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ASSESSMENT OF THE MICRO-TENSILE BOND STRENGTH OF RESIN TO BLEACHED ENAMEL USING DIFFERENT ANTIOXIDANTS

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ABSTRACT

Background and Aim: Microtensile bond strength testing using non-trimming method has been accepted as the gold standard for investigating integrity of an adhesive tooth-resin bond. Present study was done with an aim to compare and evaluate the micro-tensile bond strength of resin to bleached enamel using different antioxidants. **Material and Methods:** One hundred fifty four extracted permanent incisors without caries, cracks and fractures were obtained, cleaned of debris and stored in saline. The application of antioxidant agents after bleaching has been recommended as a way to reduce the waiting period between bleaching and restorative procedure by eliminating the reactive oxygen from the dental substrate. The specimens were randomly divided into one control and six experimental groups, with 22 samples in each group. **Results:** The mean micro-tensile bond strength values are highest for group-1 (15.90 MPa) and least for group-2 (5.10MPa). The mean values are in descending order group1 followed by group-7, group-3 and group-4, group-5, group-6, group-2. There was no statistical difference in micro-tensile bond strengths between group-1, group-3, group-4, group-5 and group-7. There was a statistical significant difference between group-1, group-2 and group-6. **Conclusion:** There was significant improvement in the bond strength of composite to bleached enamel was seen with the application of antioxidant agents. Further, among the experimental groups Quercetin application showed highest mean bond strength values when compared to others.

KEYWORDS: Antioxidant Agent, Bond Strength, Enamel, Microtensile bond strength.

INTRODUCTION

Smile is considered as a person's ability to express a range of emotions with the structure and movement of the teeth and lips.[1] One of the most common clinical issues related to aesthetics is pigmentation of dental structures. Mankind has always tried to improve the facial features, for looking appealing, since the appearance and alignment of teeth influence the personality of a person. With increasing understanding of aesthetic dentistry, bleaching has become a very common procedure.[2]

Tooth discoloration is caused by intrinsic and extrinsic factors, which can be treated by bleaching, enamel microabrasion, porcelain veneers, crowns.[3,4,5] Hydrogen peroxide (HP), an oxidizing agent diffuses into the tooth and dissociates to produce free radicles which attack the organic pigmented molecules within the tooth tissue. As a result, the chromophore molecules become smaller and less heavily pigmented and there will be a shift in the absorption spectrum, thus resulting in the bleaching of the tooth.[6,7] It has been proposed that HP penetrates enamel and dentin to reach the pulp cavity, and the residual oxygen from the bleaching agent inhibits resin polymerization. So, to prevent the deleterious effects of peroxide on the resin, a waiting period of 1-3 weeks is recommended.[6] However, due to the immediate aesthetic requirements of the patients, other methods of reversing the reduced bond strength of enamel after bleaching becomes important.

It has been proposed that residual oxygen from the bleaching process remains within the tooth structure for up to 2 weeks and is believed to interfere with polymerization of the dentin adhesives.[8] Although the immediate negative effect of bleaching on bond strength have been known through multiple studies, it is reported that waiting for at least 7 days, dentin bond strength values return to baseline.[9-11] Although bleaching effects on dentin aren't long-term, antioxidants like sodium ascorbate is used to counteract the adverse effects of bleaching to permit immediate placement of a restoration.[12-14]

Several studies have found no significant difference between the use of antioxidant surface treatment immediately after the bleaching procedure and delaying the restorative bonding for at least two weeks. So the application of antioxidant agents after bleaching has been recommended as a way to reduce the waiting period between bleaching and restorative procedure by eliminating the reactive oxygen from the dental substrate.[15,16]

Microtensile bond strength testing using non-trimming method has been accepted as the gold standard for investigating integrity of an adhesive tooth-resin bond.[17] Investigators in previous reports have ground or sandblasted the surface enamel to create a more even or flat bonding surface, however this results in issues of depth of enamel reduction, differences in surface and subsurface enamel and dentine encroachment. Enzymatic agents such as Super Oxide dismutase and non-enzymatic agents such as Sodium Ascorbate (SA), Flavonoids, Lycopene, and Vitamin E have antioxidant properties.

Present study was done with an aim to compare and evaluate the micro-tensile bond strength of resin to bleached enamel using different antioxidants.

MATERIAL AND METHODS

One hundred fifty four extracted permanent incisors without caries, cracks and fractures were obtained, cleaned of debris and stored in saline. Later, prophylaxis of the teeth was carried out and disinfected in 0.9% thymol for 24 hours. The crowns were sectioned from the root using a lowspeed rotary saw under water irrigation. The crown specimens were embedded in self-cure acrylic resin with the labial surfaces parallel to the horizontal plane. The buccal surfaces were flattened with silicon carbide sandpaper with decreasing granulations under abundant irrigation with water for 10 seconds with each sand paper and for 60 seconds with the 600 grit sandpaper, to obtain a flat surface and maintain approximately 1.5 mm of enamel remnant. The specimens were randomly divided into one control and six experimental groups, with 22 samples in each group.

Group 1: Control group

In this group, unbleached specimens were stored in distilled water for one week and etched with Gluma etch (KULZER) for 20 sec and water sprayed to completely rinse off. The enamel surface must be dried and should appear chalky white, followed by the application of bonding agent and light-cured for 20 seconds with Bluephase N at 400- 500mw/cm. A Teflon mold of (5mmx5mmx5mm) was placed approximately in the center of the labial surface and 5mm thick submicron hybrid composite (CHARISMA) was incrementally cured for 20 seconds. All the specimens were stored in distilled water untill they were tested.

Group 2: Bleached and restored immediately

In this group, teeth were exposed to a 1mm layer of 37.5% of HP gel for 8 minutes and thoroughly cleaned with a soft micro-brush under flowing distilled water and placed in artificial saliva for remaining time of the day. A total of 5 applications were done with one-day interval. At the end of 5 th day, they were restored with composite similar to that of the control group protocol and stored in distilled water.

Group 3: Bleached and restored after one week

In this group, teeth were bleached similar to the protocol which was followed in group-2 and stored in distilled water for one week, later composite build up was done following the same protocol as in control group and stored in distilled water.

Group 4: Bleached and 10%Sodium Ascorbate (SA) application

In this group, teeth were bleached similar to the protocol which was followed for Group-2. 10% SA solution was freshly prepared by adding 10gm of SA powder in 100ml of distilled water. The Solution was applied onto the bleached surface with the help of micro-brush and after 10 min the specimens were rinsed and dried and restored with composite similar to that of the control group and stored in distilled water.

Group 5: Bleached and 5% Proanthocyanidin (PA) application

In this group, teeth were bleached similar to the protocol which was followed for Group 2.5% PA solution was freshly prepared by adding 5gm of PA powder to 100ml of distilled water. The Solution was applied for 10 minutes on bleached enamel and rinsed with a jet of water and restored with composite similar to that of the control group and stored in distilled water.

Group 6: Bleached and Super Oxide Dismutase (SOD) application

In this group, teeth were bleached similar to the protocol which was followed for Group 2. After the bleaching protocol, SOD readily available solution was applied and rinsed after 10 minutes on bleached enamel and they were restored with composite similar to that of the control group and stored in distilled water.

Group 7: Bleached and 6.5% Quercetin application

In this group, teeth were bleached similar to the protocol which was followed for Group 2. Quercetin solution was freshly prepared by adding 10gms of Quercetin powder in 10ml of Dimethylsulfoxide and 40 ml of Phosphate Buffered Saline. Solution was applied on bleached enamel and rinsed with a jet of water after 10 minutes and was restored with composite similar to that of the control group and stored in distilled water.

The composite was placed on the enamel surfaces incrementally using a resin instrument to create a block of approximately 5x5x5 mm. Each increment of 2mm composite resin was cured for 20 seconds. The bonded specimens were placed in distilled water at 37ºC for 24 hours. Subsequently, the enamel-composite bonded specimens were sectioned occluso-gingivally into slabs using hard tissue microtome. Sectioning the slabs mesiodistally generated multiple —resin-enamel-dentin beams with a cross sectional surface area of approximately (1.2 x 1.2 x 8mm) + 0.2mm. Molds of PVC-Poly vinyl chloride pipes were made to prepare acrylic cylinders of diameter 1 cm and height 3.5cm. Three-fourth of the cylinder was filled with self-cure acrylic in dough stage, and one end of the tooth resin rod was embedded and remaining one-fourth of the cylinder was filled self-cure polymer and monomer in sprinkle on method. Similarly the other end of the tooth-resin rod was also embedded in another acrylic cylinder as mentioned above. Both ends of resin tooth rod were embedded in 2 acrylic blocks, such that 3mm of resin-tooth rod was exposed, which consisted of 1.5mm of enamel and 1.5mm of resin. The tooth–resin rod was secured with a cyano-acrylate resin to a modified testing jig attached to UTM-Universal testing machine. The specimens were stressed in tension mode at a crosshead speed of 0.5mm per minute until failure. The failure strength was determined as the quotient of Load on failure / Adhesive area (A) and was recorded in MPa.

Statistical analysis

The recorded data was compiled and entered in a spreadsheet computer program (Microsoft Excel 2007) and then exported to data editor page of SPSS version 15 (SPSS Inc., Chicago, Illinois, USA). For all tests, confidence level and level of significance were set at 95% and 5% respectively.

RESULTS

The mean micro-tensile bond strength values are highest for group-1 (15.90 MPa) and least for group-2 (5.10MPa). The mean values are in descending order group1 followed by group-7, group-3 and group-4, group-5, group-6, group-2. (Table 1). There was no statistical difference in micro-tensile bond strengths between group-1, group-3, group-4, group-5 and group-7. There was a statistical significant difference between group-1, group-2 and group-6.

The results of stereomicroscopic examination revealed three types of failure modes adhesive, cohesive, and mixed. Adhesive failure indicates that the failure occurred due to lack of sufficient bond strength of the composite material to the tooth. A cohesive failure on the other hand, indicates that the bond strength was greater than the strength of composite materials used. A mixed failure generally indicates when the bond strength and strength of the supporting composite materials were close enough to each other to exhibit both failure modes.

Group-1 (control) showed 39% adhesive failures, 19%cohesive failures, 39% mixed failures. Group-2 (immediate bonding after bleaching) showed 59% adhesive failures, 12% cohesive failures, 26% mixed failures. Group-3 (delayed bonding of bleached enamel) showed 46% adhesive failures, 26% cohesive failures, 39% mixed failures. Group-4 (SA) showed 32% adhesive failures, 39%cohesive failures, 26% mixed failures. Group-5 (PA) showed 32% adhesive failures, 26% cohesive failures, 39% mixed failures. Group-6 (SOD) showed 39% adhesive failures, 12%cohesive failures, 46% mixed failures. Group-7 (Quercetin) showed 39% adhesive failures, 46% cohesive failures, 26% mixed failures. Adhesive failure occurred purely at the restorative/ dentin interface resulting in reduced bond strength between resin and dentin which is unfavorable. In the present study, cohesive failure within the restorative material exhibited by group-7 (Quercetin) and group-4 (SA) are considered favorable compared to group-6,5and 2 which showed more mixed failures and is least desirable in the clinical conditions.

Table 1: The mean and standard deviation values and confidence intervals of microtensile bond strength in (MPa).

Groups	N	Mean	SD	P value
Group 1	22	15.90	4.1	0.001*
Group 2	22	5.10	1.20	
Group 3	22	15.20	5.2	
Group 4	22	15.09	4.02	
Group 5	22	12.02	7.6	
Group 6	22	6.01	1.8	
Group 7	22	15.98	5.65	

*** indicates statistically significance at p≤0.05 Test applied ANOVA**

DISCUSSION

In the past, investigators have used ground flat enamel surface for bonding rather than the intact enamel surface used in the present investigation. The amount and depth of enamel removed during grinding procedure can be difficult to standardize as shape and curvature of individual teeth vary and may well lead to dentine encroachment. Bar specimen from the coronal 2/3rd of the enamel was subjected to microtensile testing, as bond strengths of RBC to enamel from the occlusal third are known to be significantly higher than those made to the cervical enamel.[18]

Although bleaching has proved to be meritorious in different ways, de Lima et al. (2009) and Dias Ribeiro et al. (2009), in their studies stated that it may present certain undesirable side effects such as pulp toxicity, microleakage, external root resorption, changes in the structure of tooth, decrease in microhardness as well as the bond strength reduction of composite resin to tooth structures. After the bleaching treatment, patients usually desire for a more esthetic procedures such as laminate veneers or composite resin restoration for restoring deficiencies.[19] The residual oxygen remaining on tooth surfaces after bleaching as stated by Rueggeberg and Margeson, (1990) can interfere with adhesion of the adhesive system applied for restorative procedures. All in all, the decrease in bond strength may evoke several other factors which include an increase in microleakage, changes in fracture resistance as given by Khoroushi and Fardashtaki, (2009)[20] and increase in tooth surface porosity stated by Titley et al. (1991).[21]

However, Titley et al.[21] showed that the bleaching process may result in residual peroxide, which has an immediate adverse effect on bond strength of adhesive restorations to tooth structures. In a systematic review by Feiz A et al. (2017)[22], the results obtained showed that the use of antioxidant agents, regardless of their type, form, concentration and duration of application, can improve the shear bond strength after bleaching. However, to ensure adequate bond strength using two different bonding strategies i.e total etch technique and self-etch technique can be evaluated by testing the microtensile bond strength.

Post-operative sensitivity is one of the most common complications of bleaching, which accounts for 62.5%. Clinical researches reported a prevalence of 18-78% of sensitivity either with at-home tray delivery or in office procedures.[23,24] Another complication of bleaching is decreased bond strength of composite resin to enamel. The decrease in the bond strength is significant when the composite restoration is performed immediately after bleaching. It was observed that bleaching may increase the solubility of glass-ionomer and other cements. The bond strength between enamel and resin based fillings was reduced by 40-55% when bonded immediately to bleached enamel.[10,11]

Various techniques have been recommended to counteract the impaired resin-adhesion problem. Commonly recommended solution is delaying the restorative treatment by 1-3 weeks to allow the degradation of residual oxygen.[25] According to some authors, the recommended waiting period for restorative procedures on enamel after bleaching with 10% CP is 24 hours.[26] Some authors suggested a waiting period of one week.[27] Many authors claimed that a period of 24 hours is not enough to re-establish the bond strength to enamel for higher concentrations like 9.5, 25,30,35 and 38% HP and at least 1-3 weeks is necessary.[14,15,21]

Among the various anti-oxidants used, Sodium Ascorbate is the most commonly used antioxidant which became the standard of care. Another antioxidant is άTocopherol, which is the most active component of vitamin E Complex. Quercetin is one of the flavonoids with antioxidant property. It is available in fruits and vegetables. Various storage medium can be used to store the testing specimens like distilled water, saline, artificial saliva or formaldehyde solution.[28] Saline was used in our study to store the teeth after extraction, surface preparation and after specimen preparation as other solutions were reported to effect the resultant bond strength. The teeth used for in vitro bonding studies are mainly obtained from humans and bovines. In the present study, a crosshead speed of 0.5mm/min was chosen as no clear advantage or relationship of scatter of data was observed using lower cross head speeds to debond the specimens. Under the experimental conditions of the present study the results demonstrate that the mean micro-tensile bond strength value of group I (Control) is higher than the experimental groups with no statistical difference between control group and Quercetin. This was in accordance with the study done by Mana Shamsed in et al. who observed that these experimental concentrations of Quercetin were capable of improving the SBS to normal levels.

In our study, the mean micro-tensile bond strength of group 2 was least when compared to those of control and experimental groups with a statistically significant difference. There is 68.5% decrease in bond strength when compared to group 1. This is in accordance with the studies done by Amal Suleiman et al, Fahim et al, Moosavi et al. who observed that peroxides and their by-products present inside the dental structure are capable to interfere with the polymerization process of the adhesive material thereby decreasing the bond strength.

In the present study the mean micro-tensile bond strength of group 3 was 5.6% lower than the control group and there was no statistical difference between group 3 and 4. There was statistical difference present between group 3 and 2. These were in accordance to the previous studies Murad et al, Mehdi et al, Hamid et al. They concluded that the anti-oxidizing ability of SA helped to neutralize and reverse the oxidizing effects of the bleaching agent.

In the present study there was no significant difference in the mean micro-tensile bond strength values of group 4 (SA) and group 5(PA) which was in accordance with the previous studies. Zahra et al, Vidya et al, Manoharan et al, observed that the specificity of OPC for hydroxyl free radicals and the presence of multiple donor sites on OPC trap superoxide radicals.

In the present study, the mean micro-tensile bond strength values of group 6 (SOD) were less than group 5 (PA). This was in contradictory to the study done by Megha Nair et al, who observed that the application of SOD to the bleached enamel produced higher µTBS values than the application of PA.

There was 68.5% reduction of bond strength values of group 2 compared to that of control group. There was a difference of 5.6% in mean µTBS values between group 3and 1, with group 1 higher than that of group 3.

CONCLUSION

There was significant improvement in the bond strength of composite to bleached enamel was seen with the application of antioxidant agents. Further, among the experimental groups Quercetin application showed highest mean bond strength values when compared to others. Application of Quercetin resulted in bond strength nearly equal to the control group. In terms of mode of failure, Quercetin application group showed more favorable kind of failure than other groups.

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