhesive (TETRIC N BOND, IVOCLAR VIVADENT) was applied to all specimens according to manufacturer's instruction. and then light cured for 20 sec, using a Blue phase light curing unit with an intensity out put of 400-500 mW/cm2, and the intensity was verified with Radiometer . A plastic tube with

internal diameter of 2mm and 4mm height was placed onto bonded specimens and securely holded and the tube was filled with Nano Hybrid composite (TETRIC NCERAM , IVOCLAR VIVADENT) by Composite packing instruments and cured using blue phase curing unit with an intensity out put of 900-1000 mWcm2, and intensity was verified using a Radiometer.

Nano hybrid Composite was packed in two increments , and each increment is light cured for 40s. Then plastic tubes were removed after curing of composite resin. Then the height of the composite cylinder is checked with Digital vernier Calipers. The specimens are stored in deionised water at for 24 at 37°C hrs prior to Shear Bond strength testing.

Analysis of Shear Bond Strength

The shear bond strength was measured with Instron Universal testing machine. A knife edge shearing rod with cross – head speed of 0.5mm/min was used. The shear bond strengths of the specimens were calculated and expressed in MPa.

Statistical Analysis

The values obtained were statistically analyzed usingcomputer software Statistical Package for Social Sciences(SPSS) version 22. One-way analysis of variance (ANOVA)followed by Post Hoc Tukey test was used to analyze the data. Significance was established at $P < 0.01$ level.

RESULTS

Table 1 Mean and SD of Shear bond strength in various groups

Group	N	Minimum	Maximum	Mean	Std. Deviation
Group 1	10	7.23	11.98	9.72	1.56
Group 2	10	19.14	25.78	22.19	2.31
Group 3	10	21.42	28.73	25.54	2.57
Group 4	10	26.37	35.06	31.11	2.80
Group 5	10	31.06	41.44	36.93	3.50

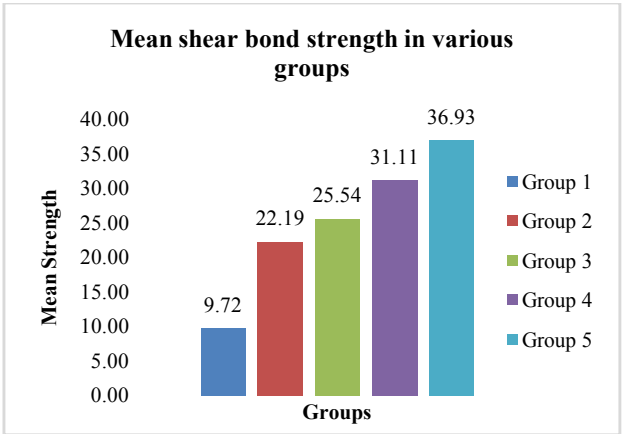


Table 2 Intergroup comparison of mean Shear bond strength using one way ANOVA test

Comparison	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	4213.995	4	1053.499	152.627	<0.001**
Within Groups	310.609	45	6.902		
Total	4524.604	49			

**-Statistically highly significant (P<0.01)

Inference

There is statistically significant differences present in the mean bond strength values among the various groups compared. ANOVA table signifies overall summary of the comparison to know the comparisons between the individual groups we have to perform post hoc Tukey test.

Table 3 Post Hoc tukey test for individual intergroup comparisons

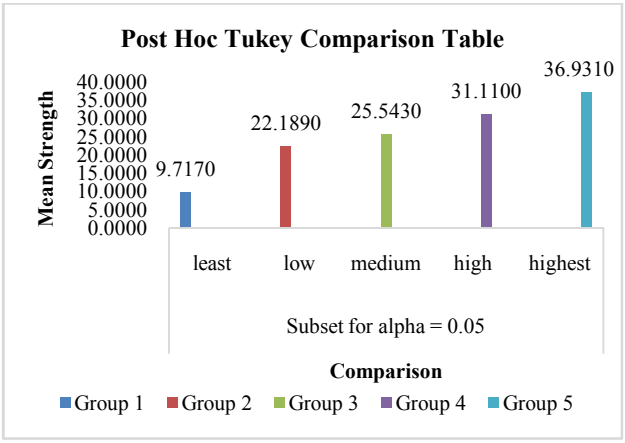
Comparison	Mean Difference	Sig
Group 1 vs Group 2	12.47200*	<0.001**
Group 1 vs Group 3	15.82600*	<0.001**
Group 1 vs Group 4	21.39300*	<0.001**
Group 1 vs Group 5	27.21400*	<0.001**
Group 2 vs Group 3	3.35400*	0.048*
Group 2 vs Group 4	8.92100*	<0.001**
Group 2 vs Group 5	14.74200*	<0.001**
Group 3 vs Group 4	5.56700*	<0.001**
Group 3 vs Group 5	11.38800*	<0.001**
Group 4 vs Group 5	5.82100*	<0.001**

*-Statistically significant (p<0.05), **- Highly significant (p<0.01)

Inference

The following Inferences can be drawn from the above table

1. There are statistically significant differences present in the mean Shear bond strength values when comparisons are made between Group 1 with all other groups individually (p<0.05)
2. There are statistically significant differences present in the mean Shear bond strength values when comparisons are made between Group 2 with all other groups individually (p<0.05)
3. There are statistically significant differences present in the mean Shear bond strength values when comparisons are made between Group 3 with all other groups individually (p<0.05)
4. There are statistically significant differences present in the mean Shear bond strength values when comparisons are made between Group 4 with all other groups individually (p<0.05)
5. There are statistically significant differences present in the mean Shear bond strength values when comparisons are made between Group 5 with all other groups individually (p<0.05)



The order of Mean Shear bond strengths is

Group 5 > Group 4> Group 3> Group 2> Group 1

DISCUSSION

Smile is a universal means of showing welcome, friendship and acceptance. The creation and maintenance of a beautiful

smile has always been a part of dental practice. Patients demand not only a healthy mouth but also what they consider is a perfect smile. A healthy, bright and beautiful smile does have an impact on individual beauty, self-consciousness, self-image as well as self-confidence. With the increasing demand for a white smile, conservative techniques such as bleaching for the treatment of discoloured teeth has gained importance .[17] Many authors have investigated and published the effects of bleaching on bond strengths of composite to enamel and dentin. For brightening discolored teeth, the use of peroxide releasing agents such as hydrogen peroxide, carbamide peroxide or sodium perborate has become a popular treatment modality, which is comparatively safe. [18] This is possible because hydrogen peroxide is of low molecular weight and can denature proteins, which increases tissue permeability and allows ions to move through the tooth. Studies show that bleaching causes changes in the organic enamel matrix, a loss of calcium and a decrease in micro-hardness that are potential causes for the reduction of bond strength.[18, 19]

It is further claimed that residual oxygen, released by the bleaching agent, interferes with resin infiltration and subsequently affects bond strength. [20] Treatment of bleached teeth is challenging for dentists because they cannot immediately perform a resin restoration on bleached teeth due to the presence of oxygen or peroxide residues on the surface, since they prevent complete polymerization of adhesive resin . [21.] However, by postponing the composite restoration for two weeks following bleaching, no reduction in bond strength would occur .[22 ,23, 24] But, sometimes it is not possible for the patient to wait that long. Therefore, use of antioxidants like ascorbic acid or sodium ascorbate is one method to immediately increase the bond strength of composite to bleached enamel.[15,25]

It has been demonstrated that use of herbal antioxidants such as green tea and grape seed is an effective alternative strategy for this purpose.[24] Ascorbic acid and its compounds are derivatives of Vitamin C and are well‑known antioxidants. They have been shown to have the ability to quench the reactive free radicals in biological systems.[26] Lai *et al*. hypothesized that application of an antioxidant may inhibit the incorporation process of the peroxide ions itself, thereby limiting the structural aberrations. The antioxidant (sodium ascorbate) restores the altered redox potential of the oxidized bonding substrate thereby reversing the compromised bonding. Thus, the main purpose of antioxidant treatment of the teeth post bleaching is to eliminate any residual oxygen trapped inside the dental hard tissues, thereby neutralizes the oxidizing effects of the bleaching agent.[27] Thus, the results obtained in this study are in agreement with these findings as the bond strength of the teeth treated with sodium ascorbate postbleaching was found to be increased significantly.[28]

Oligomericproanthocyanidins are a class of polyphenolic bioflavonoids found in fruits and vegetables and are present in grape seed extract, pine bark extract, cranberries, lemon tree bark, hazel nut tree leaves, etc. They have free radical scavenging and antioxidant activity. They also have antibacterial, antiviral, anti-inflammatory, antiallergic, anticarcinogenic, and vasodilatory actions.[29] Pine bark extract contains phenolic compounds broadly divided into monomers (catechin, epicatechin, and taxifolin) and condensed flavonoids (oligomeric to polymeric proanthocyanidins).[30]

Flavonoids can perform scavenging action on free radicals like superoxide, hydroxyl, and 1,1-dipheny l-2-picrylhydrazyl (DPPH) and have metal chelating properties. The presence of the functional group OH in the structure and its position on the ring of the flavonoid molecule determines its antioxidant capacity. [31]The antioxidant properties of grape seed extract and pine bark extract could be due to the flavonoids present.[32]

Aloe Vera is one of the most versatile plants on the earth. The Egyptians called Aloe “the plant of immortality.” Today, the Aloe Vera plant is being used for various purposes. Pharmacologically it shows antibacterial, anti-inflammatory and antioxidant properties . [33]In this study leaf extract of aloe vera was used as an antioxidant, as it contain ascorbic acid along with tocopherol and vitamin A. It also contains vitamin B12, folic acid, and choline. Its antioxidant properties neutralize free radicals. Moreover anthraquinones present in the aloe vera reported to exert antioxidant property .[33,34 ,35]. It contains calcium, chromium, copper, selenium, magnesium, manganese, potassium, sodium and zinc. These are essential for the proper functioning of various enzyme systems in different metabolic pathways and few are antioxidants.[34] Antioxidants like 10% sodium ascorbate have also been used to reverse the reduced SBS of bleached enamel.[4,5]

Since, there is a limited information available on the use of the newer natural antioxidant agents like Oligomeric Proanthocyanidin Complexes (OPCs) that they increases the bond strength of composite resin to bleached enamel. OPCs present in natural antioxidants like grape seed extract and pine bark extract have free radical scavenging activity [4 ,36]. Hence, the aim of this in-vitro study was to evaluate and compare the effect of 10% Sodium Ascorbate , 10 % pine bark extract, and Aloe vera leaf solution on the SBS of composite resin to bleached enamel.

The interest in natural antioxidants of plant origin has greatly increased in recent years. Oligomeric proanthocyanidin. complexes present in natural antioxidants like grape seed extract and pine bark extract have free radical scavenging activity. [32, 37]

Antioxidants used in this study were capable of reversing the compromised bond strength of composite resins to bleached enamel. So this could be used to avoid the waiting period before bonding to bleached enamel which makes it clinically significant. Even though studies on antioxidants showing their capability of reversing bond strength to bleached enamel has been done, it has not been compared with pine bark extract and Aloe vera in this aspect. However, this being an *in vitro* study, it cannot mimic the *in vivo* conditions. In the oral cavity, the interface between the restoration and the tooth is exposed to diverse forces. that act simultaneously. During its life time, a restoration is subjected to cyclic loading; each load is insufficient to provoke failure, but in the long-term can possibly lead to marginal deterioration and loss of restoration. Therefore, fatigue testing of dental adhesives is expected to better predict their *in vivo* performance. Nevertheless, the more expensive and long-lasting clinical trials remain necessary to validate the laboratory observations. Since 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

Under the limitations of this in-vitro study, it can be concluded that, treatment of the bleached enamel surface with 10% sodium ascorbate ,or 10% pine bark extract or 10% Aloe vera reverses the reduced Shear Bond Strength of composite resin to bleached enamel.

Among antioxidants, the use of 10% Aloe vera significantly increases the SBS of composite resin to bleached enamel than that of 10% pine bark and 10% sodium ascorbate solution.

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