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Toothbrushing abrasion susceptibility of enamel and dentin bleached with calcium-supplemented hydrogen peroxide gel

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ABSTRACT

The objective of this study was to evaluate enamel and dentin susceptibility to toothbrushing abrasion, after bleaching with 7.5% hydrogen peroxide (HP) gel supplemented or not with 0.5% calcium gluconate (Ca). Toothbrushing was performed immediately and 1 h after bleaching, with two suspensions (high and low abrasivity). Bovine enamel and dentin specimens were divided into 12 groups (n = 10) according to the bleaching gel (with and without Ca), slurry abrasivity (high or low) and elapsed time after bleaching (immediately and after 1 h). As control, a group was not bleached, but abraded. The treatment cycle (7 d) consisted of bleaching (1 h) and toothbrushing (135 strokes/day) immediatelly or after 1 h of artificial saliva exposure. Surface roughness and surface loss (μm) were measured by profilometry and analysed by three-way ANOVA (5%). Surface roughness means were significantly influenced by slurry abrasivity ($p < 0.0001$). For enamel loss, significant triple interaction was observed ($p < 0.0001$). HP-bleached groups and immediately brushed with high-abrasive slurry exhibited increased loss (1.41 ± 0.14) compared to other groups (μm). Control and HP + Ca-bleached groups brushed after 1 h with low abrasive slurry presented the lowest loss ($0.21 \pm 0.03/0.27 \pm 0.02$). For dentin loss, significant interaction was observed for bleaching and interval factors ($p < 0.001$). 7.5%HP-bleached groups and immediately brushed showed significantly higher loss (8.71 ± 2.45) than the other groups. It was concluded that surface roughness increased when high abrasive was used, independently of bleaching. 7.5%HP increased enamel and dentin loss, mainly with high abrasive slurries. Calcium supplementation of bleaching gel reduced surface loss. Additionally, in order to minimize tooth wear susceptibility, it is recommended to delay brushing after bleaching.

Clinical relevance: After bleaching gel application, postponing toothbrushing is recommended, as well as brushing with low abrasive dentifrices. Additionally, supplementation of hydrogen peroxide gel with calcium-based remineralizing agent potentially reduces tooth loss after abrasion.

1. Introduction

Hydrogen peroxide has been widely used as a treatment agent for discolored teeth. Due to its low molecular weight and high instability, hydrogen peroxide is able to diffuse through enamel and dentin and decompose, releasing free radicals [1,2]. Its mechanism of action is based on the oxidative destruction of chromophores. This occurs by chemical degradation of molecular moieties responsible for absorbing visible eletromagnetic radiation, resulting in the increase of total reflectance of the substrate, and consequently, in its brighter appearance [3].

Although studies have proven the whitening efficacy of bleaching agents, adverse effects on dental tissues are also reported and must be carefully evaluated so that their use can be considered safe [4,5].

Dental bleaching has been previously related to microstructural tooth alterations, such as microhardness reduction [6] and changes in chemical composition of tooth [7]. Morphology defects, such as irregularities, depresssions and porosity formation have also been reported [8]. Although these changes were usually assigned to mineral loss due to the low pH of bleaching gels [4], degradation of

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the organic matrix by oxidation reaction was reported [9], with the described alterations also observed when near neutral agents were used [7,8]. In order to reduce the bleaching gel demineralization potential, the addition of remineralizing agents such as calcium and fluoride in the bleaching agents has been proposed [10–12].

Daily oral hygiene (brushing with regular toothpaste) is usually considered safe and not related to enamel potential harm [13]. However, individuals undergoing bleaching treatment may associate it with high abrasivity whitening toothpastes, potentially increasing its harmful effects on tooth surface [14]. Although abrasivity represents the predominantly mode of action of whitening toothpastes [15], it is known that the degree of dentifrice abrasivity may be related to potential wear, mainly on dentin [16]. Previous studies have reported a higher wear susceptibility of bleached enamel [17] and dentin after brushing [18–20].

Additionally, the effect of toothbrushing abrasion after bleaching on tooth surface roughness has been an issue of concern, since alterations on surface texture can lead to increased susceptibility to staining and bacterial adhesion, and consequently, further discoloration [21,22].

It has been shown that saliva is able to reharden demineralized bleached enamel [11]. Additionally, the supplementation of bleaching agents with remineralizing agents could contribute to reduce any potential harmful effect on bleached tooth [8]. Since bleaching may be related to tooth surface microstructural alterations and its association with toothbrushing can potentially increase surface loss, there is a concern regarding the optimal interval between the removal of the whitening gel and brushing, in order to reduce the possible interaction between the bleaching agent and the abrasive process on dental tissues.

Thus, the aim of this in vitro study was to investigate if the elapsed time between bleaching with 7.5% hydrogen peroxide (associated or not with calcium) and brushing (with high and low abrasivity slurries) would affect the enamel and dentin roughness and wear. The null hypotheses tested were that: a) 7.5% hydrogen peroxide, associated or not with calcium would not affect the substrate surface roughness and its susceptibility to abrasive wear; b) the elapsed time between bleaching and brushing would not interfere with roughness and wear, and; c) the slurry abrasivity would not influence roughness and wear.

2. Materials and methods

2.1. Experimental design

This study followed the complete factorial $3 \times 2 \times 2$ randomized design, with three experimental factors: 1. bleaching at 3 levels (no bleach, 7.5% hydrogen peroxide-HP, and HP with the addition of 0.5% calcium gluconate); 2. slurry abrasivity at 2 levels according to the RDA values (high and low); and 3. elapsed time between bleaching and abrasion at 2 levels (immediately and 1 h after bleaching), in a bleaching-abrasion cycling model using bovine enamel and dentin specimens. The specimens were randomly assigned into 12 groups (n = 10). The model was conducted for a total of seven consecutive days, and response variables were arithmetic mean surface roughness (Ra) and surface loss (in μm) measured by contact profilometry.

2.2. Enamel and dentin specimens preparation

Freshly extracted and intact bovine incisors were stored until required in 0.1% thymol solution, refrigerated at 4 °C. Cylindrical enamel and dentin specimens (3 mm diameter) were prepared from the labial surfaces of crowns and roots, respectively, using a custom-made diamond trephine mill. The specimens were then embedded in auto-polymerizable acrylic resin using cylindrical silicone molds (6 mm diameter, 3 mm depth), with the labial surface exposed for treatments, as previously described [23].

Embedded specimens were ground flat and polished with water-cooled sequential aluminum oxide abrasive papers (1200, 2400 and 4000 grit FEPA P; Struers, Ballerup, Denmark) in a polishing device (DP 10, Panambra, Sao Paulo, SP, Brazil). After each grindpaper, specimens were sonicated in deionized water for 5 min. The prepared specimens were examined in stereomicroscope (20X—Carl Zeiss, Stemi 2000, Tokyo, Japan) to verify the absence of cracks or other surface defects and then stored in ultrapure water to prevent dehydration.

The baseline profiles of the enamel and dentin surfaces were measured using a contact profilometer (MarSurf GD 25, Mahr, Göttingen, Germany). In order to maintain the reference surfaces for lesion-depth determination, and allow the exact superimposition of the baseline and post-treatment profiles, two parallel grooves were marked as guides on the resin at the sides of the embeded tooth structure. The specimens were positioned into a custom-made specimen holder attached to the profilometer, which allows the exact repositioning of the sample after the treatments. The diamond stylus moved from the first reference (resin) to the enamel or dentin area and then over to the other reference area (4.2 mm long). Three profile measurements were performed for each specimen at intervals of 0.25 mm.

For baseline superficial roughness analysis, the mean surface roughness values (Ra) were determined with a cut-off value of 0.8 mm, a transverse length of 0.8 mm, and a stylus speed of 0.1 mm/s, in the previously described profiles.

2.3. Bleaching and abrasive procedures

The specimens were randomly allocated into 12 groups (n = 10). The first group division was according to the bleaching gel: NB- no bleach; HP- 7.5% hydrogen peroxide gel (pH 5.62); HP + Ca- 7.5% HP gel with the addition of 0.5% calcium gluconate (pH 5.60). The experimental gels were modified by the manufacturer (FGM, Joinville, SC, Brazil), by adding or not the calcium compound in a 7.5% HP-based bleaching gel.

A 2 mm thick layer of the bleaching gel was daily applied to the specimens' surface and remained for 1 h. After this period, the gel was removed with a suction tip and the surface rinsed with ultrapure water for 20 s. In the non-bleached groups, the specimens remained in ultrapure water during the period corresponding to bleaching procedure.

After the described procedures, the specimes of each described group were divided according to the elapsed time after bleaching into two subgroups: immediatelly and one hour after bleaching for performing toothbrushing. The specimens of the group 1 h were kept in artificial saliva (1.5 mM $CaCl_2 \times 2H_2O$; 0.9 mM KH_2PO_4; 130 mM KCl; 20 mM of HEPES; pH adjusted to 7.0 with 1 M KOH solution) [24].

The specimens were finally divided into two subgroups according to the slurry abrasivity: H- high and L- low. The abrasive challenge was performed using an automatic toothbrushing machine (SEM-2T, Odeme Dental Research, Luzerna, SC, Brazil), which imparted reciprocating motion to standard medium bristle toothbrush stiffness (Sanifill Ultraprofissional, Sao Paulo, Brazil). The brushes were angled 15° in relation to the specimen surface to minimize grooves formation. During brushing, the right and left sides of the specimens, corresponding to acrylic resin with the reference groves, were protected with a stainless steel mask (0.1-mm thick), with an opened window of 2-mm wide, leaving an exposed area in the center of each specimen, preventing the abrasion of reference areas for the profilometric analysis.