Demineralization Effects of Hydrogen Heroxide on Bovine Enamel and Relation to Shear Bond Strength of Brackets

Santiago González-López^a/Carmen Lucia Soares G. de Medeiros^b/Camejo-Aguilar Defren^c/ Maria Victoria Bolaños-Carmona^d/Purificación Sanchez-Sanchez^e/Mario Menendez-Nuñez^f

Purpose: To measure the demineralization capacity of 37% phosphoric acid and shear bond strength (SBS) of brackets attached on bovine enamel at different times after bleaching with 30% hydrogen peroxide.

Materials and Methods: Four equally-sized pieces of each crown from 18 bovine incisors were randomly distributed among 7 groups (n = 10). After bleaching with 30% hydrogen peroxide for 1 h, specimens were stored in artificial saliva for 0 h, 24 h, or 1, 2, 3, or 4 weeks before bonding specimens to brackets. An unbleached group of specimens served as controls. Shear bond strength (SBS in MPa) was measured with a universal testing machine. Adhesive Remnant Index (ARI) scores were determined after failure of bracket bonds. To measure demineralized Ca²⁺, four 4 x 4 mm sections from each of 15 bovine incisors were randomly distributed among 4 groups (n = 15). Specimens were stored in artificial saliva for 0 h, 24 h or 7 days after bleaching and then immersed in 37% phosphoric acid solution. After 15 s, 30 s, and 60 s, 5-ml aliquots of solution were removed for spectrophotometry. Unbleached specimens served as controls.

Results: Larger amounts of Ca^{2+} were extracted from the enamel by phosphoric acid up to 24 h after application of hydrogen peroxide, when there was also a significant decrease in bracket-enamel SBS. After 1 week, there were no changes in amounts of Ca^{2+} extracted, and SBS values returned towards unbleached values.

Conclusion: Lower bracket-enamel SBS values at 24 h after bleaching are closely correlated with the larger amounts of Ca_2^+ extracted from the enamel.

Keywords: 30% hydrogen peroxide, shear bond strength, brackets, decalcifying, phosphoric acid, spectrophotometry.

J Adhes Dent 2009; 11: 461-467. doi: 10.3290/j.jad.a17651 Submitted for publication: 28.02.08; accepted for publication: 20.07.08.

Tooth bleaching has been considered a safe procedure by some authors, who observed no evidence of deleterious effects on enamel or dentin after the longest applications of the highest concentrations of hydrogen peroxide in normal clinical use for this purpose.^{12,22,23} Nevertheless,

- ^a Associate Professor, Department of Pathology and Dental Therapeutics, University of Granada, Spain.
- ^b Associate Professor, Department of Restorative Dentistry, University of Paraiba, Paraiba, Brazil.
- ^c Assistant Professor, Department of Operative Dentistry, University of Los Andes, Mérida, Venezuela.
- $^{\rm d}$ Associate Professor, Department of Integrated Pediatric Dentistry, University of Granada, Spain.
- Associate Professor, Department of Inorganic Chemistry, University of Granada, Spain.
- ^f Associate Professor, Department of Orthodontics, University of Granada, Spain.

Correspondence: Dr. Santiago González López, Department of Pathology and Dental Therapeutics, Faculty of Dentistry, University of Granada, Campus de Cartuja, E, 18071, Granada, Spain. Fax: +34-958-240-908. e-mail: sglopez@ugr.es

bleaching agents may adversely affect the bond strength of composite to acid-etched enamel. Various studies have reported significant reductions in enamel bond strength of resin composite restorations²⁴ and resin-bonded brackets when bonding is preceded by tooth bleaching.^{10,20,42} These reductions are greater at higher concentrations of bleaching agent.⁴³

Several investigations have attempted to elucidate this decrease in enamel bond strength after bleaching. Due to its low molecular weight, hydrogen peroxide can penetrate enamel and be retained in the bleached enamel.¹ Residual oxygen released from the bleaching agent inhibits polymerization of the resin, resulting in an incomplete curing of the adhesive, and interferes with resin infiltration into the etched enamel.¹⁵ It may therefore be responsible for the time-dependent reduction in the quality of resinenamel bonds.⁴¹

There is no general agreement concerning the deleterious effects of bleaching agents on enamel and dentin. Changes to the microstructure of surface enamel have been reported that are proportionally more severe with longer treatment times and higher hydrogen peroxide concentrations.⁷ In addition, bleaching agents may alter enamel microhardness, with loss of mineral content from the outer tooth structure.^{2,3,47} The release of calcium and phosphorous ions increases with higher hydrogen peroxide concentrations, leading to a reduction in the Ca:P ratio in bleached samples.² After prolonged use, the bleaching agent reduces the fracture resistance of dentin,³⁹ and these reductions are greater for longer application times and higher bleach concentrations.⁴⁰

Carbamide peroxide has an intrinsic demineralizing effect.^{17,29} Previous bleaching with 30% carbamide peroxide increases the amount of Ca²⁺ extracted from enamel after etching with phosphoric acid, with highest losses observed at 24 h after bleaching.²⁸ Many clinical procedures, eg, bonding of orthodontic brackets, porcelain veneers, composite veneers, or composite restorations, require acid etching of the enamel only a short time after completion of bleaching. The effectiveness of these procedures may be affected by this higher demineralizing power of phosphoric acid on bleached enamel.

In the present study, the null hypothesis was that bleaching treatment with 30% hydrogen peroxide does not modify either the decalcifying ability of phosphoric acid in enamel acid etching or the shear bond strength of brackets to enamel. The objectives were: 1. to determine the shear bond strength of metal orthodontic brackets bonded to hydrogen peroxide-bleached enamel after various postbleaching intervals (immediately, or at 24 h, 1 week, 2 weeks, 3 weeks, and 4 weeks postbleaching); 2. to evaluate by Atomic Absorption Spectrometry the decalcifying effects of 37% phosphoric acid on enamel after clinically relevant application times (15 s, 30 s, and 60 s) following three postbleaching intervals (no interval, 24 h, and 1 week); and 3. to explore the relationship between the decalcifying effects of 37% phosphoric acid and shear bond strength.

MATERIALS AND METHODS

Shear Bond Strength (SBS)

Eighteen bovine incisors with no fractures or defects on the vestibular enamel surface were selected and stored in thymol under refrigeration until use. All roots were cut off at the cementoenamel junction using an Accutom-50 diamond cutter (Accutom Hard Tissue Microtome, Struers; Ballerup, Denmark). Each crown was then cut (in the incisocervical then mesiodistal direction) into 4 equallysized sections. Using 20-ml disposable syringes (with conical ends removed) as molds, each specimen was then attached to the central part of the syringe plunger using Coltene utility wax (Whaledent; Mahwah, NJ, USA). The plunger was withdrawn to leave a 1-cm deep container that was then filled with transparent self-curing resin (Ortocryl, Dentaurum; Ispringen, Germany), prepared according to the manufacturer's instructions. During polymerization, the mold was submerged in water to compensate for the exothermic reaction. The resulting cylinder was expressed from the mold by using the syringe plunger. The exposed enamel surface of specimens was polished with 500- then 1200-grit silicon carbide paper disks in order to remove the surface aprismatic enamel layer and obtain a flat surface.

Specimens were randomly divided into seven treatment groups (n = 10). Group 1 was an unbleached control group. The other six groups were bleached with Illumine Office bleaching agent (Dentsply Detrey; Konstanz, Germany) according to the manufacturer's instructions. After mixing the two components, the resulting gel was applied to the polished surface of the enamel for a period of 60 min, during which the specimens were kept in an oven maintained at 37°C and low relative humidity. Although the manufacturer's information leaflet describes this widely used product as 30% hydrogen peroxide, the mixed gel has a concentration of 15%. After bleaching, specimens were washed with abundant distilled water and dried with absorbent paper. Groups 3 to 7 were then stored in artificial saliva²⁸ in an incubator at 37°C for the required time periods (24 h, 1, 2, 3, or 4 weeks). Group 2 was bonded immediately.

A 37% phosphoric acid gel (3M Dental Products; St Paul, MN,USA) was used to etch specimens for 30 s. They were then rinsed with abundant water and dried with an air syringe until the frosty white appearance of etched enamel was seen. MBT Victoria Series metal brackets for maxillary lateral incisors (3M Uniteck; Puchheim, Germany) were used, with a combined surface area of 12.2 mm². They were attached with Transbond XT bracket adhesive (3M Dental Products; St Paul, MN, USA) following the manufacturer's recommendations.

After surface preparation. Transbond XT liquid primer (3M Dental Products) was applied to the etched surfaces. Sufficient bonding paste was applied to cover the bracket bases. The brackets were then seated firmly on the enamel surface and excess resin was removed with a dental explorer. Light curing was done with an Astralis 10 halogen lamp (Ivoclar Vivadent; Schaan, Liechtenstein) at 700 mW/cm² for 20 s. After bonding, the specimens in all groups were stored in artificial saliva at 37°C for 24 h to allow complete resin polymerization. SBS was tested by a method that avoids the generation of leverage forces (Fig 1), using an Electrotest Model 500 universal traction machine (IBERTEST; Madrid, Spain) at a speed of 1 mm/min. The device allows a sharpened blade to be moved in only one direction on the smooth surface of the specimen, always cutting along the bracket/enamel interface in all specimens.

After debonding, all specimens and brackets were examined under 10X magnification and scored on the Adhesive Remnant Index (ARI).⁶ The ARI scale has a range of 1 to 5. A score of 5 indicated no composite remaining on the enamel; 4: < 10% of composite remaining; 3: 10% to 90% remaining; 2: > 90% remaining; 1: all of the composite remained on the tooth, bearing the impression of the bracket base.

Spectrometry

Fifteen bovine incisors were stored in distilled water with thymol crystals until use. All roots were cut off at the cementoenamel junction using an Accutom-50 diamond cutter (Accutom Hard Tissue Microtome, Struers; Ballerup,

462

González-López et al

Denmark). Crowns were polished with silicon carbide paper disks on a polisher (Exakt-Apparatebau; Norderstedt, Germany) to obtain a flat vestibular surface, and each crown was then cut into 4 equally-sized 4 x 4 mm sections (sample specimens) as described above. After polishing the inner surfaces to remove the dentin, sample specimens were weighed on a Sartorius BL60S precision balance (accuracy of \pm 0.0001 g; Sartorius; Mugió, Italy). Maximum weight equality among specimens was achieved by reducing weight when necessary with 600-grit silicon carbide paper disks (WS 18-B, Struers), always reducing the inner surface of the section to avoid modifying its geometry. An Olympus stereomicroscope (Olympus Optical España; Barcelona, Spain) was used to verify that all exposed surfaces were enamel.

One sample specimen from each crown was assigned to one of four groups (n = 15): group I, no bleaching agent (control group); groups II, III, and IV, bleached with 30% hydrogen peroxide for 60 min. Groups III and IV were then stored in artificial saliva for 24 h and 1 week, respectively.

For bleaching, specimens were immersed in 30% hydrogen peroxide (Illuminé Office, Dentsply Detrey) for 60 min, replacing all of the gel at 30 min as per manufacturer's instructions. Specimens were washed with abundant distilled water and dried with absorbent paper. Group III and IV specimens were then kept in artificial saliva at 37°C for the specified time period (24 h or 7 days). Group II specimens were immersed in 37% phosphoric acid solution immediately after bleaching. The 37% phosphoric acid solution was prepared by dissolving 311 ml of 85% phosphoric acid (Scharlan Cemise; Barcelona, Spain) with a concentration of 1.71 g/ml in 689 ml of distilled water to obtain 1000 ml of 37% acid solution with a pH of 0.14, measured with a Micro pH 2000 pH meter (Crisol; Alella, Spain).

A blank 20 ml sample of phosphoric acid solution was first prepared to determine Ca^{2+} levels in the absence of a specimen. All unbleached and bleached specimens underwent the same experimental procedure, ie, immersion in the phosphoric acid solution followed by constant agitation using a magnetic multi-stirrer (SBS A-09 series C, Scientific Basic Solutions; Barcelona, Spain) to uniformly mix the Ca^{2+} extracted into the solution. At 15 s, 30 s, and 60 s, 5-ml aliquots were removed, using a calibrated micropipette that was replaced after each extraction. Extracts were placed in hermetically sealed, labelled glass tubes. By these means, three extracts were obtained for each sample specimen.

Ca²⁺ concentrations in the solutions were determined by Atomic Absorption Spectrometry (Perkin Elmer, model 5100ZL; Waltham, MA, USA) using the flame technique. Extract readings were expressed in ppm and subsequently transformed into mg of Ca²⁺ lost per g of specimen by applying the following formula as described elsewhere:²⁸

mg Ca²⁺/g = [(ppm Ca₂⁺)·10⁻³ l/ml·V]/W

where ppm Ca^{2+} are ppm of Ca^{2+} in each time period, V is volume of solution in ml (at 15 s, V1; at 30 s, V2; and at 60 s, V3), and W is the weight of the sample in mg. For the



 $\ensuremath{\mbox{Fig}}\xspace1$ SBS-testing device that avoids the generation of leverage forces.

15-s period, the volume of solution (V1) was 20 ml, whereas at 30 s, the volume (V2) was 15 ml, since 5 ml of solution had previously been removed. Therefore, the mg of Ca²⁺ corresponding to 5 ml of V1 solution were added to the mg of Ca²⁺ obtained at 30 s. Likewise, since the volume was 10 ml at 60 s, the mg of Ca²⁺ corresponding to 5 ml of V1 solution plus 5 ml of V2 solution were added to those obtained at 60 s.

Statistical Analysis

The effect of storage interval on enamel-bracket SBS was analyzed using one-way ANOVA and the post-hoc Dunnet's t test, with the unbleached control group as the reference category. A three-way ANOVA was used to explore the relationships among bleaching treatment, acid exposure, and extracted calcium, followed by two-way ANOVA and the post-hoc Dunnet's t test. Frequencies in each ARI category were calculated. The distribution of ARI scores in groups was compared by using a chi-square test. The significance level was set at p < 0.05.

RESULTS

Shear Bond Strength

Table 1 shows the means and standard deviations of SBS measurements. The ANOVA revealed significant effects (F = 9.137; p <0.001) of the time elapsed before bracket bonding to enamel (bleaching to bonding interval). Dunnet's t-test showed significant differences in SBS values between the control group and groups with bleaching-to-bonding intervals of 24 h and 1 week. SBS values in the 24-h group were one-third of SBS values began to recover towards unbleached control values and had doubled after 1 week, although they remained significantly lower than controls. No significant differences in SBS values were ob-

 Table 1
 Mean shear bond strength (in MPa) for each bleaching-to-bonding interval

Shear bond strength (MPa)	Bleaching-to-bonding interval							
	Control	0 h	24 h	1 week	2 weeks	3 weeks	4 weeks	
Mean (SD)	37.20	28.96	13.38	26.60	32.74	32.52	32.76	
	(4.42)	(7.06)	(3.44) a	(8.99) b	(11.64)	(5.51)	(11.31)	

Table 2 Comparisons of adhesive remnant index (ARI) among different groups

Bleaching-to-bonding		n				
interval	1	2	3	4	5	
Unbleached	5	-	-	1	4	10
0 h	-	-	2	3	5	10
24 h	-	-	-	-	10	10
1 week	2	-	-	4	4	10
2 weeks	1		1	2	6	10
3 weeks		1	2	3	4	10
4 weeks			2	3	5	10
Chi-square = 41.194, p = 0	.16					

served between the unbleached control group and groups with a bleaching-to-bonding interval greater than 1 week. When brackets were attached immediately after bleaching, SBS values appeared slightly lower compared to the unbleached control group, but the difference was not statistically significant.

Analysis of failure type (Table 2) revealed that all failures were adhesive along the resin/enamel interface (ARI score of 5) after a bleaching-to-bonding interval of 24 h, with no resin remaining adhered to the enamel. Fractured specimens from the other postbleaching groups showed various types of failure (around 75% with ARI scores of 4 or 5). All of the composite remained bonded to the enamel surface in 50% of unbleached specimens (score of 1) but in none of the bleached groups. The distribution of fractured specimens across ARI categories significantly differed among the groups ($\chi^2 = 41.195$; p < 0.05).

Ca²⁺ Demineralization by Phosphoric Acid Etching

Table 3 shows the cumulative amounts of Ca²⁺ extracted from the enamel specimens after subtracting the amount of Ca²⁺ found in the study solution without a specimen. The three-way ANOVA (bleaching, acid exposure time, and sample) showed significant differences between experimental groups (F = 5.124, p < 0.002) and acid exposure (etching) times (F = 115.300, p < 0.001), with no significant interactions.

The amounts of Ca²⁺ extracted from bleached and unbleached specimens increased with longer etching time, and significantly differed between all etching time periods (60 s > 30 s > 15 s). Unbleached and bleached groups showed similar demineralization kinetics.

The total extracted Ca²⁺ was greater for groups with bleaching-to-etching intervals of 0 h and 24 h than for the control group, although this difference was only significant (p < 0.05) for the 24-h group (p < 0.10 for the 0-h group). Similar amounts of Ca²⁺ were extracted between the group etched at 7 days after bleaching and the unbleached control group. For a given exposure time, groups did not significantly differ in the amounts of Ca²⁺ extracted.

DISCUSSION

Contemporary bleaching agents are typically either hydrogen peroxide or carbamide peroxide. In-office bleaching generally uses relatively high levels of whitening agents for short time periods. For this study, the in-office bleaching agent Illuminè (Dentsply), usually applied for 60 min in the

Table 3 Amounts of Ca²⁺ in mg extracted (as determined by spectrometry) in relation to bleaching-to-etching intervals and acid exposure time

Bleaching-to-etching	Acid exposure time					
	15s	30s	60s	Total		
	(n=60)	(n=60)	(n=60)	(n=180)		
Group I	0.614	0.952	1.601	1.056		
(control)	(0.282)	(0.287)	(0.456)	(0.538)		
(n = 45)	a	b	c	A		
Group II	0.749	1.089	1.846	1.228		
(B- 0 h)	(0.388)	(0.351)	(0.740)	(0.726)		
(n = 45)	a	b	c	A		
Group III	0.731	1.151	1.880	1.257		
(B- 24 h)	(0.442)	(0.628)	(0.803)	(0.729)		
(n = 45)	a	b	c	B		
Groups IV	0.579	0.875	1.520	0.991		
(B- 7 days)	(0.167)	(0.238)	(0.353)	(0.473)		
(n = 45)	1	2	3	A		
Total B-E (n = 180)	0.668 (0.336) a	1.017 (0.454) b	1.714 (0.622) c			

B-E: Bleaching-to-etching interval. Different capital letters indicate statistically significant differences among experimental groups. In each row, different lower-case letters indicate statistically significant differences among etching times in each bleaching group.

clinical setting, was applied on the enamel of bovine teeth, widely recognized as a reliable substitute for human teeth in this type of investigation.³³ Although demineralization is faster in bovine than human enamel¹⁸ and undergoes greater erosion and erosion-abrasion,⁵ it has been described as a suitable substitute to evaluate enamel demineralization.³⁰ Published studies have supported the use of bovine teeth as a reliable substitute for human teeth in microleakage³² and adhesion tests on enamel and dentin,19,33-35 obtaining comparable bond strength measurements in bovine and human teeth. SEM observations showed that the morphology of bovine and human teeth and their enamel was similar after etching with 35% H₃PO₄.³³ Bovine enamel was used in the present study because a histologically similar group of teeth can be more readily obtained and four equal-sized enamel sections can be cut from a single bovine tooth, unlike in human teeth, enhancing comparison conditions. Nevertheless, due caution should be exercised in extrapolating results from bovine to human teeth.

Several factors might influence the bond strength values after tooth bleaching, including the bonding technique,³⁸ the bleaching agent and its concentration,⁴³ and the delay between bleaching and bonding.^{13,37,45} Enamelbracket SBS is known to depend on the point at which the force is applied.²⁵ In the present study, a test method that avoids the generation of leverage forces was used.

It is well documented that bleaching significantly reduces the immediate bond strength of composite to enamel,^{24,31,45} which has been related to alterations in the attachment surface area, resin/enamel interface, and resin quality.²⁶ Sundfeld et al³⁷ found inferior formation of tags, which were shorter, thinner, less frequent, nonuniform, and poorly defined, and a lesser penetration of the adhesive material into the enamel in comparison with unbleached controls. Dishman et al¹⁵ reported a high concentration of residual oxygen in the pores of bleached tooth enamel. This oxygen-rich layer is 5 to 10 µm thick, and may not only inhibit polymerization of the adhesive but also increase the porosity of the resin material by the release of gaseous oxygen. Resin/enamel interfaces on bleached enamel may exhibit more extensive nanoleakage as a result of this interaction.27

In this study, SBS values were lower in brackets attached immediately after bleaching than in the unbleached group, but this difference was not significant. However, Unlu et al⁴⁵ found significantly lower SBS values for specimens bonded immediately after bleaching than for unbleached specimens. This disparity may be explained by their use of a self-etching adhesive (Clearfil SE Bond), since the susceptibility of adhesives to bleaching may depend on the aggressiveness of the demineralization process.¹¹ The pH of Illuminé is 6.5, much higher than the 1.9 of Clearfil SE Bond adhesive. When the latter is used immediately after the former, the higher pH of the former could interfere with the effectiveness of the adhesive. When Uysal et al⁴⁵ used the same adhesive protocol as in the present study (etching with phosphoric and Tansbond XT), they found that in-office bleaching with 35% hydrogen peroxide did not adversely affect the bond strengths of brackets bonded immediately or at 30 days after bleaching. These results are in agreement with the present findings for these time periods. However, they did not study bonding of brackets to enamel at 24 h after bleaching, when we found the weakest SBS values (and largest Ca²⁺ losses during etching), which were around one-third lower than for unbleached controls. One week later, the SBS had increased to reach the value of the control group, and SBS values then remained the same for all groups up to 1 month. It should be borne in mind that the increase in SBS might have occurred earlier, since it was not evaluated between three and six days after bleaching.

There are concerns that tooth-whitening procedures irreversibly damage tooth structure. Bleaching has been reported to produce microstructural changes to the surface and subsurface layers of enamel,²³ mainly localized in the outer enamel and diminishing in inner enamel layers,¹⁶ associated with a loss of Ca²⁺ from the enamel and a significant decline in calcium:phosphate ratios.⁸ A greater loss of Ca²⁺ is produced by 37% phosphoric acid etching of enamel after bleaching,²⁴ resulting in loss of prismatic form and a more extensive etching pattern.^{18,22} Although the rougher surface and greater retention obtained might be expected to increase the bond strength, it is in fact reduced because the hydrogen peroxide alters the organic matrix of the enamel,^{17,21} favoring Ca²⁺ loss and weakening the etched enamel.

Josey et al,²³ using light microscopy, found that the bleaching process resulted in a loss of mineral from the enamel that was evident 24 h after bleaching, similar to the present finding. Although hydrogen peroxide has a high capacity for diffusion,⁴⁶ a time interval is required for the structural alteration of hydroxyapatite that produces Ca^{2+} losses, which are observed after 24 h. In a previous study, Medeiros et al²⁸ found no significant difference in Ca^{2+} loss between a control group and a group etched immediately after bleaching treatment. The pH of the 30% hydrogen peroxide gel used in their study was 6.5, therefore the demineralization observed in the present study cannot be attributed to the low pH of the gel.

We found no statistically significant differences in pair comparisons between groups, which may be explained by the low final concentration of 15% hydrogen peroxide in the bleaching gel used (Illuminé Office, Dentsply Detrey). Significant differences may also have emerged with a longer exposure time, as found in a previous study by our group that used an 11% final concentration of hydrogen peroxide for 90 min, 30 min more than in the present study.²⁸ Efeoglu¹⁷ found a significant reduction in the mineral content of surface and subsurface enamel specimens after application of high-concentration in-office bleaching for 2 h, the maximum application time recommended by the manufacturer. This demineralization was clearly greater in the outer 150 μ m of enamel. The quantity of ions released increased with higher hydrogen peroxide concentrations.² These greater Ca²⁺ losses might be explained by changes in the chemical composition of dental enamel caused by the bleaching agents.¹⁵

opyr

Although Josey et al²³ suggested that the loss of mineral from enamel was sustained after 12 weeks of storage in artificial saliva, it is also widely reported that microstructural changes in bleached enamel can be reversed by salivary components,^{4,7,14} with consequent restoration of the bond strength to enamel.^{10,43} In the present study, the artificial saliva functioned as a remineralization solution. For this reason, the present findings are similar to those of Unlu⁴⁵ and Machado,¹³ who found that SBS values and levels of extracted Ca²⁺ returned to levels found for unbleached enamel by 1 week after the use of hydrogen peroxide and then remained stable until the end of the study at 4 weeks.

Several methods have been proposed to meet the clinical challenge posed by the effects of bleaching on bond strength. Dishman¹⁵ suggested that the oxygen-rich layer would be removed by the action of the phosphoric acid applied before application of resin-based adhesive. In contrast, Cadenaro¹¹ proposed that etching and rinsing procedures do not eliminate residual oxygen from the surface and that polymerization of the adhesive is reduced. Composite-enamel bond strength was significantly improved by the pre-bonding application of antioxidant agents, eg, 10% sodium ascorbate, catalase or ethanol,²⁶ which were claimed to reduce or completely remove residual peroxides and other oxygen radicals. Our results demonstrate that the reduction in SBS is not solely a result of the persistence of oxygen in the enamel. Thus, lowest SBS values were at 24 h, when the largest extraction of Ca²⁺ by phosphoric acid etching was observed. Moreover, the fracture patterns observed indicated poor adhesion. Therefore, the bleached enamel surface did not allow a strong and stable bond to be formed with the composite, and our null hypothesis must be rejected.

We recommend a time interval of at least 7 days between the utilization of peroxide bleaching materials and restorative procedures that require acid-etching and adhesive bonding,⁹ allowing sufficient time for penetration of the adhesive system into the enamel surface, with formation of regular tags.¹³

CONCLUSIONS

At 24 h after application of 30% (final concentration 15%) peroxide, a larger amount of Ca^{2+} was extracted by 37% phosphoric acid from bleached than unbleached enamel, coinciding with a significant decrease in the shear bond strength of brackets to enamel. After one week, the shear bond strength with bleached enamel progressively recovered to the value observed with unbleached enamel, and extracted Ca^{2+} values stabilized.

González-López et al

REFERENCES

- Adibfar A, Steele A, Torneck CD, Titley KC, Ruse D. Leaching of hydrogen peroxide from bleached bovine enamel. J Endod 1992;18:488-491.
- Al Salehi SK, Wood DJ, Hatton PV. The effect of 24h non-stop hydrogen peroxide concentration on bovine enamel and dentine mineral content a microhardness. J Dent 2007;35:845-850.
- Attin T, Vollmer D, Wiegand A, Attin R, Betke H. Subsurfaces microhardness of enamel and dentin after different external bleaching procedures. Am J Dent 2005;18:8-12.
- Attin T, Kielbassa AM, Schwanenberg M, Hellwing E. Effect of fluoride treatment on remineralization of bleached enamel. J Oral Rehabil 1997;24: 282-286.
- Attin T, Wegehaupt F, Gries D, Wiegand A. The potential of deciduous and permanent bovine enamel as substitute for deciduous and permanent human enamel: Erosion-abrasion experiments. J Dent 2007;35:773-777.
- Bishara SE, VonWald L, Olsen ME, Laffoon JF. Effect of time on the shear bond strength of glass ionomer and composite orthodontic adhesives. Am J Orthod Dentofacal Orthop 1999;116:616-620.
- Bistey T, Nagy IP, Simó A, Hegedus C. In vitro FT-IR study of the effects of hydrogen peroxide on superficial tooth enamel. J Dent 2007;35:325-330.
- Bizhang M, Seemann R, Duve G, Römhild G, Altenburger MJ, Jahn KR, Zimmer S. Demineralization effects of 2 bleaching procedures on enamel surfaces with and without post-treatment fluoride application. Oper Dent 2006;31:705-709.
- Bulut H, Kaya AD, Türkün M. Tensile bond strength of brackets after antioxidant treatment on bleached teeth. Eur J Orthod 2005;27:466-471.
- Bulut H, Turkum M, Kaya AD. Effect of an antioxidizing agent on the shear bond strenth of brackets bonded to bleached human enamel. Am J Orthod Dentofacial Orthop 2006;129:266-272.
- Cadenaro M, Breschi L, Antoniolli F, Mazzoni A, Di Lenarda R. Influence of whitening on the degree of conversion of dental adhesives on dentin. Eur J Oral Sci 2006;114:257-262.
- Dadoun MP, Bartlett DW. Safety issues when using carbamide peroxide to bleach vital teeth – a review of the literature. Eur J Prosthodont Restor Dent 2003;11:9-13.
- Da Silva Machado J, Cândido MS, Sundfeld RH, De Alexandre RS, Cardoso JD, Sundefeld ML. The influence or time interval between bleaching and enamel bonding. J Esthet Restor Dent 2007;19:111-119.
- 14. De Freitas PM, Basting RT, Rodriguez AL, Serra MC. Effect of 10% carbamide peroxide bleaching agents on dentin microhardness at different time intervals. Quintessence Int 2002;33:370-375.
- 15. Dishman MV, Covey DA, Baughan LM. The effects of peroxide bleaching on composite to enamel bond strength. Dent Mater1994;10:33-36.
- Efeoglu N, Wood D, Efeoglu C. Microcomputerised tomography evaluation of 10% carbamide peroxide applied to enamel. J Dent 2005; 33:561-567.
- 17. Efeoglu N, Wood DJ, Efeoglu C. Thirty-five percent carbamide peroxide application causes in vitro demineralization of enamel. Dent Mater 2007;23:900-904.
- Featherstone JD, Mellberg JR. Relative rates of progress of artificial carious lesion in bovine, ovine and human enamel. Caries Res 1981;15:109-114.
- Fowier CS, Swartz ML, Moore BK, Rhodes BF. Influence of selected variables on adhesion testing. Dent Mater 1992;8:265-269.
- Garcia-Godoy F, Dodge WW, Donohue M. O'Quinn JA. Composite resin bond strength after enamel bleaching. Oper Dent 1993;18:144-147.
- Jiang T, Ma X, Wang Y, Tong H, Shen X, Hu Y, Hu J. Investigation of the effects of 30% hydrogen peroxide on human tooth enamel by Raman scattering and laser-induced fluorescente. J Biomed Opt 2008;13:014019.
- Joiner A. Review of the effects of peroxide on enamel and dentine properties. J Dent 2007;35:889-896.
- Josey AL, Meyers IA, Romaniuk K, Symons AL. The effect of vital bleaching techniques on enamel surface morphology and the bonding of composite resin to enamel. J Oral Rehabil 1996;23:244-250.
- 24. Kaya AD, Türkün M. Reversal of dentin bonding to bleached teeth. Oper Dent 2003;28:825-829.
- Klocke A, Nieke B. Influence of force location in orthodontic shear bond strength testing. Dent Mater 2005;21:391-396.

- 26. Kum KY, Lim KR, Lee CY, Park KH, Safavi KE, Fouad AF et al. Effects of removing residual peroxide and other oxygen radicals on the shear bond strength and failure modes at resin-tooth interface after tooth bleaching. Am J Dent 2004;17:267-270.
- Lai SCN, Tay FR, Cheung GSP, Mak YF, Carvalho RM, Wei SHY, Toledano M, Osorio R, Pashley DH. Reversal of compromised bonding in bleached enamel. J Dent Res 2002;81:477-481.
- Medeiros CL, González-López S, Bolaños-Carmona MV, Sanchez-Sanchez P. Effects of phosphoric acid on bovine enamel bleached with carbamide peroxide. Eur J Oral Sci 2008;116:66-71.
- McCracken MS, Haywood VB. Demineralization effects of 10 percent carbamide peroxide. J Dent 1996;24:395-398.
- Mellberg JR. Hard-tissue substrates for evaluation of cariogenic and anticariogenic activity in situ. J Dent Res 1992; 71:913-919.
- 31. Nour El-din AK, Miller BH, Griggs JA, Wakefield C. Immediate bonding to bleached enamel. Oper Dent 2006;31:106-114.
- Reeves GW, Fitchie JG, Hembree JH Jr, Puckett AD. Microleakage of new dentin bonding systems using human and bovine teeth. Oper Dent 1995; 20:230-235.
- Reis AF, Giannini M, Kavaguchi A, Soares CJ, Line SR. Comparison of microtensile bond strength to enamel and dentin of human, bovine, and porcine teeth. J Adhes Dent 2004;6:117-121.
- Saleh F, Taymour N. Validity of using bovine teeth as a substitute for human counterparts in adhesive tests. East Mediterr Health J 2003;9:201-207.
- Schilke R, Bauss O, Lisson JA, Schuckar M, Geurtsen W. Bovine dentin as substitute for human dentin in shear bond strength measurements. Am J Dent 1999;12:92-96.
- Sulieman M, Addy M, Macdonald E, Rees JS. A safety study in vitro for the effects of an in-office bleaching system on the integrity of enamel and dentine. J Dent 2004; 32:581-590.
- Sundfeld RH, Briso ALF, De Sá PM, Sundfeld MLMM, Bedran-Russo AKB. Effect of time interval between bleaching and bonding on tag formation. Bull Tokyo Dent Coll 2005;46:1-6.
- Sung EC, Chan SM, Mito R, Caputo AA. Effect of carbamide peroxide bleaching on the shear bond strength of composite to dental bonding agent enhanced enamel. J Prosthet Dent 1999;82:595-599.
- Tam LE, Kuo VY, Noroozi A. Effect of prolonged direct and indirect peroxide bleaching on fracture toughness of human dentin. J Esthet Restor Dent 2007;19:100-110.
- Tam LE, Noroozi A. Effects of direct and indirect bleach on dentin fracture toughnes. J Dent Res 2007;86:1193-1197.
- Torneck DC, Titley KC, Smith DC, Adibfar A. The influence of time of hydrogen peroxide exposure on the adhesion of composite resin to bleached bovine enamel. J Endod 1990;16:123-128.
- Türkkahraman H, Adanir N, Günör Y. Bleaching and desensitizer application effects on shear bond strengths of orthodontic brackets. Angle Orthod 2007;77:489-493.
- Türkün M, Kaya AD. Effect of 10% sodium ascorbate on the shear bond strength of composite resin to bleached bovine enamel. J Oral Rehabil 2004;31:1184-1191.
- Unlu N, Cobankara FK, Ozer F. Effect of elapsed time following bleaching on the shear bond strength of composite resin to enamel. J Biomed Mater Res B Appl Biomater 2008;84:363-368.
- Uysal T, Basciftci FA, Usumez S, Sari Z, Buyukerkmen A. Can previously bleached teeth be bonded safely? Am J Orthod Dentofacial Orthop 2003;123:628-632.
- Wattanapayungkul P, Yap AU. Effects of in-office bleaching products on surface finish of tooth-colored restorations. Oper Dent 2003; 28:15-19.
- Zantner C, Beheim-Schwarzbach N, Neumann K, Kielbassa AM. Surface microhardness of enamel after different home bleaching procedures. Dent Mater 2007; 23:243-250.

Clinical relevance: A minimum interval of 7 days is recommended between the use of peroxide bleaching material and the adhesive bonding of orthodontic brackets which requires acid etching. Copyright of Journal of Adhesive Dentistry is the property of Quintessence Publishing Company Inc. and its content may not be copied or emailed to multiple sites or posted to a listserv without the copyright holder's express written permission. However, users may print, download, or email articles for individual use.