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By Ali Alabaş, Fulya Toksoy Topçu & Elif Aybala Oktay

University of Health Sciences Gulyhane

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Ali Alabaş ^a, Fulya Toksoy Topçu ^a & Elif Aybala Oktay^b

Abstract- The aim of this in-vitro study was to evaluate the application of different concentrations of sodium ascorbate (SA) on the strength of bonding to enamel after bleaching with hydrogen peroxide (HP). Forty human anterior teeth were randomly divided into four groups. Group 1 was treated with no bleaching application (negative control), and the treatments applied in the other groups were: Group 2; 35% HP for 30 min + 40% SA for 10 min, group 3; 35% HP for 30 min + 20% SA for 10 min, and group 4; 35% HP for 30 min + 10% SA for 10 min. Specimens were restored with adhesive All-Bond Universal (Bisco, Schaumburg, IL, USA), and a Beautifil-2 (Shofu Inc., Kyoto, Japan) giomer-resin material build-up was created. The specimens were subjected to shear bond testing by a universal machine. The data were analyzed with the Kruskal Wallis test followed by the Mann Whitney test, and $p < 0.05$ was deemed to indicate statistical significance. The unbleached teeth showed the highest shear bond strength, followed by the bleached teeth treated with 40% SA. Within the limitations of this study, it can be concluded that SA increased the potential bond strength associated with bleached enamel. Notably however, none of the different concentrations of SA tested restored the bond strength values to those of the unbleached teeth.

Keywords: sodium ascorbate, shear strength, dental resin, bleaching.

I. INTRODUCTION

The bleaching of vital teeth is a method used for eliminating the discolorations found on the surface and in the inner structure of teeth. It is a reliable and accepted treatment, and its popularity is on the increase. However, clinicians should be aware of the potential interactions between bleaching treatments and procedures involving adhesives, performed to treat aesthetic irregularities.

Bleaching agents of various concentrations are used to obtain rapid and aesthetic results. Hydrogen peroxide (HP) and carbamide peroxide have been successfully used for many years, to attain the desired tooth color. Postoperative susceptibility, pulpal irritation, changes in tooth structure, and microleakage due to existing restorations may be considered among the contraindications for bleaching treatments. Another

important complication that occurs after bleaching is a reduction in the strength of the bond between composite resin and the enamel.¹

Were a composite resin restoration is planned for a patient after the bleaching process, treatments such as laminate veneers and adhesion of an orthodontic bracket can be adversely affected by the prior bleaching, with regard to bonding to the enamel.² Several studies have shown that the bonding values of composite adhesive restorations performed on the surfaces of teeth after bleaching are considerably lower when compared with the surfaces of teeth to which no bleaching has been applied. The reason for this is the presence of a residual oxygen layer that occurs as a result of the bleaching process, and it has been reported that removing this residual oxygen layer increases the strength of bonding between the tooth and the composite material.³ The general approach for avoiding the compromised bond strength that occurs after bleaching is to observe a waiting period, which can be as little as 24 hours or can extend to up to 3 weeks.³

Recently conducted research suggests that the reduction in the strength of the bond between the adhesive and the tooth surface after bleaching can be ameliorated via the application of the antioxidant agent sodium ascorbate (SA). Antioxidant agents neutralize oxygenic free radicals and eliminate their negative biological effects.⁴

The objective of the current study was to evaluate bond strength between the tooth enamel and giomer-resin in teeth that had first been subjected to HP treatment, then treatment with one of three different concentrations of SA, then had the restorations performed.

II. METHODOLOGY

The protocol of this study was reviewed and approved by the Gulhane Military Medical Academy Research Ethics Committee (Registration number 08/236), and informed consent was obtained from the tissue donors. Forty undecayed human anterior teeth extracted for orthodontic and periodontal reasons were used in our study. The teeth were randomly divided into 4 groups of 10 teeth per group. In group 1 (negative control), giomer-resin blocks were built over the surface

Author a: DDS, Specialist, University of Health Sciences Gulhane Faculty of Dentistry, Department of Restorative Dentistry Ankara, Turkey. e-mail: alialabas@gata.edu.tr

Author a p: Assoc. Prof, Department of Restorative Dentistry Kecioren, Etilk, Ankara 06018 Turkey. e-mails: f.toksoytopcu@gmail.com, eaybala@yahoo.com



of the enamel without applying any bleaching agent or an antioxidant solution to the teeth. In group 2, giomer-resin blocks were built over the surface of the enamel after the application of a 35% bleaching agent containing HP for 30 min and a 40% solution containing the antioxidant agent SA for 10 min. In group 3, giomer-resin blocks were built over the surface of the enamel after the application of a 35% bleaching agent containing HP for 30 min and a 20% solution containing the antioxidant agent SA for 10 min. In group 4, giomer-resin blocks were built over the surface of the enamel after the application of a 35% bleaching agent containing HP for 30 min and a 10% solution containing the antioxidant agent SA for 10 min.

Immediately after being extracted, all teeth used in the study were immersed in a 10% formaldehyde solution, where they were stored until the time of study. Particular attention was paid to ensure that no decay, cracks, or fractures were present on the labial enamel surfaces of the teeth. The teeth were cleaned of debris with the aid of a periodontal curette, and were then immersed in de-ionized distilled water.⁵ Under water cooling, with the aid of a low-speed engine (Isomet Buehler Ltd, Chicago, IL, USA) all the dental crowns were cut and detached from the root parts by means of double-sided diamond discs, such that they remained 2 mm below the cementoenamel junction (CEJ). The root canal outlets that had been opened on the cut-off section were closed with a composite resin-based filling material. The crown pieces that were cut were kept in de-ionized distilled water at all non-experimental stages of the trial, to negate any dehydration. The teeth were embedded in cold acryl put into PVC-blocks of 3.0 cm in height and 2.5 cm in diameter, such that the labial enamel surface of each crown fragment was left exposed.⁶ The labial surfaces of the teeth were rubbed with 600-grid silicon carbide emery (abrasive) paper under water, and smooth, flat enamel surfaces 3 mm in diameter were obtained. The samples were then put into de-ionized distilled water.

The bleaching gel used incorporates a syringe system. Groups 2–4 were subjected to a bleaching agent containing 35% HP for 30 min (Yotuel Special, Biocosmetics Laboratories, Madrid, Spain). At the end of the bleaching treatment, the gel on the samples was removed with the aid of a surgical aspirator; then, the samples were washed in de-ionized distilled water. The different antioxidant treatments were then performed.

For group 2, 60 mL of de-ionized distilled water was put into a 100-mL glass tube, and SA was added until the volume reached 100 mL; hence, a 40% SA solution was obtained. For group 3, 80 mL of de-ionized distilled water was put into a 100-mL glass tube, and SA was added until the volume reached 100 mL; hence, a 20% SA solution was obtained. For group 4, 90 mL of de-ionized distilled water was put into a 100-mL glass tube, and SA was added until the volume reached 100

mL; hence, a 10% SA solution was obtained.⁷ Then, 1 mL of the relevant solution was applied to each of the samples every minute for a period of 10 min, thus, renewal of the solution and constant wetness of the enamel surface were ensured.^{8,9} At the end of this 10-min period, the enamel surfaces were washed in distilled water for 30 sec, and immediately thereafter, the construction of giomer-resin blocks over tooth enamels was performed.

At the resin block construction phase, the tooth enamels were first acidified with 37% orthophosphoric acid gel (Super Etch, SDI Ltd., Victoria, Australia) for 15 sec. In order to remove the acid gel after the acidification process, the tooth enamels were washed in water for 15 sec, and the remaining water on the surface was eliminated via a moist cotton pellet. A thin-layered adhesive (All Bond Universal, Bisco Inc., Schaumburg, IL, USA) was then applied to the acidified enamel surface with a micro-brush for 10 sec, after which it was subjected to air-drying for 10 sec, irradiated for 10 sec, and lastly it was polymerized. Following this, cylindrical plastic tubes with an inner diameter of 2.5 ± 0.02 mm and a height of 3.0 ± 0.02 mm were placed onto the tooth enamel surfaces.^{2,4} The plastic tubes were then filled with giomer-resin material (Beautifil 2, Shofu Inc., Kyoto, Japan), and polymerization was performed for 20 sec by means of a composite pistol emitting visible blue light with a wavelength of 430–490 nm (Bluephase C5, Ivoclar Vivadent, Liechtenstein, Switzerland). The plastic tubes were cut and removed with a bistoury (scalpel) end; then, resin blocks were obtained. The samples were kept in de-ionized distilled water for a period of 1 day prior to bond strength testing.

Bond strength testing was performed with an Instron 3363 (Norwood, MA, USA) device. The acrylic blocks were positioned such that the specialized end of the Instron device that takes the form of a knife-edge rested in the tooth enamel surface-junction area of the giomer-resin blocks. The samples were subjected to shear force such that the "crosshead" speed/rate was 1 mm/min.⁷ The data were recorded in megapascals (mpa).

a) Statistical Analysis

Statistical analyses of the data were performed via the SPSS 22.0 for Windows software package (SPSS Inc., Chicago, USA). As the number of samples in each group was small, the median, minimum, and maximum values were used in descriptive statistical analyses. Kruskal-Wallis analysis, which is the non-parametric equivalent of one-way analysis of variance, was used to compare the groups. As a post-hoc multiple comparison method, the Mann-Whitney U test with Bonferroni correction was performed, to reduce the error level. In the statistical analyses, $p < 0.05$ was deemed to indicate statistical significance.

III. RESULTS

The descriptive statistics derived from each group are shown in Table 1. There was a significant difference in the ultimate strengths between the groups, in terms of mpa values, as identified via the Kruskal-Wallis test.

Table 1: The descriptive statistics derived from each group via the Kruskal-Wallis test

	Median	Minimum	Maximum	<i>p</i>
Group 1 – Control	28.20	25.28	29.90	< 0.001
Group 2 – 40%	21.19	18.96	25.13	
Group 3 – 20%	13.66	12.81	17.18	
Group 4 – 10%	12.06	10.83	13.59	

Comparisons of the mean scores in each group suggest that the most successful experimental (non-control) group was group 2 (40% SA). When group 2, group 3 (20% SA), and group 4 (10% SA) were compared with one another, a statistically significant difference was observed. The values that were closest to those of group 1 (the control group) were obtained in group 2, and group 4 exhibited the lowest ultimate strength.

IV. DISCUSSION

In today's society, people are aware of the importance of teeth in human aesthetics. Discolored teeth that distort aesthetic appearance can negatively affect the psychological wellbeing and social lives of individuals. For these reasons, interest in tooth bleaching treatments is rapidly increasing.

The bleaching process on its own is not sufficient for achieving the necessary aesthetics in patients with diastema or distortion in the shape and form of their teeth. In such cases, composite restorations along with bleaching and aesthetic adhesive procedures such as laminate veneers are applied as a combined treatment. For better color harmony, adhesive practices such as diastema closure, or adjusting the form of canines or arranging the positions of lateral incisors can be performed following the bleaching process. For the long-term clinical success of adhesive restorations, sufficient adhesion must be ensured in the tooth structure. Any factor likely to adversely affect adhesion will also affect the aesthetic appearance and durability/stability of adhesive restorations.¹⁰

In the literature, it is stated that bleaching with peroxide reduces the adhesive strength of the composite over the tooth enamel.^{11,12} Those reports recommend a waiting period of 1–3 weeks before an adhesive procedure after bleaching. Such waiting

periods are not appropriate in cases in which early aesthetic arrangements are required.

It has been suggested that the main reason for the decline in adhesive bond strength after bleaching is that residual oxygenic free radicals remain in the tooth tissue in the wake of bleaching, and these prevent resin polymerization.^{13,14} Those studies utilized electron and optical microscopes and showed that the resin tags on bleached enamel surfaces are scattered, weak-looking, and structurally insufficient. It has also been suggested that the adhesive agent applied on the bleached enamel surface is not fully polymerized, such that the mechanical retention weakens and the strength of the adhesive diminishes.^{15,16} In those studies, papilla bubble-like structures and gaps were observed within the adhesive layer, and the authors surmised that the reason for this was that the oxygen released from HP remains within the adhesive layer in the course of the irradiation of the bonding agent.

In previous studies, various antioxidant agents have been used to eliminate the oxygen radicals that remain in the hard tissues of teeth after bleaching. These include SA,^{1,3,17,19} catalase,¹⁷ glutathione peroxidase,¹⁷ acetone,¹⁷ ethanol,¹⁷ sodium bicarbonate,¹⁷ grape seed extract,³ pine bark extract,³ proanthocyanidin,¹ lycopene,¹ malvidin chloride,¹⁹ pelargonidin chloride,¹⁹ and α -tocopherol.¹⁹ SA is the sodium salt form of ascorbic acid, which is known to be an effective antioxidant, and it has the ability to reduce various oxidative compounds.⁷ A previous study suggested the potentially protective effect of SA against damage caused to biological tissues by HP.²⁰ While ascorbic acid also exhibits a high antioxidant effect, it is not appropriate for use in clinical procedures due to its very low pH of 1.8. In contrast, the pH of SA, the antioxidant activity of which is the same as that of ascorbic acid, is 7.4, which is more compatible with biological tissues.²¹ Thus, we deemed it suitable to study SA, which we consider to be a biocompatible antioxidant agent with the potential for use after bleaching.

An appropriate duration period for the application of SA to bleached enamel surfaces has been determined to be 10 min in several studies.^{1,3,22} Where restorative tooth therapy is performed on a patient after a procedure with an antioxidant agent, the amount of time that patient will spend in the clinic will be extended accordingly. It is thought that a short span of time with regard to the application of the antioxidant agent will be more convenient in terms of clinical use.

There are several reports of increases in adhesive bonding values when SA was used as an antioxidant agent.^{1,19} On the other hand, in the studies in which different methods were applied, increases in adhesive bonding values have not been consistent. There are also studies showing that SA is insufficient for increasing adhesive bond strength.¹⁷

In the current study, the group that yielded a median value closest to the 28 mpa median of the control group was group 2 (40% SA), with a median of 21 mpa. In the results obtained at the end of our study, SA proved insufficient for increasing bond strength. In other studies, SA has been applied at different concentrations and for different periods of time, in efforts to ameliorate the reduction in bond strength of restorative materials observed after tooth bleaching.^{4,17,22}

Subramonian et al.³ assessed bonding values following office bleaching with 37% HP in two groups, one in which a 3-week waiting period was imposed, and the other in which 10% SA was applied for 10 min directly after bleaching. They reported that the bonding values were similar in both groups, but neither group attained the bonding values observed in the negative control group. Dabas et al.² applied 10% and 20% SA for 30, 60, and 120 min, and did not observe any significant differences in bond strengths between the 10% and 20% concentrations. They reported that bonding increased as the application period was extended, and that the adverse effect of bleaching was fully reversed after application periods of 60 and 120 min. Conversely, Tabatabaei et al.⁴ reported that 10% SA did not yield any significant increase in bonding strength after application periods of 5 or 10 min; however, they ascertained that compromised bond strength could be completely negated via a 1-week waiting period.

Miranda et al.²³ found that the application of 10% SA for 60 min after bleaching yielded the same results as a 1-week waiting period; which yielded almost the same results as those observed in the negative control (no bleaching) group. Notably however, a 2-week waiting period yielded better results than were observed in either of the aforementioned groups.

In a study in which 35% HP was used, Freire et al.²⁴ applied 35% SA for different periods of time and compared the resulting bond strength values, and measured the residual oxygen radical rates after waiting periods of 24, 48, 72, 96, 120, and 144 hours in the control group. They reported that residual oxygen radicals had decreased to a large extent after the 72-hour waiting period. The values obtained in the groups in which 35% SA was applied for 10 and 60 min suggested bond strengths approximating those of the control group. In the current study, the median value of 21 mpa observed in the 40% SA for 10 min group was low in comparison with the 28 mpa median in the control group.

Torres et al.¹⁷ reported that the mean bonding strength value in their control group after bleaching with 35% HP was 14.02 mpa, whereas it was 4.98 mpa after applying 10% SA for 20 min. Braz et al.²⁵ reported median bond strength values of 23.43 mpa in their control group after bleaching with 35% HP and 17.73 mpa after the application of 10% SA for 10 min.

Subramonian et al.³ reported that the mean bond strength in their control group after bleaching with 37% HP was 13.9 mpa, while it was 8.474 mpa after the application of 10% SA for 10 min. The median value of 28.20 mpa in the control group in our study, and the median value observed in group 4, in which 10% SA was applied for 10 minutes, seem to support the value of 12.06 mpa.

V. CONCLUSION

In the current study, even when a high SA concentration (40%) was applied for 10 min, the resulting bond strength values failed to reach those of the negative control group in which no bleaching was applied. Given that the patient is required to sit in the dental chair for almost 30 min during the bleaching process, we suggest that it will not be practical to apply an antioxidant agent for more than 10 min to enhance bond strength values. As performing a resin-filling process immediately after bleaching reduces the bond strength of adhesive restorations, dentists who aim to attain the best bond strength should take this situation into consideration.

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FIGURE LEGEND

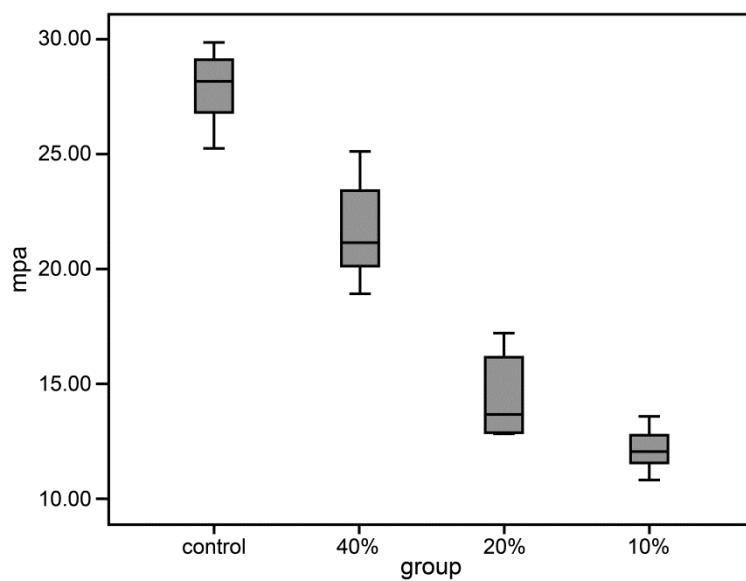


Figure 1: Box-plot comparison of the results of the control group and the experimental groups



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