EGYPTIAN
DENTAL JOURNAL

Vol. 63, 3843:3851, October, 2017

I.S.S.N 0070-9484

Fixed Prosthodontics, Dental materials, Conservative Dentistry and Endodontics

www.eda-egypt.org • Codex : 166/1710

THE EFFECT OF ANTIOXIDANTS ON THE BOND STRENGTH OF RESIN COMPOSITE TO BLEACHED ENAMEL

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ABSTRACT

Aim : This study aimed to assess the effect of treating bleached enamel with antioxidants on the value of shear bond strength of resin composite to enamel. Also, a comparison was set between the natural antioxidant used (5% pomegranate solution) and the chemical one (10 % sodium ascorbate gel).

Materials and Methods: Fifty six human freshly extracted sound upper anterior teeth were used. The specimens were randomly divided into four main groups as follow : Group I : control(no bleaching) ; Group II : bleached then treated with the chemical antioxidant ; Group III : bleached then treated with the natural antioxidant; Group IV : bleached with no antioxidant treatment. Each group was further sub-divided into two sub-groups according to whether the restoration was performed immediately or delayed (after one month incubation in artificial saliva) (n=7). The variables were as follow: No bleaching immediate restoration (control) ; no bleaching delayed restoration after incubation for one month in artificial saliva ; bleached followed by application of a chemical antioxidant (10% sodium ascorbate gel) for 10 min then immediate restoration;bleached followed by application of a chemical antioxidant (10% sodium ascorbate gel) for 10 min then delayed restoration; bleached followed by application of a natural antioxidant (5% pome granate solution) for 10 min then immediate restoration ; bleached followed by application of a natural antioxidant (5% pome granate solution) for 10 min then delayed restoration ; bleached with no application of an antioxidant then immediate restoration ; bleached with no application of an antioxidant then delayed restoration. The data were statistically analyzed using two way ANOVA test, one way ANOVA test, Tuckey's post-hoc tests and pair wise student T-test.

Results : Regarding the mean shear bond strength values (immediate restoration) ; the control sub-group (immediately restored) demonstrated the highest values followed by the bleached sub-groups treated with the natural antioxidant (5% pomegranate solution), then the bleached sub-groups treated with the chemical antioxidant (10% sodium ascorbate gel). The bleached sub-groups that were not treated with the antioxidants showed the least values. Regarding the sub-groups that undergone a delayed investigation ; the sub-groups treated with the natural antioxidant demonstrated the highest shear bond strength values followed by the sub-groups treated with chemical antioxidants then the control group while the bleached sub-groups that were not treated with the antioxidants showed the least values of mean shear bond strength

KEYWORDS : Antioxidants - bond strength – bleached enamel.

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INTRODUCTION

Today we are living in a world of increasing demand for cosmetic dentistry. Bleaching is considered a rapidly growing and important dimension of esthetic dentistry. [1-3] Apart from the various advantages offered by bleaching agents, there are few complications, the most important of which is the decreased bond strength of resin composite to enamel. [1,4-9] It was reported that the presence of oxygen ions affected the process of resin polymerization.[1,10-12] In order to remove these residual oxygen radicals from enamel, various methods were suggested eg., delaying restoration with resin composite one to three weeks after the bleaching procedure; [1] the use of antioxidants in order to reverse the procedure; [1-3, 7, 13-5] performing an alcohol treatment before restoration;[16] removal of superficial enamel;[17] and using adhesives containing organic solvents. [18-19] However according to what was previously mentioned, it was reported that using antioxidants to reverse the bleaching procedure effect on the bond strength of resin composite to enamel, was the most effective method.[1-3,7,13-15] It was further reported that the antioxidants demonstrated a great potential to scavenge the free residual oxygen radicals.[1-3,7,13-45,20-12] Of the suggested antioxidants were, proanthocyanidin, a grape seat extract ;[2,20,21] sodium ascorbate ;[1-5,24,25] and lycopene.[22,23] In this study an assessment was set regarding the use of two antioxidants;a chemical one (sodium ascorbate 10%) and a natural one (pomegranate 5%) to counter act the proposed reduction of bond strength of resin composite to enamel after bleaching. The assessment included the use of antioxidants followed by restoration of resin composite immediately after bleaching, or delayed restoration and both immediate and delayed resin composite restoration without the use of antioxidants. That in addition to immediate and delayed restorations with no bleaching procedure at all (control). A comparison was set among the different parameters of the test. The null hypothesis was to prove that the use of the antioxidants resulted in recovery of the reduced shear bond strength of resin composite to bleached enamel. Also it was to prove that effect of the natural antioxidant surpassed that of the chemical one.

MATERIALS AND METHODS

Specimens preparation

56 human freshly extracted sound upper anterior teeth were used in this study. The teeth were extracted for periodontal reasons in persons of the out patients of the dental clinics of the College of Oral and Dental Surgery – Misr University for Science and Technology. All teeth were cleaned ultrasonically, polished with silicon carbide discs to produce a flat enamel surface available for the bonding procedure, mounted on acrylic blocks and then stored in isotonic saline until use.

Grouping of samples

The specimens were randomly divided into four main groups as follow : Group I : control(no bleaching) ; Group II : bleached and treated with the chemical antioxidant ; Group III : bleached and treated with the natural antioxidant; Group IV : bleached only with no antioxidant treatment. Each group was further sub-divided into two sub-groups according to whether the restoration was performed immediately or delayed (after one month incubation in artificial saliva) (n=7). The variables were as follow:

- Control : No bleaching, immediate restoration with resin composite.
- No bleaching, delayed restoration with resin composite after one month incubation in artificial saliva.
- Bleached, followed by application of the chemical antioxidant (10% sodium ascorbate) for 10 min, then immediately restored with resin composite.
- Bleached, followed by application of the chemical antioxidant (10% sodium ascorbate)

for 10min, then delayed restoration with resin composite after one month incubation in artificial saliva.

- Bleached, followed by application of the natural antioxidant (5% pome granate solution) for 10 min, then immediately restored with resin composite.
- Bleached, followed by application of the natural antioxidant (5% pome granate solution) for 10 min, then delayed restoration with resin composite after one month incubation in artificial saliva.
- Bleached, no application of antioxidants, immediate restoration with resin composite.
- Bleached, no application of antioxidants, delayed restoration with resin composite after one month incubation in artificial saliva.

Incubation in artificial saliva

In case of delayed restoration sub-groups, teeth were incubated in artificial saliva at 37 ºC for one month during which the solution was changed daily. Before restoration the specimens were thoroughly cleaned with an air-water spray for 30s. The composition of artificial saliva was as follow : 1g sodium carboxymethyl cellulose, 4.3g xylitol, 0.1g potassium chloride, 40mg potassium phosphate, 5mg calcium chloride, 1mg potassium thiocyanate and 100g distilled deionized water with a pH 7. [5]

The bleaching method

Bleaching of specimens was performed using the cosmetic tooth light whitening (White smile – Germany) with 22% HP (3x10min)

Method of application of the prepared chemical antioxidant

Sodium ascorbate (10%) was applied to specimens belonging to group II using a brush. It was left for 10min then, rinsed and dried. Sodium ascorbate was prepared by adding sodium hydroxide to ascorbic acid. 10g of the resultant sodium ascorbate (as powder) were dissolved in 100ml distilled water to provide a 10% sodium ascorbate gel.

Method of application of the prepared natural antioxidant

Pomegranate extract powder was prepared in the Faculty of Pharmacy-Cairo University. 5g of pomegranate extract powder were dissolved in 100ml distilled water to produce a 5% pomegranate solution. This pomegranate solution was applied to specimens belonging to group III immediately after bleaching, left for 10min then rinsed and dried.

TABLE (1) Materials used in this study, components, batch no & manufacturer

Material	Components	Batch no	Manufacturer
3M Filtek™ Z250	In organic filler Zirconia / Silica (60% by volume) Matrix : BIS-GMA,UDMA and BIS-EMA resins	535294	3M ESPE Dental products St Paul, MN, USA
3M™ESPE™ Single Bond	Phosphoric acid etchant A vial of adhesive	N 628066	3M ESPE Dental products St Paul, MN, USA
WHITEsmile[e]	2x2.5 ml LIGHT WHITENING AC 22% HP 1x3 g-Gingiva Protector 1x3 ml-After Whitening Mousse	17037	WHITEsmile Germany

The bonding procedure

All specimens were acid etched using 35% phosphoric acid gel for 15sec according the manufacturer's instructions. The specimens were then rinsed and dried. The bonding agent (Single bond, 3M ESPE, USA) was applied to each specimen according to the manufacturer's instructions. Then a polyethylene cylindrical tube of diameter 2mm and height 2mm was placed on the uncured bonding agent. Light curing was then performed for 20s. Then the cylindrical tube was filled with resin composite (Filtek Z 250XT, 3M ESPE, USA) (Table1). Light curing was performed for 40s.

Shear bond strength testing

A circular interface shear test was designed to evaluate the bond strength. All samples were individually and horizontally mounted on a computer controlled materials testing machine (Model 3345; Instron Instruments, USA) with a loadcell of 5 kN and data were recorded using computer software (Blue-Hill; Instron Instruments). Samples were secured to the lower fixed compartment of testing machine by tightening screws. Shearing test was done by compressive mode of load applied at enamel substrate -resin interface using a mono-bevelled chisel shaped metallic rod attached to the upper movable compartment of the testing machine traveling at cross-head speed of 0.5 mm/min (fig1). The load required to debonding was recorded in Newtons. The shear bond strength was calculated as follow :the load at failure was divided by bonding area to express the bond strength in MPa: $\tau = P/ \pi r^2$ where τ =shear bond strength (MPa), P =load at failure(N), π =3.14 and r=radius of resin disc(mm).

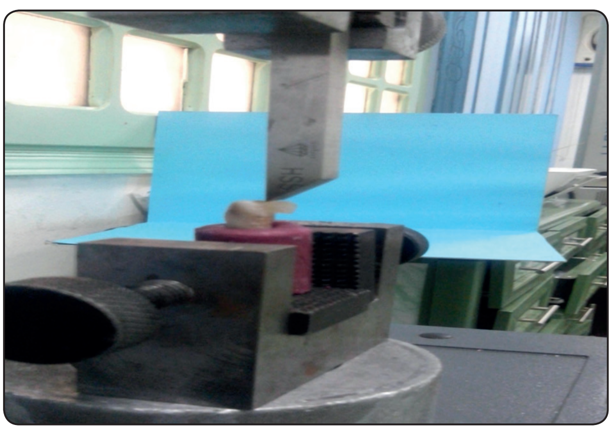

Fig. (1) : Shear bond strength testing.

Statistical analysis

Data analysis was performed in several steps. Initially, descriptive statistics for each group results. Two-way analysis of variance ANOVA test of significance was done for comparing variables (main groups, and storage time) affecting mean values. One way ANOVA followed by pair-wise Tukey's post-hoc tests were performed to detect significance between subgroups. Pair-wise student t-test was performed to detect interaction between variables of significant effect. Statistical analysis was performed using Asistat 7.6 statistics software for Windows (Campina Grande, Paraiba state, Brazil). P values ≤ 0.05 are considered to be statistically significant in all tests.

Shear bond strength results (Mean±SD) measured in mega Pascal (MPa) for all groups with different anti-oxidant (AO) and storage time are summarized in table (2) and graphically drawn in figure (2)

RESULTS

Immediate; it was found that the ***control*** group recorded statistically significantly the highest shear bond strength mean values followed by ***bleached + Natural AO*** treated group mean values then ***bleached + chemical AO*** treated group while the lowest statistically significant shear bond strength mean values were recorded with the ***Bleached*** group with no antioxidants as indicated by one-way ANOVA test($F=12.05, p<0.05$). Tukey's post-hoc pair-wise test showed non-significant ($p>0.05$) differences between (***bleached, bleached + chemical AO*** and ***bleached + natural AO*** groups). table (2) and figure (2)

Delayed; it was found that ***bleached + Natural AO*** treated group recorded statistically significantly the highest shear bond strength mean values followed by ***bleached + chemical AO*** treated group mean values then ***control*** group while the lowest statistically significant shear bond strength mean values were recorded with the ***Bleached*** group with no antioxidants as indicated by one-way ANOVA test(F=16.9, p<0.05). Tukey's post-hoc pair-wise test showed non-significant (p>0.05) differences between (***control*** and ***bleached, bleached + chemical AO*** and ***bleached + natural AO*** groups). table (2) and figure (2)

***Immediate* vs *Delayed*;**

In ***control*** *group;*it was found that the ***immediate*** subgroup recorded statistically significant higher mean value than the ***delayed*** subgroup as indicated by paired t-test **(t=3.8,** p<0.05).

In ***bleached*** *group;* it was found that the ***delayed*** subgroup recorded statistically non-significant higher mean value than the ***immediate*** subgroup as indicated by paired t-test **(t=0.78,** p>0.05).

In ***bleached + chemical AO*** *group;* it was found that the ***delayed*** subgroup recorded statistically non-significant higher mean value than the ***immediate*** subgroup as indicated by paired t-test **(t=1.6,** p>0.05).

In ***bleached + natural AO*** *group;* it was found that the ***delayed*** subgroup recorded statistically non-significant higher mean value than the ***immediate*** subgroup as indicated by paired t-test **(t=1.54,** p>0.05).

Total effect of main groups; totally regardless of storage time it was found that ***bleached + Natural AO*** treated group recorded statistically significantly the highest shear bond strength mean values followed by ***bleached + chemical AO*** treated group mean values then ***control*** group while the lowest statistically significant shear bond strength mean values were recorded with the ***Bleached*** group with no antioxidants as indicated by two-way ANOVA test (F=4.8, p=0.0134<0.05). Tukey's post-hoc pair-wise test showed no-significant (p>0.05) differences between (***control*** and ***bleached + natural AO***, ***bleached*** and ***bleached + chemical AO*** groups). table (2) and figure (2)

Total effect of storage time; irrespective of main groups, it was found that ***immediate*** subgroups recorded statistically non-significant higher mean value than ***delayed*** subgroups as indicated by two-way ANOVA test (F=1.6, p=0.2212>0.05).

TABLE (2) Shear bond strength results (Mean±SD) for all groups with different anti-oxidant and storage time

Variables		*Immediate*		*Delayed*		*t-test*
		Mean	*SD*	*Mean*	*SD*	*P value*
Control		15.92^{A}	5.34	7.87^{B}	1.51	0.0024*
Bleached		6.64^{C}	1.28	7.05^{B}	0.57	0.4491 ns
Bleached + Chemical AO		9.97^{BC}	1.67	11.26^{A}	1.36	0.1402 ns
Bleached + Natural AO		11.40^{AB}	1.19	13.35^{A}	3.14	0.2246 ns
ANOVA	***P value***	<0.0001*		<0.0001*		

AO; anti-oxidant Different letters in same column indicating statistically significant difference (p < 0.05). ns; non-significant (P>0.05) *; significant (P<0.05)

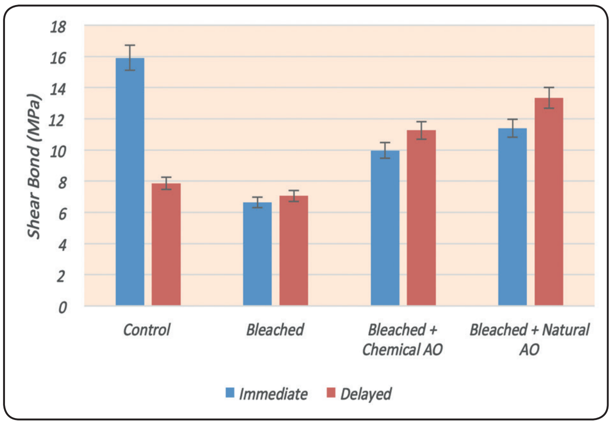

Fig. (2) Column chart of shear bond strength mean values for all groups with different anti-oxidants and storage time.

DISCUSSION

Esthetic restorations that are predominantly used in dentistry require the use of bonding procedures. It has recently been a subject of concern whether immediate bonding of those esthetic restorations, after bleaching procedures, decreased their bond strength to tooth structure.(1,4-9) That, partly, was attributed to the delayed release of oxygen which could hinder the resin in filtration into etched enamel, and, partly, was reported to be due to a possible inhibition of the polymerization reaction of resins.(1,4,5) It was reported that enamel acted as reservoir for residual oxygen, thus making its release difficult immediately after the bleaching procedure. It was further stated that it led to morphologic changes in the form of increase in porosity due to mineral loss, and loss of prismatic pattern.(26-28) Experimentally, removal of the oxygen-rich layer of enamel resulted in recovery of the bond strength of resin composite to the enamel to normal.(1) It was reported that the exact depth of the oxygen-rich enamel layer could not be detected. On the other hand, it was suggested that this layer must be deeper than 5 -10μm, other wise acid etching would have removed it.(29) In this perspective, it was suggested that during the bleaching procedure using hydrogen peroxide, the hydroxyl radicals in the apatite lattice were displaced by peroxide ions forming a peroxide apatite. It was further reported that, in two weeks time, the peroxide ions decomposed and the displaced hydroxyl radicals re-entered the apatite lattice causing a return of enamel to normal micro structurally.(2,11,24) Also it was found that the pulpal micro circulation removed the residual oxygen products.(4) According to those findings a delay in bonding or additional adhesive esthetic procedures after the bleaching procedure was recommended.(1,4-9) Studies suggested that even the dentin and the dentinal fluid could act as peroxide and oxygen free radical reservoirs until removed by pulpal micro circulation or, later, via surface diffusion.(4) The use of antioxidants produced an effect similar to that of a delay in the bonding procedure following the bleaching process. The reduced bond strength of esthetic restorations to enamel was reversed.(1-9,13-15) In this study we used two types of antioxidants, a natural one (pome granate extract) and a chemical synthetic one (sodium ascorbate preparation). The null hypothesis was that the natural antioxidant could be more powerful than the chemically prepared one. Sodium ascorbate is derived from ascorbic acid but with a neutral pH. That is why it was used instead of ascorbic acid to avoid the acidity. Sodium ascorbate was described as a potent antioxidant that produced surface free radicals in biologic medium. Added to that, since vitamin C and its salts were found to be non-toxic, this led to the wide use of sodium ascorbate as an antioxidant.(4)

Pome granate was the natural antioxidant chosen to be investigated in this study. This was because the effect of pome granate on bleached enamel has been very rarely investigated so far. Pome granate extract contains compounds that have a powerful antioxidant property, the most outstanding of which are phenolic compounds. Also, the polyphenolic bioflavonoids demonstrate a free radical scavenging property as well as a powerful antioxidant activity; in addition to reported antiviral, antibacterial, anti inflammatory, anticarcinogenic and vasodilatory actions.[6] In this study, the delayed in investigation sub-groups of specimens were incubated in artificial saliva for one month. The aim of incubation in artificial saliva was to simulate the intraoral condition on one side and to monitor the role that biodegradation could produce along with the gradual recovery of bond strength, on the other side. Incubation of specimens in artificial saliva was rarely performed in previous studies testing the effect of antioxidants on bleached enamel. Moreover, in our knowledge, incubation for a period of one month has not been investigated so far. The results of this study were as follow : the control sub-group (non-bleached) immediately investigated, demonstrated statistically significantly the highest shear bond strength mean values of all sub-groups. This is attributed to the fact that no bleaching was performed to this sub-groups of specimens and thus nothing influenced the shear bond strength of resin composite to the unaltered enamel. Within the control group (non-bleached), the immediate sub-group (mentioned above) consequently, demonstrated statistically significantly higher mean value than the delayed sub-group. In our opinion, this was attributed to the fact that the incubation of the delayed sub-group in artificial saliva for one month, caused gradual biodegradation which, in turn, affected the bond strength of resin composite to enamel. Regarding the bleached groups, it was found that both antioxidants (chemical and natural) were capable of causing recovery of the reduced shear bond strength following the bleaching procedure. However, it was found that, regardless of the storage time, the bleached groups treated with the natural antioxidant recorded statistically significantly higher shear bond strength mean values followed by the bleached sub-groups treated with the chemical antioxidant. This, as we think, is attributed to the powerful antioxidant phenolic compounds found in pome granate (the natural antioxidant). Within each of the bleached groups that were treated with antioxidants (natural or chemical), the delayed in investigation sub-groups demonstrated statistically non-significant higher shear bond strength mean values than the immediately investigated sub-groups. In our opinion, the presence of antioxidants could counter act the biodegradation process that could,possibly, occur as a result of incubation in artificial saliva. This issue may need further investigation. The bleached groups with no antioxidants use statistically significantly demonstrated the lowest shear bond strength mean values of all groups. Within those bleached groups (with no antioxidant use), the delayed sub-groups demonstrated statistically non-significant increase in shear bond strength mean values than the immediate sub-groups. It was reported that one of the methods used to remove the residual oxygen radical from the bleach enamel, is delaying restoration with resin composite. This was previously reported to be due to the fact that the residual peroxide ions as a result of bleaching, decomposed with time and that the expelled hydroxyl radicals re-entered the apatite crystal causing enamel to, micro structurally, return to normal [2,11,24]. According to the results of this study, the delayed sub-groups, demonstrated a non-significant increase in shear bond strength values compared to the immediate ones. That is to say, a sort of recovery in the reduced shear bond strength of resin composite to bleached enamel has occurred. However, a significant recovery did not occur. This, could be attributed to the bio degrading effect of artificial saliva (the incubating medium)

CONCLUSION

- Treatment of bleached enamel with antioxidants prior to resin compsite restoration, could reverse the reduced shear bond strength as a result of the bleaching process.
- The natural antioxidant demonstrated statistically significantly higher mean shear bond strength values than the synthetic chemical antioxidant.
- Incubation in artificial saliva could hinder the complete recovery of the reduced shear bond strength of resin composite to bleached enamel.

- Antioxidants (natural and chemical) could resist the biodegradation process as a result of incubation of the restored specimens in artificial saliva.

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