Original Article

Influence of Bleaching Regimen and Time Elapsed on Microtensile Bond Strength of Resin Composite to Enamel

Abstract

Objectives: The aim of this study was to evaluate the effects of time elapsed since bleaching and different bleaching regimens on the microtensile bond strength of resin composite to enamel. **Methodology:** Forty flattened buccal enamel surfaces were divided into four groups: An unbleached (control) group and three bleaching groups. Control group specimens were not subjected to a bleaching regimen (Group 1), while those in the bleaching groups were bleached as follows: opalescence 10% (Group 2), whiteness perfect 16% (Group 3), and whiteness hydrogen peroxide 35% (Group 4). Thereafter, the bleached specimens were divided into three subgroups (n = 4 teeth each) for restoration according to predetermined posttreatment time intervals (immediately, 1 week, and 2 weeks). Bonded specimens were then sectioned and subjected to µTBS testing. The data were analyzed using Kruskal–Wallis and Mann–Whitney U-tests at $\alpha = 0.05$. **Results:** There was a significant difference in the µTBS of the resin composite to enamel in groups that were bonded immediately after bleaching and in the control group ($P < 0.05$). Compared to the control group, the µTBS in Groups 2, 3, and 4 decreased significantly 1-week postbleaching ($P < 0.05$). No significant difference in µTBS was observed between the bleached and unbleached groups 2 weeks after treatment ($P > 0.05$). **Conclusions:** Adhesive restorative procedures could not be performed immediately or after 1 week irrespective of the type or concentration of bleaching system used. Composite restorations on bleached enamel surfaces should be performed after an interval of at least 2 weeks.

Keywords: *Bleaching regimen, enamel, microtensile bond strength, resin composite*

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Introduction

Tooth whitening systems were first introduced by Haywood and Heymann in 1989.[1] Since then, bleaching and adhesion have become significant topics in restorative dentistry not only because of the availability of new technologies and materials but also because of improved knowledge and understanding of the mechanisms of action of bleaching agents and adhesive systems.[2] At present, vital tooth bleaching can be accomplished by carbamide or hydrogen peroxide (HP) bleaching agents at different concentrations in esthetic dentistry.[3,4] Despite the mechanism by which teeth are whitened by HP not being fully understood, it is believed that the peroxide can diffuse into the enamel and dentin and oxidize a variety of colored organic and inorganic compounds, leading to a reduction in tooth color.[5,6] Although tooth whitening is generally regarded as safe, adverse reactions, such as tooth sensitivity and gingival irritation, may occur following whitening procedures performed at home or in the office.[7-9]

There have been reports of a relationship between bleaching agents and the strength with which composite materials bond to enamel.[10-13] Studies have reported a reduction in enamel bond strength when the bonding procedure is carried out immediately or up to 1 week after vital bleaching with HP and carbamide peroxide (CP)-based agents.[11-14] This reduction in enamel bond strength has become a concern in esthetic dentistry with regard to clinical applications that involve resin bonding.[15]

The reason for the reduction in bond strength is the presence of residual oxygen, which inhibits free radical polymerization.[11-13] Various methods have been advocated to counteract the adverse effects related to the lower bond strength following bleaching. A number of studies

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have reported on bonding of adhesive systems and resin composite materials associated with bleaching regimens, and most recommend waiting for a period after bleaching for optimal bonding of adhesive systems and composite resins.[3,4,13,14,16-18] Therefore, it is important to know how the concentration of bleaching agents would affect optimal bonding performance in clinical practice and what is the ideal waiting time to achieve this goal.

With more emphasis now placed on esthetics, tooth discoloration has become of increasing concern. Bleaching is one of the most conservative and cost-effective methods of improving the appearance of teeth and may be indicated before direct or indirect restorative treatment procedures for shade matching between teeth and the restorative material.[19,20] In addition, it has been reported that enamel bleaching before resin composite restoration would achieve a more satisfactory outcome.[21] There are a number of studies in the literature concerning the strength of enamel-resin bonding to bleached enamel and the waiting time after bleaching most suitable for optimal bond strength. However, the findings of these studies are quite variable, and the data reported for waiting times after bleaching are conflicting.

The purpose of this study was to evaluate the effects of three different bleaching regimens using different concentrations of bleaching agent (opalescence 10%, whiteness perfect 16%, and whiteness HP 35%) on the microtensile bond strength (μTBS) of resin composite to bleached enamel after three different time intervals since restorative treatment (immediately, and at 1 and 2 weeks).

The null hypothesis of the study was that there would be no significant difference between the different bleaching regimens used at different concentrations or between the different time intervals elapsed since bleaching on the μTBS of the resin composite to enamel when compared with a nonbleached control.

Methodology

Tooth selection and preparation

Forty erupted sound human third molar teeth indicated for extraction were collected from patients who had received detailed verbal and written information about the study and signed an informed consent form. After extraction, each tooth was cleaned with a hand scaler (Hu-Friedy, Chicago, IL, USA) to remove any remaining soft tissue remnants, treated with pumice prophylaxis, and then stored in isotonic saline solution containing a few crystals of thymol at 4°C for the purpose of inhibiting microbial growth and used within 2 months after extraction. The buccal surfaces of all teeth were examined for possible cracks and defects using a stereomicroscope (Olympus SZ61, Olympus Optical Co., Tokyo, Japan) at ×40 magnification before preparation of the enamel surface. Any of the teeth showing such defects were excluded from the study. The buccal surfaces of the teeth were flattened using a polishing machine (Metaserv 250, Buehler Ltd., Lake Bluff, IL, USA) with wet 300-grit silicon carbide paper for 20 s to form a uniformly flat enamel surface. The flattened surfaces of all teeth were then polished with wet 600-grit silicon carbide paper for 60 s to create a standardized smear layer.[18] Next, all surfaces were washed with tap water to remove any particles of silicon carbide paper. Each tooth was then carefully reexamined under the stereomicroscope to confirm that the flattened enamel surface was secure and that the dentin surface was not exposed. The crown of each tooth was sectioned from the roots 2 mm below the cementoenamel junction using a slow-speed diamond blade (Isomet 1000, Buehler Ltd.) with copious water irrigation, and the coronal pulp tissue was removed using a hand instrument. After completion of these procedures, the teeth were randomly divided into four groups, i.e., an unbleached (control) group (n = 4 teeth) and three bleaching groups (n = 12 teeth per group). The flattened buccal surface of each tooth was bordered with an area of nail polish approximately 5 mm × 5 mm (25 mm^2) where the bleaching and adhesive restorative procedure would be performed.[17] All the preparations were performed by the same investigator to standardize the procedure. The materials used in the study are listed in Table 1.

Bleaching procedure

The prepared specimens were then randomly divided into four treatment groups according to the bleaching agents and concentrations used as shown in Table 2.

After completion of the bleaching procedures, all specimens except for those in the control group was divided into three subgroups (n = 4 teeth in each subgroup) for restoration according to predetermined posttreatment intervals as follows: (a) immediately after bleaching; (b) after immersion in isotonic saline for 1 week; and (c) after immersion in isotonic saline for 2 weeks.

Restorative procedures

After the bleaching regimens were completed, four specimens from each of the respective posttreatment groups and the control group were submitted for the adhesive restorative procedure. The flattened enamel surfaces were washed with water spray and air-dried with an air syringe. The flat areas were etched with 32% phosphoric acid gel (Scotchbond Universal etchant; 3M ESPE, St Paul, MN, USA) for 15 s, rinsed with water spray, and air-dried. A multimode adhesive system (Single Bond Universal; 3M ESPE, Neuss, Germany) was then applied with a disposable microbrush tip to the acid-etched enamel surface according to the manufacturer's instructions. The adhesive-coated surfaces were light cured for 10 s with a halogen light-curing unit (Optilux 501; Kerr Corp., Danbury, CT, USA) operating in standard mode and emitting not <650 mW/cm^2 as measured with a light meter (Hilux; Benlioglu Dental Inc., Ankara, Turkey) before beginning polymerization. Next, three layers of A2 shade nanohybrid resin composite (Filtek Z550; 3M ESPE) were placed on the flattened enamel surfaces incrementally using a hand instrument until a buildup approximately 5 mm high was achieved. Each layer of the resin composite was placed <2 mm thick and light-cured separately for 40 s. The restored specimens were then stored in isotonic saline at 37°C for 24 h before the µTBS test. The bleaching procedure and details of specimen preparation for the µTBS test are schematically illustrated in Figure 1.

Microtensile bond strength test

After storage in isotonic saline, the bonded specimens were positioned as perpendicular as possible in relation to the diamond saw of the machine and then serially sectioned in a vertical (occlusogingival) following mesiodistal direction with a low-speed diamond saw (Isomet 1000; Buehler Ltd) under water cooling to obtain multiple enamel-resin beams with a cross-sectional surface area of approximately 1 mm^2 (1 mm × 1 mm) for the µTBS testing procedure. A total of thirty beams were created per group and subjected to the µTBS test. For µTBS testing, the ends of the obtained beams were fixed with cyanoacrylate glue (Zapit, Dental Ventures of America, Corona, CA, USA) onto a µTBS testing device (Bisco, Schaumburg, IL, USA), and the fixed specimens were stressed to tensile force loading at a crosshead speed of 0.5 mm/min until failure. After failure, the specimens were carefully removed from the holder and the adhesive area at the fracture site was measured with a digital caliper (Mitutoyo, Tokyo, Japan). The bond strength was determined by dividing the

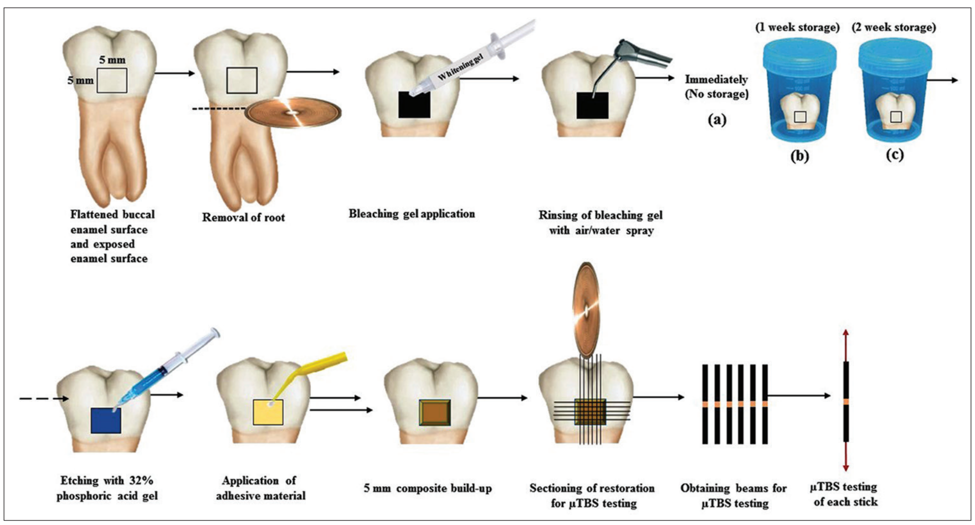

Figure 1: Schematic illustration of the bleaching procedure used and specimen preparation for testing of microtensile bond strength

Table 1: Materials used in the study according to manufacturer's specifications

Product	Classification	Composition	Lot #	Manufacturer
Opalescence	Home bleaching	10% carbamide peroxide	b7jxx	Ultradent Products Inc., South Jordan, UT, USA
Whiteness Perfect	Home bleaching	16% carbamide peroxide	120913	FGM, Joinville, SC, Brazil
Whiteness HP	Office bleaching	35% hydrogen peroxide	270413	FGM, Joinville, SC, Brazil
Filtek Z550	Nanohybrid Universal Restorative	Matrix: Bis-GMA, UDMA, Bis-EMA, PEGDMA, TEGDMA Filler: Zirconia/silica particles, 67.8% (v/v), 81.8% (w/w), Average filler size 0.1–10 (µm)	432323	3M ESPE, St. Paul, MN, USA
Single Bond Universal	Total-etch, Self-etch, Selective-etch – no matter	MDP phosphate monomer, dimethacrylate resins, HEMA, Vitrebond™ copolymer, filler, ethanol, water, initiators, silane	494756	3M ESPE, St. Paul, MN, USA

Bis-GMA: Bis-phenol A diglycidylmethacrylate; UDMA: Urethane dimethacrylate; Bis-EMA: Bis-phenol A polyethoxylated dimethacrylate; PEGDMA: Polyethylene glycol dimethacrylate; TEGDMA: Triethyleneglycol dimethacrylate; HEMA: 2-hydroxyethylmethacrylate failure load by the adhesive area, with the result expressed in MPa.

After completion of μTBS testing, the failure mode was observed using a stereomicroscope (Olympus SZ61) at × 40 magnification and classified as cohesive failure in enamel, adhesive failure between the enamel-composite interface, or cohesive failure in the composite.[18,22]

Statistical analysis

The μTBS data were initially analyzed using the Shapiro–Wilk test for normality of the data distribution. The data were not confirmed to be normally distributed for opalescence 10% for 2 weeks and compliance could not be achieved as a result of the transformations that were performed, so subsequent statistical analyses were performed using nonparametric Kruskal–Wallis and Mann–Whitney U-tests at a significance level of 5%. The Student's *t*-test was used to test for differences in mean μTBS values between the control and treatment groups at each time interval except for opalescence 10%. All statistical analyses were carried out using Statistical Package for Social Sciences software version 21 (IBM SPSS Statistics, Chicago, IL, USA).

Results

The minimum, maximum, and mean μTBS values for each study group and their significance are presented in Table 3 and Figure 2. The results of the data analysis revealed that the bond strength values were significantly different between the control and experimental groups immediately and 1 week after bleaching ($P < 0.05$). The μTBS of the composite resin to enamel immediately after bleaching showed significantly reduced bond strength, irrespective of the bleaching regimen applied when compared with the control group ($P < 0.05$). The μTBS values for opalescence 10%, whiteness perfect 16%, and whiteness HP 35% indicated significantly decreased bond strength 1-week posttreatment when compared with the control group ($P < 0.05$), with no significant between-group differences (opalescence 10% versus whiteness perfect 16%; $P > 0.05$). Whiteness HP 35% exhibited significantly increased

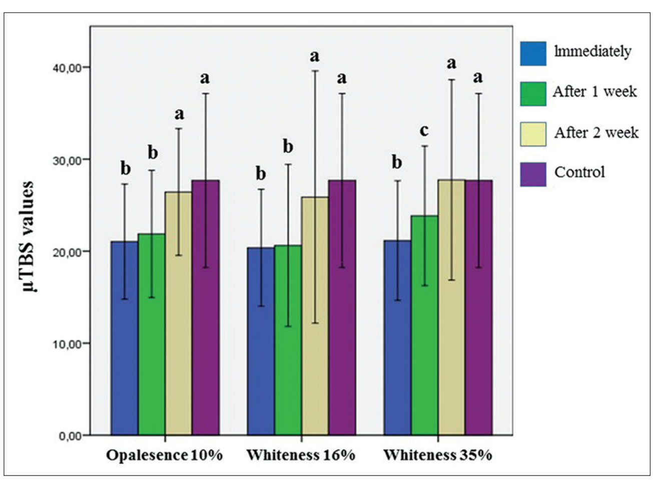

Figure 2: Mean microtensile bond strength values for each study group and significance

Table 2: Treatment groups and application procedures of the bleaching agents used in the study

Group	Application procedure
Group 1	This was the control group, with no bleaching treatment applied (*n*=4 teeth)
Group 2	Specimens were bleached with 10% CP home bleaching gel (*n*=12 teeth), i.e., 10% Opalescence (Ultradent Products, Salt Lake, UT, USA). The bleaching gel was applied onto the flattened buccal enamel surface according to the manufacturer's instructions at a thickness of approximately 1-2 mm. Bleaching was performed for 8 h daily on 14 consecutive days; after each bleaching session, the specimens were thoroughly rinsed with air/water spray for 30 s to remove the bleaching gel. The teeth were stored in isotonic saline at 37°C for the remainder of each treatment day
Group 3	Specimens were bleached with 16% CP home bleaching gel (*n*=12 teeth), i.e., 16% Whiteness Perfect (FGM Dental Products, Joinville, SC, Brazil). The bleaching gel was applied onto the flattened buccal enamel surface according to the manufacturer's instructions at a thickness of approximately 1-2 mm. Bleaching was performed for 4 h daily on 14 consecutive days; after each bleaching session, the specimens were thoroughly rinsed with air/water spray for 30 s to remove the bleaching gel. The teeth were stored in isotonic saline at 37°C for the remainder of each treatment day
Group 4	Specimens were bleached with 35% hydrogen peroxide bleaching gel (*n*=12 teeth), i.e., 35% Whiteness HP (FGM Dental Products). The gel was mixed according to the manufacturer's instructions and applied onto the flattened buccal tooth surface at a thickness of approximately 1-2 mm. Application of bleaching gel was performed in three sessions, with a between-session interval of 3 days. At each session, the bleaching gel was applied to the enamel surface on two occasions, each lasting 15 min (i.e., a total of 30 min of bleaching in each session) separated by a 5-min interval without light activation. The specimens were then thoroughly rinsed with air/water spray for 30 s to remove the bleaching gel. The teeth were stored in isotonic saline at 37°C for the remainder of each treatment day

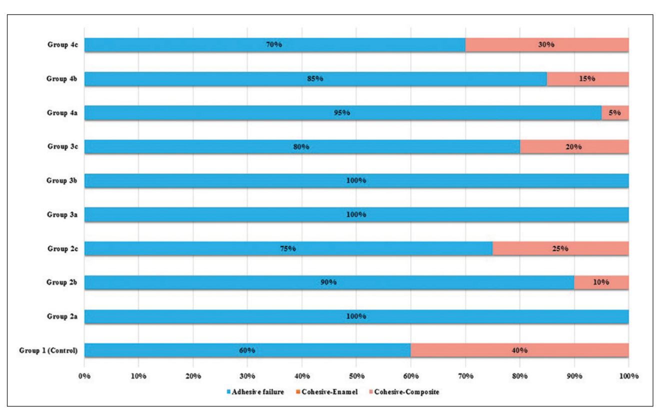

Figure 3: Distribution of failure modes (%) according to group

Table 3: Minimum, maximum, and mean microtensile bond strength values for the experimental groups

Groups	Subgroups	*n* (beams)	Minimum	Maximum	Mean±SD
Group 1 (control)	-	30	19.20	39.42	27.68±4.73[a]
Group 2	Group 2a	30	15.34	26.19	21.03±3.13[b]
	Group 2b	30	15.42	29.11	21.87±3.46[b]
	Group 2c	30	18.65	33.42	26.42±3.44[a]
Group 3	Group 3a	30	14.00	26.84	20.37±3.18[b]
	Group 3b	30	14.63	33.24	20.62±4.40[b]
	Group 3c	30	13.65	46.06	25.88±6.85[a]
Group 4	Group 4a	30	14.82	26.85	21.15±3.25[b]
	Group 4b	30	16.29	30.20	23.84±3.79[c]
	Group 4c	30	19.61	37.42	27.74±5.45[a]

Note: Mean values identified with a different superscript letter in the same column indicate a statistically significant difference ($P<0.05$). SD: Standard deviation

bond strength when compared with opalescence 10% and whiteness perfect 16% at 1-week posttreatment ($P < 0.05$) with a significantly lower bond strength value when compared with control ($P < 0.05$). After 2 weeks, the μTBS values for the composite resin to enamel in all the bleaching groups returned to a value comparable with that of the control ($P > 0.05$), with no significant differences between the bleached groups ($P > 0.05$). No significant differences were detected in μTBS with regard to the type and concentration of bleaching agents used for any of the time intervals ($P > 0.05$) except for the whiteness HP 35% bleaching agent after 1 week.

The distribution of type of failure mode in each group is presented in Figure 3. Adhesive failure was the predominant type of failure in the bleached groups. The control group showed a 40% cohesive failure rate for the resin composite. No cohesive failure in enamel was observed in the control group or in the bleached groups at any posttreatment time interval.

Discussion

A variety of bleaching systems are now available for dental professionals and come in different concentrations and formulations (carbamide or HP) for conservative and safe whitening of tooth discoloration.[9] It has been previously reported that several factors can influence the enamel-resin bond strength after the bleaching process, including the adhesive system used, the type of bleaching system and its concentration, and the wait time after bleaching.[4,23-25] The present study evaluated the effects of three bleaching regimens with different concentrations (opalescence 10%, whiteness perfect 16%, and whiteness HP 35%) and postbleaching time (immediately, and at 1 and 2 weeks) on the μTBS of resin composite to enamel. According to our results, the null hypothesis must be rejected.

In esthetic restorative dentistry, many patients ask their dentist to replace existing older tooth-colored resin immediately after the bleaching procedure due to differences in color between restorations and tooth enamel since the existing restorations did not change color with bleaching.[17] Therefore, it is important for dentists to know the effect of the most commonly used bleaching agents on enamel-resin composite bond strength. In the present study, we found significantly reduced μTBS of resin composite to enamel in bleached groups compared with the control when the restorative procedures were performed immediately after bleaching either in carbamide or HP form. These results are in agreement with previous investigations reporting a significant decrease in bond strength when restorative procedures are done immediately after enamel bleaching.[4,13,14,17] According to previous investigations, even low concentrations of bleaching agents, such as 10% CP, as used in this study, decrease the enamel-resin bond strength.[3,14,26] The most frequently cited reason for this is the presence of residual oxygen in the bleached enamel, which hinders polymerization of the resinous materials.[11,13] In addition, physical and chemical alteration might occur in enamel immediately after the bleaching procedure,[3,12,13,15,23] so the enamel-resin bond strength is reduced. In contrast with our findings, it has been reported that the adverse effects of residual oxygen can be removed from tooth enamel pores by application of phosphoric acid etching.[27] Similarly, Dishman *et al*.[13] have suggested that the oxygen rich layer in enamel after bleaching can be removed by the action of phosphoric acid etching before application of adhesive. However, a positive enamel-composite bond strength action of phosphoric acid etching before application of adhesive on bleached enamel was not observed in the present study. In addition, it has been reported that use of an alcohol-based bonding agent can minimize the inhibitory effects of the bleaching process through the interaction of alcohol with residual oxygen in the enamel pores.[10,23,28,29] In contrast with our findings, Sung *et al*.[23] reported that use of alcohol-based bonding systems resulted in less compromised bond strength when restorations were done immediately after the bleaching procedure. In the present study, Single Bond Universal, which contains an ethanol/water-based solvent, was used as a bonding agent after acid etch application in all groups. However, in contrast to their results, our findings indicate a significantly reduced immediate bond strength when compared with the control, and this might be attributed to use of phosphoric acid etching followed by application of an ethanol-based adhesive system were not eliminated the bonding inhibitory effects of the bleaching agents.

The commercially available bleaching agents contain variable CP and HP concentrations ranging from 10% to 22% and from 30% to 40%, respectively.[3,4] Higher concentrations of bleaching agents are generally preferred when rapid bleaching is desired.[26] However, when the mechanism of action of these bleaching systems and the ease with which their low molecular free radicals diffuse through the dental substrates are considered, more marked dental structural alterations can be expected when more highly concentrated bleaching systems are used.[26,30] Previous studies have examined the effects of different bleaching systems in different concentrations on enamel-resin bond strength in some depth, and most recommend a wait time ranging from 1 week to 6 weeks after bleaching to prevent reduced bond strength of restoration work, even when using a low-concentration bleaching system (e.g. 10% CP).[2,3,13,14,17,18,20,31-33] However, the optimal wait time after bleaching is still controversial in the literature. Some investigators suggest that a 1-week wait time after bleaching is enough to reverse reduced bond strength[4,17] while others recommend a 2-week wait time for complete restoration of bond strength.[3,14] In the present study, enamel-composite bonding procedures were also performed 1 and 2 weeks after the bleaching procedure, and during these times, samples were kept in isotonic saline that was renewed daily. In contrast with previous findings, we observed that the μTBS of the resin-enamel composite applied 1 week after bleaching was significantly reduced in all the bleaching groups, with no significant differences between the 10% CP and whiteness perfect 16% group. In the whiteness HP 35% group, a significant increase in the μTBS of the resin-enamel composite was observed when compared with the other treatment groups 1 week after bleaching, and this can be attributed the presence of thickener (carbopol) in whiteness HP 35%, which might improve the adhesion and extend the release of active bleaching molecules,[31,34] such that bonding performance is increased. One of the possible explanations for the reduced bond strength 1 week postbleaching in the present study is that residual oxygen was retained at the enamel surface and subsurface which might have allowed the whitening action to continue and did not completely leach out during the storage period, resulted in incomplete polymerization of the adhesive material both on the enamel surface and at the base of the adhesive layer.[13,14,35] In addition, the ongoing oxidizing effect of free radicals and metabolites from the bleaching agents might have affected the enamel surface and interprismatic regions by altering the enamel crystallites, resulting in decreased enamel-resin bond strength.[14] Therefore, we can assume that the ongoing oxidizing effect of bleaching gels,[13,14,35] regardless of the concentration, and structural defects in the enamel surface and subsurface due to loss of calcium and phosphate minerals[32,33,36-38] affected the retentive quality of the adhesive material by 1-week postbleaching in the present study. In addition, application of phosphoric acid to the altered surface structure of the enamel might also result in a more aggressive etching pattern, which could weaken the enamel by increasing its porosity and prevent optimal bonding.[39] Bistey *et al*.[40] reported that bleaching agents used at home or in the office, regardless of concentration, can produce structural alterations in enamel, which is in agreement with the present findings. On the other hand, Barcellos *et al*.[26] reported that the extent of the damage done by bleaching products to tooth structures depends on the concentration of the bleaching agent used, with more highly concentrated agents causing lower tooth restoration bond strength, which is not consistent with the results of our study.

Although bleaching agents have adverse effects in terms of enamel-resin bond strength, this effect is temporary, and the enamel recovers its adhesiveness after a period of delay. In the present study, enamel-resin bond strength returned to normal levels, comparable with those of nonbleached samples, after 14 days of storage in isotonic saline. Our present findings, in this regard, are in agreement with previous studies showing that storage of samples in artificial saliva or distilled water results in complete recovery of reduced enamel bond strength.[2,3,14,18,20,25,33]

When the failure pattern of the groups was determined, the unbleached groups showed a predominantly adhesive (60%) and subsequent composite fracture (40%) pattern, so the high bond strength obtained was consistent with the fracture pattern. The groups that underwent bonding immediately after bleaching and 1 week later had very similar fracture patterns, being predominantly adhesive failure, which appears consistent with the μTBS results. These fracture patterns also confirm that resin bonding to bleached enamel was not negatively affected by the high concentration of bleach used in the whitening procedure performed 1 week before bonding. When the delay after bleaching was 2 weeks, all groups showed similarly predominant adhesive (range 70%–80%) and cohesive (range 20%–30%) behavior in composite failure, suggesting that enamel recovered its adhesiveness.[17,36]

It is important to emphasize that adhesion to enamel after bleaching remains a major problem in esthetic dentistry. Therefore, further investigations, especially *in vivo* and *in situ*, are necessary to corroborate our findings, considering that saliva and its high mineral content as well as enzymes such as peroxidase might positively affect bond strength results and the effect of the time lapse since treatment with the various bleaching regimens.

Conclusions

Based on the findings of this study, it may be concluded that immediate and after 1-week of bonding of composite to bleached enamel compromises μTBS even when the enamel is etched with phosphoric acid and an alcohol-based bonding agent is subsequently used, irrespective of the type or concentration of the bleaching system used. It is advisable that composite restorations on bleached enamel surfaces be performed after an interval of at least 2 weeks, regardless of the concentration of bleaching agent used. The fracture patterns in the different groups in our study seemed to be consistent with the bond strength values obtained in each bleaching group.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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