

ORIGINAL ARTICLE

Efficacy of whitening oral rinses and dentifrices on color stability of bleached teeth

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Abstract

Objective: This study aimed to evaluate the effect of whitening toothpastes and mouthrinses on the color stability of teeth bleached with 16% carbamide peroxide (CP) after immersion in coffee solution.

Materials and methods: Specimens obtained from bovine incisors were bleached with 16% CP for 14 days. After bleaching, the specimens were stained in coffee solution for 24 h and randomly divided into eight groups according to the following products ($n = 10$): distilled water (control group, DW), Scope White mouthrinse (SW), Crest 3D White mouthrinse (CWR), Crest 3D White toothpaste (CWT), Crest 3D White toothpaste and Crest 3D White mouthrinse (CWT + CWR), Listerine Whitening toothpaste (LWT), Listerine Whitening mouthrinse (LWR), and Listerine Whitening mouthrinse and Listerine Whitening toothpaste (LWR + LWT). Color measurements were conducted using a spectrophotometer. The data were assessed by analysis of variance for repeated measures and Tukey's multiple comparison test ($p < 0.05$).

Results: Immersion in coffee solution after bleaching caused perceptible staining on tooth specimens ($\Delta E > 3.46$). The whitening effect of CWR on teeth stained after bleaching was significantly greater than that in the other groups ($p < 0.001$). Tooth whitening (ΔE) in each group showed no significant difference from 6 to 12 weeks ($p > 0.05$). The combination of mouthrinse and toothpaste did not increase the degree of tooth whitening.

Conclusion: Whitening mouthrinse and toothpaste had similar effects on the control group in terms of whitening of teeth stained after bleaching. Nevertheless, Crest 3D White mouthrinse produced the greatest recovery whitening effect among all the products tested.

Keywords

Bleaching, carbamide peroxide, tooth whitening

History

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Introduction

Tooth bleaching, a conservative and effective method for the esthetic treatment of discolored teeth, has become an integral component of aesthetic dentistry.[1] Fundamental vital tooth bleaching techniques can be generally classified as at-home (dentist-supervised night-guard bleaching), in-office or power bleaching (professionally administered), and over-the-counter (OTC) or mass market products.[2] The home application procedure involves applying oxygenating gels, usually for a period ranging from 2 to 6 weeks, and usually includes up to 15% hydrogen peroxide (HP) or 22% carbamide peroxide (CP) as active agents.[3,4] Although there are variations in the duration of home bleaching agents, at least 1 h per day for 2 weeks provides a whitening effect of up to 90% in most patients.[5] This method is the most widely used and introduced technique in investigations.[4]

Various factors influencing the efficacy of whitening have been examined, and previous studies have shown that increasing the time of contact with the teeth and the frequency of using bleaching gels develops efficacy in terms of tooth

color change.[6,7] Active concentration of the bleaching agents is another important factor to consider.[1] On the other hand, patient collaboration is also an important factor in the success of whitening treatments, particularly in association with the home bleaching procedure.[8]

Previous studies have reported that bleaching agents can alter the texture and morphology of the enamel surface. There may be a loss of organic components from bleached enamel and dentin surfaces, and these alterations may facilitate the recurrence of extrinsic tooth discoloration.[9,10] It is known that contact with some dietary factors, such as coffee and tea, can lead to significant tooth staining,[11] particularly when there are pores or superficial defects in the dental structure.[12]

Color relapse of bleached teeth is an important issue with different reported results. Some studies have reported a regression of color at the end of the observation period,[13,14] whereas others have reported that tooth color remained stable throughout the observation period.[15,16] Comparing these studies is difficult due to the various study designs, bleaching

agents, and techniques used. While the most important factor is considered to be exposure to staining in clinical trials, there is no exposure to staining in *in vitro* studies, making them less susceptible to color regression after bleaching.[1] Haywood [17] reported a 26% color regression at 18 months with home bleaching, while Matis et al. [18] found a mean color regression rate of 65% after 6 weeks.

Coffee, tea, wine, nicotine and cola-based soft drinks might contribute to the color regression of bleached teeth. In order to prevent color relapse after bleaching treatments, patients have been shown to have better success using a power toothbrush with a whitening toothpaste rather than a manual toothbrush.[19] The whitening effect can be maintained through the use of whitening toothpastes and mouthrinses,[5] but there are no published studies investigating the effectiveness of these whitening agents after bleaching treatment. Thus, this study aimed to investigate the effect of whitening toothpastes and mouthrinses (two whitening dentifrices and three mouthrinses) on the color stability of bleached teeth. The null hypothesis states that the mouthrinses and toothpastes tested have no effect on the color change of stained teeth after bleaching.

Materials and methods

Specimen preparation

Eighty bovine incisors were collected from a slaughterhouse and cleaned. Teeth were immersed in 0.5% chloramines-T solution for 48 h for disinfection. They were then stored in distilled water until they were used within 1 week of extraction. Teeth with spots and fractures were excluded from the study. The tooth crowns were sectioned with a water-cooled diamond disk (Imppect PC10; Equilam Lab. Equip., Diadema, SP, Brazil) in order to obtain 5 × 5 mm enamel–dentin specimens; the thickness of each specimen was standardized at 3.5 ± 0.2 mm, checked with a digital caliper (Mitutoyo, Tokyo, Japan). Using molds, each specimen was individually embedded in transparent acrylic resin to expose the enamel surface. Each specimen was polished for 30 s across the buccal surface, using a prophylaxis paste applied with a polishing brush under manual pressure on a low-speed contra angle, and then washed. All the specimens were stored in distilled water for 4 days before bleaching in order to ensure rehydration.

Bleaching procedure

To simulate the home bleaching treatment, the specimens were bleached with 16% CP (Opalescence PF; Ultradent Products, Inc., South Jordan, UT), according to the manufacturer's instructions, for 14 days. The bleaching gel was applied to each specimen surface in a 2-mm layer, using a dispenser tip for 4 h daily at 37 °C. The bleaching gel was removed from the specimen surface using cotton, after which the specimen was washed with distilled water. Between bleaching sessions, the specimens were maintained at 37 °C in artificial saliva,[20] changed daily. After the specimen surface was covered with bleaching gel, the specimen was placed in a silicon mold filled with distilled water to prevent dehydration. After the bleaching treatment was completed, the specimens were stored in artificial saliva for 24 h and then immersed for 24 h in a

coffee solution prepared with 18 g of coffee (Nescafe 3 in 1; Karacabey, Bursa, Turkey) in 200 mL of boiling distilled water. The specimens were then washed in distilled water.

Group divisions and brushing procedures

The ingredients of the mouthrinses and toothpastes used in this study are presented in Table 1. The specimens were randomly divided into 8 groups of 10 specimens each as follows:

- Group DW (control): The specimens were brushed with distilled water using soft electric toothbrushes[21] in daily mouth cleaning mode (Oral B Triumph; Braun, Kronberg, Germany).
- Group SW: The specimens were brushed with distilled water, as in group DW, and then immersed in a whitening oral rinse (Scope White mouthrinse) for 1 min twice daily for 12 weeks.
- Group LWT: The specimens were brushed with a whitening toothpaste (Listerine Whitening toothpaste). Each brushing cycle was performed with a freshly prepared toothpaste mixture, with one part of toothpaste in three parts of deionized distilled water.[22] The specimens were placed in a silicon mold specially made for each specimen and were then brushed with the prepared toothpaste mixture.
- Group CWT: The procedure was the same as that of group LWT, with a different toothpaste (Crest 3D White toothpaste).
- Group CWR: The procedure was the same as that of group SW, with a different whitening rinse (Crest 3D White Multi-care whitening rinse).
- Group LWR: The procedure was the same as that of group SW, with a different whitening rinse (Listerine Whitening mouthrinse).
- Group CWR + CWT: The specimens were brushed with whitening toothpaste (Crest 3D White toothpaste) and then immersed in whitening oral rinse for 1 min (Crest 3D White Multi-care Whitening rinse), according to the manufacturer's recommendation, following the same procedures as in groups CWT and CWR.
- Group LWR + LWT: The specimens were brushed with whitening toothpaste (Listerine Whitening toothpaste) and then immersed in whitening oral rinse for 1 min (Listerine Whitening mouthrinse), according to manufacturer's recommendation, following the same procedures as in groups LWT and LWR.

In all groups, the brushing procedures were performed twice daily, for 2 min each, for 12 weeks. The toothbrush was fixed on a steel rod with clamp, and brushing was performed with a typical force of 200 g,[23] which was measured with an orthodontic gauge (Correx, Haag-Streit, Koeniz, Switzerland). Different toothbrushes having similar properties were used for each group and changed monthly. Between brushing/immersion procedures, the specimens were immersed in artificial saliva at 37 °C.

Color assessment

Color measurements were performed under D65 illumination with a contact-type digital spectrophotometer (VITA

Table 1. The compositions of whitening mouth rinses, toothpastes and artificial saliva.

Product name (code)	Manufacturer	Compositon
Scope White mouthwash (SW)	Procter & Gamble, Cincinnati, OH	Water, glycerin, alcohol (5%), 1.5% HP, hexametaphosphate, poloxamer 407, sodiumcitrate, flavor, sodium saccharin, citric acid
Crest 3D White toothpaste (CWT)	Procter & Gamble, Cincinnati, OH	Sodium fluoride, water, sorbitol, hydrated silica, disodium pyrophosphate, sodium lauryl sulfate, cellulose gum, sodium hydroxide, sodium saccharin, carbomer, polyethylene, mica, titanium dioxide, blue 1 lake
Crest 3D White multi-care whitening rinse (CWR)	Procter & Gamble, Cincinnati, OH	Water, 1.5% HP, propylene glycol, sodium hexametaphosphate, poloxamer 407, sodium citrate, flavor, sodium saccharin, citric acid
Listerine Whitening mouthrinse (LWR)	Johnson & Johnson Healthcare Products, Skilman NJ	Water, alcohol (8%), HP, tetrapotassium pyrophosphate, pentasodium triphosphate, citric acid, poloxamer 407, flavors, sodium saccharin, sucralose
Listerine Whitening toothpaste (LWT)	Johnson & Johnson Healthcare Products, Skilman NJ	Sodium monofluorophosphate, sorbitol solution, water, hydrated silica, glycerin, PEG-32, sodium lauryl sulfate, cellulose gum, sodium saccharin, eucalyptol, methyl salicylate, thymol, phosphoric acid, menthol, zinc citraete, sodium phosphate, xanthan gum, benzoic acid, flavors, blue 1
Artificial saliva	RTEU Laboratories of Biochemistry	Calcium chloride 0.166 g, sodium benzoate 1 g, cellulose 10 g, magnesium chloride 0.05 g, potassium chloride 0.62 g, sodium chloride 0.025 g, sorbitol 42 g, distilled water 944.52 mL, potassium phosphate 1.1 g

Easyshade Advance; Zahnfabrik, Bad Säckingen, Germany) with a 5-mm diameter probe. Prior to conducting the measurements, the spectrophotometer was calibrated using a calibration plate according to the manufacturer's instruction. The probe was placed 90° to the specimen surface and the unit was activated. Mean L*, a* and b* values of each specimen were obtained after three consecutive measurements.

Color measurements were taken at baseline, after immersion in the coffee solution, and 6 and 12 weeks after brushing or whitening. The color difference at the different time intervals was measured as ΔE from the Commission Internationale de L'Eclairage and calculated as follows:

$$\Delta E = \left[(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2 \right]^{1/2}$$

$$= \left[(L_1 - L_0)^2 + (a_1 - a_0)^2 + (b_1 - b_0)^2 \right]^{1/2},$$

where the subscripts 0 and 1 denote initial and final, respectively.

According to Ghinea et al.,[24] the difference is clinically perceptible when ΔE values are over 3.46 units. For each group, the ΔE , ΔL , Δa and Δb values were calculated using two procedures: (i) bleached versus stained (after immersion in coffee, Table 2) and (ii) stained versus brushing or whitening at Weeks 6 and 12 (Table 3).

Statistical analysis

ΔE , ΔL , Δa and Δb values were statistically analyzed. Parametric tests were performed, as the data were normally distributed on the results of the Kolmogorov–Smirnov test. After staining, the data obtained were analyzed by one-way analysis of variance (ANOVA) to determine whether a significant difference among the groups could be found. ANOVA for repeated measures and Tukey's multiple

Table 2. Means \pm standard deviations of parameters after staining of bleached teeth.

Groups	ΔE	ΔL	Δa	Δb
DW	5.10 \pm 3.31 ^a	-3.64 \pm 2.54 ^a	0.84 \pm 0.71 ^{ab}	2.98 \pm 2.74 ^b
SW	3.99 \pm 1.75 ^a	-3.23 \pm 1.39 ^a	0.42 \pm 0.50 ^{ab}	2.02 \pm 1.50 ^{ab}
LWT	5.31 \pm 2.10 ^a	-3.94 \pm 2.85 ^a	0.91 \pm 1.17 ^b	2.19 \pm 1.47 ^{ab}
CWT	4.77 \pm 1.47 ^a	-4.48 \pm 1.57 ^a	-0.36 \pm 0.66 ^a	0.57 \pm 1.31 ^{ab}
CWR	5.24 \pm 2.06 ^a	-4.43 \pm 2.36 ^a	-0.35 \pm 0.36 ^{ab}	1.74 \pm 1.93 ^{ab}
LWR	4.01 \pm 0.84 ^a	-2.89 \pm 1.33 ^a	0.17 \pm 0.59 ^{ab}	1.71 \pm 1.97 ^{ab}
CWR + CWT	5.06 \pm 1.21 ^a	-4.76 \pm 1.06 ^a	0.29 \pm 0.98 ^{ab}	0.24 \pm 1.52 ^a
LWR + LWT	4.64 \pm 2.02 ^a	-3.60 \pm 2.42 ^a	0.13 \pm 1.46 ^{ab}	2.00 \pm 1.10 ^{ab}

Different superscript letters determine significant differences among groups.

comparison tests were conducted to compare the color of the stained specimens with the color obtained after the brushing or whitening process in the 6th and 12th weeks. The level of significance was set to $p < 0.05$.

Results

The mean color changes (ΔE) and standard deviations after immersion in coffee solution according to L*, a* and b* values after bleaching are listed in Table 2. After immersion, decreased ΔL values and increased Δb and Δa values could be observed. No statistically significant differences were found among groups in terms of ΔE values and ΔL values ($p > 0.05$). Coffee solution after bleaching caused visually perceptible tooth discoloration ($\Delta E > 3.46$).

The statistical evaluation and color change values from the staining period (according to L*, a* and b* values) to the 6th and 12th weeks of the treatment period are presented

Table 3. Means \pm standard deviations of ΔE , ΔL , Δa and Δb values at the time point evaluated after whitening.

Groups	ΔE		ΔL		Δa		Δb	
	6 weeks	12 weeks	6 weeks	12 weeks	6 weeks	12 weeks	6 weeks	12 weeks
DW	3.00 \pm 2.20 ^a	2.48 \pm 1.43 ^a	2.17 \pm 2.29 ^{a+}	-0.39 \pm 2.43 ^{a+}	-0.40 \pm 0.52 ^{ab+}	0.27 \pm 0.50 ^{bcd+}	-0.64 \pm 1.85 ^a	0.22 \pm 1.54 ^{ab}
SW	2.46 \pm 1.69 ^a	3.26 \pm 2.15 ^a	1.41 \pm 2.31 ^a	1.17 \pm 2.52 ^{ab}	-0.02 \pm 0.40 ^{bc}	-0.08 \pm 0.44 ^{bc}	-0.18 \pm 1.34 ^a	-0.23 \pm 2.88 ^{ab}
LWT	3.56 \pm 1.88 ^{ab}	3.45 \pm 2.92 ^a	1.75 \pm 2.85 ^a	2.00 \pm 2.05 ^b	-0.63 \pm 0.80 ^a	-0.51 \pm 0.66 ^a	-0.42 \pm 2.19 ^{a+}	0.65 \pm 4.12 ^{b+}
CWT	3.07 \pm 1.64 ^a	3.08 \pm 1.61 ^a	2.45 \pm 2.15 ^a	2.55 \pm 1.44 ^b	0.31 \pm 0.59 ^c	0.39 \pm 0.63 ^{cd}	0.16 \pm 1.14 ^a	-0.18 \pm 1.81 ^{ab}
CWR	5.17 \pm 2.39 ^b	5.91 \pm 2.81 ^b	4.89 \pm 2.27 ^b	5.42 \pm 2.88 ^c	0.39 \pm 0.32 ^c	0.50 \pm 0.38 ^{cd}	-0.91 \pm 1.60 ^a	-1.66 \pm 1.51 ^a
LWR	2.81 \pm 1.06 ^a	3.14 \pm 1.07 ^a	2.19 \pm 1.36 ^a	2.19 \pm 1.51 ^b	0.29 \pm 0.35 ^c	0.61 \pm 0.35 ^d	-0.65 \pm 1.40 ^a	-0.99 \pm 1.70 ^a
CWR + CWT	3.08 \pm 1.53 ^a	3.61 \pm 1.32 ^a	2.50 \pm 1.39 ^a	2.90 \pm 1.29 ^b	0.10 \pm 0.32 ^{bc}	-0.21 \pm 0.76 ^{ab}	-1.16 \pm 1.55 ^a	-1.56 \pm 1.38 ^a
LWR + LWT	2.89 \pm 1.95 ^a	2.85 \pm 1.83 ^a	2.35 \pm 2.23 ^a	2.29 \pm 2.63 ^b	-0.29 \pm 0.89 ^{ab}	0.18 \pm 0.79 ^{ab}	-0.63 \pm 0.77 ^a	0.58 \pm 1.41 ^{ab}

For a given time, different superscript lower letters indicate significant difference among groups.

⁺Statistical significant difference between 6 and 12weeks for each color parameter ($p < 0.05$).

in Table 3. After 6 and 12 weeks, the CWR group was statistically different from the other groups in terms of color change (ΔE), except the ΔE of LWT for 6 weeks. All groups, with the exception of CWR, showed similar color changes to those of the control group at the time point evaluated. The results of the repeated measures of ANOVA showed no significant difference in the color change (ΔE) between 6 and 12 weeks for all groups ($p > 0.05$).

After 6 and 12 weeks of treatment, the ΔL values in the CWR group were significantly different from those in the other groups. After 12 weeks of treatment, the ΔL values in only the DW group decreased significantly compared with the values after 6 weeks ($p = 0.001$). The Δa values in the DW group increased significantly between 6 and 12 weeks of treatments ($p = 0.03$). At 6 weeks of treatment, no statistical significant differences were found in the Δb values among the groups ($p = 0.22$). After 6 weeks, the increase in Δb was statistically significant for the LWT group compared with that in 12 weeks of treatment ($p = 0.025$).

Discussion

This study evaluated the effectiveness of whitening dentifrices and mouthrinses containing HP on the color stability of bleached teeth. The whitening of specimens in the CWR group was significantly different from that in the other groups. Therefore, the null hypothesis that mouthrinses and toothpastes had no effect on the color stability of bleached teeth was rejected.

Coffee, tea, red wine and cola-based soft drinks have been used to create staining in several *in vitro* studies. In the present study, a 24-h coffee immersion was chosen, as the best staining with this time was reported in a previous study.[25]

The surfaces of the specimens were not flattened prior to the experiment in order to simulate natural conditions. This situation might have led to greater variation among the specimens in terms of adsorption of color pigments and measurement of the color, due to irregularities in the surface textures of the specimens.[26] On the other hand, removing the superficial layer of specimens can make them more sensitive to staining after bleaching treatment, as the aprismatic layer is more resistant to staining than subsurface enamel.[27] Furthermore, it would decrease the thickness of the enamel layer and impact the penetrability of the hard tissue of the teeth. The susceptibility of the specimens to

coffee was different among the groups, but not statistically ($p = 0.80$).

Clinically applicable digital devices for color measurement have been advertised to obtain objective color values. Spectrophotometers measure the amount and spectral composition of reflected or transmitted light. Reflection or transmission graphs must then be converted into tristimulus data.[28] They present the color data on various parameters, including the CIELab color system, which is currently the most widely used system in dental studies. The advantage of this system is related to the evaluation of the color change from the variation of the coordinates L^* , a^* and b^* .[4] A previous study reported that Easysshade Advance, particularly used in this study, can be used to evaluate tooth color changes with good reliability.[28]

Li et al. [29] noted that most color regression was stimulated by the L^* -value, but Meireles et al. [30] observed that a^* and b^* values increased in a group treated with 16% CP when compared to the end of the treatment. In our study, the ΔL values were higher than the Δa and Δb values after staining, brushing, and immersion. After staining, the Δa values of the CWT and CWR groups decreased depending on the remaining oxygen radicals in the tooth structures, causing further whitening. After 6 and 12 weeks of treatments, the ΔL values in some groups decreased while the Δa and Δb values increased, depending on the color regression.

The whitening of specimens in all the groups increased with the removal of extrinsic surface stains during 6 weeks of brushing or whitening after the staining period. The three mouthrinses used in this study contain HP, which cause a breakdown of intrinsic stains, as well as the pigments responsible for color alteration.[31] However, we obtained different results with the mouthrinses. The variance among brands with regard to color change may be explained by the different product compositions.

The color changes in the DW, LWT and LWT + LWR groups slightly decreased from the 6th to the 12th week after treatment. This decrease could be due to the previously oxidized substances that became chemically reduced and to the abrasive properties of toothpaste. Haywood [17] reported that whitening toothpastes have more abrasive ingredients than conventional toothpastes; therefore, vigorous brushing with whitening toothpastes not only facilitates stain removal but may also promote excessive wear.

The stain removal ability of dentifrices is due to the large quantity of abrasives in their ingredients, which remove and control superficial extrinsic stains. The active ingredients of whitening dentifrices contain agents that break down the organic molecules of biological film; they rarely contain CP or HP. Abrasives such as alumina, dicalcium phosphate dehydrate and silica are also present in the formulation. Hydrated silica has great cleaning ability and can remove stains.[32] The abrasiveness of a dentifrice depends not only on the inherent hardness of the particles, but also on the particle size and shape of the abrasive components and the pH of the dentifrice. High amounts of abrasives in dentifrices, however, can damage hard and soft tissues and dental restorations. Therefore, their abrasiveness must be controlled.[33] Ozkan et al.[34] reported that when bleaching treatment with 10% CP and 10% HP was combined with dentifrices, the roughness of the enamel surface increased. After bleaching, the use of mouthrinse and toothpaste may increase surface roughness or irregularities; as a result, the whiteness of teeth may decrease over time depending on the use of these products.

Mouthrinses have become a very popular OTC whitening agent because of the ease of application, low cost, and wide availability.[35] These agents generally include low concentrations of HP and sometimes, sodium hexametaphosphate, in order to remove stains from tooth surfaces. A previous study reported that different peroxide-based whitening rinses did not have a bleaching effect on stained teeth.[36] In our study, after 12 weeks of treatment, the color changes in the LWR and SC groups showed no statistical difference from those in the control group.

The continuous exposure of teeth to acidic products may result in a number of complications, such as tooth sensitivity in areas of exposed dentin and incorporation of pigments in the tooth structure, with consequent discoloration.[31] Because there is a concern regarding the possible tumor-promoting ability of HP with tobacco carcinogens, patients should avoid alcohol and smoking during the whitening treatment.[37] Nevertheless, HP is present in low concentrations in mouthrinses used in this study, which would no damage the mucosa.[38] Indeed, a previous study suggested that products containing HP produce no damage to oral hard and soft tissues and no significant risk of adverse long-term effects.[39]

Conclusions

With the findings of this *in vitro* study, it can be concluded that:

- (1) After 12 weeks, the color change in the CWR group was significantly higher than that in all the other groups.
- (2) For all the groups, the results of whitening in teeth stained after bleaching were similar to those for the control group at the time point evaluated, except for the CWR group.
- (3) The results of the study revealed no significant difference in color change (ΔE) between 6 and 12 weeks for all the groups.
- (4) Further clinical studies are needed to prove the safety and longevity of these products.

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