Original Article

Effect of Time Interval between Bleaching and Bonding on Tag Formation

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Abstract

The objective of this study was to assess penetration of adhesive material in enamel bleached with 35% hydrogen peroxide using optical polarized light microscopy. Extracted human teeth were randomly assigned to 5 groups, each representing a specific time interval between bleaching and the application of an adhesive material. They were designated as: (TC) the control group—restorations in unbleached teeth; (T0) comprising restorations carried out immediately after bleaching; (T7) comprising restorations 7 days after bleaching; (T14) comprising restorations 14 days after bleaching; and (T21) comprising restorations 21 days after bleaching. Length of resin tags was measured with an Axiophot photomicroscope at a $\times 400$ magnification, and the results subjected to an ANOVA for a comparison between groups, with a p value of < 0.05. Differences between the groups were verified using a Tukey test at a confidence level of 5%. The specimens in the control group (TC) and experimental groups T7, T14 and T21 showed better penetration of adhesive material into enamel in comparison with experimental group T0. This suggests that a gap of at least 7 days should be left between bleaching enamel with 35% hydrogen peroxide and placing adhesive bonding agents and undertaking resin composite restoration work.

Key words: Enamel—Dental bleaching—Time intervals—Tags formation

Introduction

In addition to the interest of dental professionals in solving the problem of chromatic inconsistencies, the continuous search and demand for an esthetically healthy and harmonious smile, in the majority of cases encouraged by modern society, has given rise to the development of dental bleaching techniques.

The methods employed for dental bleaching are relatively simple, and when performed correctly, make it possible to obtain surprisingly good esthetic results, while preserving the integrity of dental structure.

In 1989, Haywood and Heymann⁷⁾ proposed the application of bleaching products based on carbamide peroxide. These are delivered in an acetate custom-fitted tray, which, when properly indicated, applied and supervised by a dentist, produce highly satisfactory esthetic results^{1,7,8,11,13-17)}.

As an alternative, we may also use bleaching products based on 35% hydrogen peroxide. These should be applied while isolating the operating field in order to protect soft tissue from being adversely affected by the bleaching material^{1,2,11}. Clinically, there is no significant alteration in the color or structure of an esthetic restoration after bleaching. Therefore, replacement of esthetic restorations should be carried out after bleaching, as alteration in color is more pronounced in hard dental tissues than in restorative material^{7,13–17}.

However, there is some disagreement as to the degree to which bleaching affects enamel and the optimum time interval between bleaching and the placement or replacement of restorations^{9,10,21)}. Various studies have reported *in vitro* marginal leakage¹⁰⁾ and measured bond strength of adhesive material to enamel after dental bleaching treatment^{5,20,22,23)}. However, there is no conclusive evidence concerning the adaptation and formation of resin tags in bleached enamel.

The purpose of this study was to analyze the penetration of adhesive material in enamel bleached with 35% hydrogen peroxide (Opalescence X-TRA) using light optical microscopy. The penetration of the bonding agent was evaluated at various time intervals between bleaching and restorative treatment.

Materials and Methods

Thirty sound, recently extracted human

premolars were collected from patients ranging in age from 11 to 16 years. The study protocol was approved by the Human Subject Review Committee of the University of the State of São Paulo, Araçatuba School of Dentistry (Araçatuba, SP, Brazil).

The teeth were cleaned, washed and polished with a slurry of pumice and water, and then stored in distilled water at room temperature until use. They were randomly assigned to 5 groups (n=6). Apart from the control group, each group represented a specific time interval between bleaching and the application of adhesive material (Table 1). The roots of the teeth were embedded in acrylic resin, and the coronal portion left exposed. All samples were treated on both the buccal and lingual surfaces.

The control specimens were first given a prophylaxis using pumice and water and a prophylaxis cup operated at low speed¹²). This was followed by rinsing with an air/water spray, and drying with compressed air. Next, the enamel surfaces were conditioned with acid for 1 min¹²⁾ using 35% phosphoric acid gel. The enamel surfaces were then washed thoroughly for 15 sec, and dried with an air spray. Subsequently, Scotchbond Multi-Purpose (3M-ESPE) was applied with a microbrush, air-dispersed for 5 sec and then polymerized for 20 sec. Z100 composite resin (3M-ESPE) was applied to an approximate thickness of 2 mm, followed by light curing for 40 sec. All light curing in this experiment was carried out with an Ultralux Lens (Dabi Atlante) with an output of $450 \,\mathrm{mW/cm^2}$.

The teeth in each of the experimental groups were also given a prophylaxis as described above, followed by the application of Opalescence X-TRA (Ultradent Products). After washing and drying the surfaces, an approximately 1 mm thick layer of Opalescence X-TRA was applied using a brush. Immediately after application, the buccal and lingual surfaces were exposed to two sources of halogen light (Ultralux) with an output of 450 mW/cm² for 30 sec.

This was repeated 10 times, with a 30-sec light exposure interval after each application.

Components	Amount
Carboxymethylcellulose	0.4%
Liquid Sorbitol	6%
Potassium Chloride	1 g
Sodium Chloride	1 g
Sodium Fluoride	2 mg
Magnesium Chloride	$50\mathrm{mg}$
Calcium Chloride	150 mg
Potassium Phosphate	400 mg
Nipagin	0.2%
Distilled water	1,000 ml

 Table 1 Composition of artificial saliva, modified from Bo krasse³⁰, 1986

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Table 2 Control and experimental group averages (μm) and results of Tukey test

Time interval	Average (µm)	Standard Deviation	N	Result of Tukey test*
Control (TC)	12.35	2.44	6	В
Immediate (T0)	8.22	2.99	6	А
7 days (T7)	12.36	2.13	6	В
14 days (T14)	11.98	1.60	6	В
21 days (T21)	13.22	2.86	6	В

 * Same letters indicate no significant statistical difference between groups (p<0.05).

Next, the bleaching material was left in place for an additional period of 10 min. This time, however, the surfaces were not exposed to halogen light.

After the required time interval had elapsed, the material was rinsed off with an air/water spray for 60 sec. The teeth assigned to time interval T0 were immediately treated with 35% phosphoric acid gel, rinsed and treated with an adhesive, as described for the control group (TC).

For the other groups, adhesive treatment was completed after 7 days (T7), 14 days (T14) and 21 days (T21) following bleaching. During storage time, the specimens were kept in an incubator at 37°C in artificial saliva³ (Apothicário Pharmacy of Manipulation, Araçatuba, Brazil) which was changed daily (Table 1).

At the end of the experimental period, each tooth was cut into 5 bucco-lingual plane sections to a thickness of approximately 200 μ m (Isomet 2000, Buehler). Three sections were then selected and further polished using aluminum oxide abrasive paper with grit numbers of 80, 360 and 600 to a thickness of approximately 100 μ m, as measured by a digital caliper. The sections were decalcified in 40% nitric acid for approximately 60 sec, i.e. until the enamel dissolved, leaving the adhesive and resin tags behind. The specimens were then immersed in distilled water, mounted on glass and covered with a glass cover slide. The edges were sealed with Canadian oil¹².

The resin sections of each tooth were ana-

lyzed and measured using an Axiophot light microscope (ZEISS DSM-940 A, Oberkochen, Germany) at $\times 400$ magnification and an optical 40/075 micrometric. The resin tags of each vestibular and lingual resin section were measured with a single calibrated examiner. The resin tags of each section were measured at three pre-determined sites: the occlusal third, middle third and cervical third. The final lengths of the resin tags for each specimen represents the average of all the measurements made for each section. These, average tag lengths were then submitted to statistical analysis using an ANOVA to give a comparison between the groups at a 5% level of significance (p < 0.05).

Differences among the groups were verified using a Tukey test with a confidence level of 5%. The most representative resin tag regions from each group was photographed with an Axiophot photo-microscope at a magnification of $\times 400$ using a micrometric ocular 40/075.

Results

The ANOVA showed some significant differences between the groups. A Tukey test (p < 0.05) was then used to verify those differences (Table 2).

As shown in Table 2, no significant differences were found between the specimens from the control group (TC) and those of experimental groups (T7), (T14), (T21). On the other hand, groups TC, T7, T14, T21 showed superior penetration of adhesive material into enamel in comparison with that



Fig. 1 Resin tags of control enamel surface, where mean length of tags was 12.35μm (Original magnification × 400). CR: Composite Resin, T: Tags.

in the experimental group (TO). Unbleached control enamel (TC) and experimental groups (T7, T14 and T21) offered an excellent environment for the formation of resin tags, presenting numerous uniform resin tags of similar lengths (Figs. 1, 2 and 3) and contiguity with the composite resin.

Placement of an adhesive restoration immediately after bleaching (T0) resulted in a statistically significant decrease in the penetration of the adhesive material into the enamel in comparison with that in the control group (TC) and experimental groups (T7, T14 and T21) (Table 2 and Fig. 3).

Discussion

Dental esthetics have benefited much from technological advances, and there is much literature available on techniques and restorative materials that offer superior esthetic results^{1,4,7,13-17)}.

The credit for this scientific achievement should go to Buonocore⁴⁾, who observed that conditioning enamel with acid allowed resin material to penetrate and adhere to enamel. At the time, he suggested that this was because such conditioning created porosity in the enamel. This greatly increased the available surface area and formed a new surface through the precipitation of new substances, removing layers of inert enamel and exposing surfaces more favorable to adhesion. Further-



Fig. 2 After 7 days (T7) quality and adaptation of resin tags much better than that observed in samples bonded immediately after bleaching, where mean length of tags was 12.36µm (Original magnification × 400). CR: Composite Resin, T: Tags.



Fig. 3 Quality of resin tags in enamel of immediate time interval (T0), where mean length of tags was 8.22µm (Original magnification ×400). CR: Composite Resin, T: Tags.

more, Buonocore emphasized that acid conditioning not only promoted a rough surface, but also the creation of a considerable number of micropores in the enamel, approximately 30,000 to 40,000 mm², into which adhesive material penetrated by capillary action. This allowed the adhesive material to penetrate into deeper layers by means of the formation of resin tags, no longer remaining confined to the surface. This method is a clinically conservative, non-traumatic, biocompatible means by which resins, polymerizable under oral conditions, can be bonded durably to enamel⁶.

In addition, the success of acid etching of enamel prior to the placement of adhesive materials has had a profound effect on the long-term behavior of those materials in the oral cavity^{12,18)}.

It has been observed that when teeth with composite resin restorations are bleached, a color discrepancy is created which frequently requires the replacement of existing restorations^{1,7,10,11,13-17)}. Thus, in view of what has been reported, and supported by the results of this investigation, we believe that it is important to observe the most favorable time interval before carrying out an adhesive restoration on bleached teeth^{9,10,21)}. This allows for the release of peroxide oxidation products, such as residual oxygen⁹⁾.

In this experiment, the control group (not bleached) showed numerous tags that were well defined and contiguous to the resin material, demonstrating good penetration of the adhesive material into the enamel (Fig. 1). This observation is in agreement with Titley et al.¹⁹, who using similar experimental conditions reported the same findings obtained by means of a scanning electron microscopy analysis. In the present study, tag formation in the samples at time intervals of 7 (T7), 14 (T14), and 21 days (T21) showed the same extent of penetration in conditioned enamel. The tags were longer than those seen in the immediate time group, contiguous to the resin material and had satisfactory definition and distribution (Fig. 2). This data differs from that for the group that had the adhesive system applied immediately after bleaching. Here we found inferior tag formation that was smaller, thinner, less frequent, lacking in uniformity and poorly defined (Fig. 3). This would account for the decreased adhesive potential of the resinous material to the bleached enamel, and possibly also the reduced average values for shear bond strength in the specimens treated with 35% hydrogen peroxide²¹⁾.

These findings may explain the difference in the appearance and length of the resin tags between the control and experimental groups (7, 14 and 21 days) and immediate time interval group (T0), which demonstrated a diverse and inferior appearance. This supports the possibility of an interaction between the resin adhesive and the peroxide located on, or close to, the bleached enamel surface. This is in agreement with the findings of Titley *et al.*¹⁹⁾, who proposed that this interaction could inhibit polymerization of the adhesive due to the presence of oxygen, as well as increase the porosity of the resin material as a result of the release of oxygen.

This proposition has been further substantiated by Dishman *et al.*⁵⁾ who reported a high concentration of residual oxygen in enamel pores, and that this interfered with the polymerization of adhesive in enamel that had been bleached. They also suggested that the exact depth of the oxygen-rich layer is unknown, however, it must exceed from 5 to 10μ m in depth, otherwise it would be removed by the action of the phosphoric acid that is applied prior to the application of the resinbased adhesive.

Based on this premise, we suggest that the extension of tags of adhesive material may be compromised when applied immediately after bleaching and acid conditioning. Another factor that should be considered is that the teeth in this experiment were stored for a prolonged period of time in artificial saliva prior to the application of the adhesive. In other words, the storage time may have contributed to the complete elimination of the residual peroxide.

The results of the present study suggest that a period of at least 7 days should be left between the use of 35% hydrogen peroxide bleaching material and restorative procedures that require acid etching and adhesive bonding materials.

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