Braz Dent J (2005) 16(2): 103-106

ISSN 0103-6440

Effect of a Carbamide Peroxide Bleaching Gel Containing Calcium or Fluoride on Human Enamel Surface Microhardness

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This *in vitro* study evaluated the surface microhardness of human enamel submitted to bleaching with 10% carbamide peroxide (CP) containing calcium or fluoride. Ninety-eight dental blocks (5 x 5 mm^2) with polished enamel surfaces were randomly assigned to 7 treatment groups (n=14), as follows: without bleaching and storage in artificial saliva (control); 10% CP; 10% CP + 0.05% calcium; 10% CP + 0.1% calcium; 10% CP + 0.2% calcium; 10% CP + 0.2% fluoride; and 10% CP + 0.5% fluoride. During 14 days, enamel surfaces were daily exposed to a 6-h bleaching regimen followed by storage in artificial saliva. Surface microhardness was measured before (baseline), during (7th day), immediately after bleaching (14th day) and 1 week post bleaching. Data were analyzed by two-way ANOVA and Tukey's test ($p<0.05$). All treatments reduced SM significantly during the bleaching cycle (7th day), immediately after bleaching (14th day) and 1 week post bleaching, compared to baseline and to the unbleached control group. In conclusion, in spite of the addition of calcium and fluoride, all bleaching treatments affected the enamel surface microhardness.

Key Words: bleaching, enamel, microhardness, fluoride, calcium.

INTRODUCTION

The use of self-administered carbamide peroxide (CP)-containing bleaching gels at home has become increasingly popular for whitening of stained and discolored teeth, since their introduction by Haywood and Heymann (1). However, the effect of bleaching procedures on dental enamel is not yet clear and is still a controversial subject.

Studies have reported neither significant adverse effects of bleaching agents on tooth surface morphology using scanning electron microscopy, nor significant changes in hardness on subsurface levels of human enamel (2-4). Other investigations have demonstrated minor morphological changes on enamel treated with 10% carbamide peroxide (5). However, it has been reported that 10% carbamide peroxide bleaching may decrease calcium and phosphate enamel content, as well as its fluoride concentration (6).

These effects may possibly be overcome by addition of calcium and fluoride to bleaching agents' compositions. Fluoride and calcium ions would increase the bleaching gel saturation and decrease mineral loss during bleaching action and exchange of ions. It is generally accepted that the major effects of fluoride ions in enhancing crystal growth and retarding dissolution of dental enamel minerals are derived from their role in the solution phase, increasing supersaturation or decreasing undersaturation (7).

The purpose of this study was to test the null hypothesis that the enamel microhardness is not altered when exposed to carbamide peroxide bleaching gels containing calcium or fluoride.

MATERIAL AND METHODS

This research proposal was reviewed by the Research Ethics Committee of the Faculty of Dentistry

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of Piracicaba, State University of Campinas, and the designed methodology was approved.

Ninety-eight human enamel blocks were obtained from erupted third molars stored in 0.02% thymol solution for at least 1 month. The enamel blocks (5 x 5 mm^2) were individually embedded in self-curing polystyrene resin cylinders leaving the enamel surface exposed for further polishing. Initial surface microhardness was determined to select the blocks and compare them after treatment. The blocks were randomly assigned to 7 carbamide peroxide bleaching treatment groups (n=14), as follows: without treatment (control); 10% CP; 10% CP + 0.05% calcium; 10% CP + 0.1% calcium; 10% CP + 0.2% calcium; 10% CP + 0.2% fluoride (NaF); and 10% CP + 0.5% fluoride (NaF). The blocks were submitted to the bleaching treatment for 14 days. Surface microhardness was measured during the bleaching cycles (7th day), immediately after bleaching (14th day) and 1 week post bleaching.

The enamel blocks from the experimental groups were exposed to one daily application of the bleaching agents for 6 h during 14 consecutive days. All carbamide peroxide gels had neutral pH. In each specimen, 0.1 mL of bleaching agent and 0.05 mL of artificial saliva were applied onto enamel surface and covered with an individual tray (8). During bleaching, the enamel blocks were maintained in 100% relative humidity at 37°C and after each daily treatment, the specimens were thoroughly rinsed with air/water spray for 10 s and stored in artificial saliva at 37°C (9). Once the 14-day period was completed, specimens were stored in artificial saliva for additional 7 days. The control group did not receive any treatment and was stored in artificial saliva at 37°C. The artificial saliva was changed every day.

Surface microhardness was tested before the bleaching treatments (baseline), at the 7th and 14th day of treatment and at the 7th day post bleaching. Three microhardness indentations were done on enamel surface with a microhardness tester (Future Tech-FM, Tokyo, Japan) and Knoop indentator using a 25-g load for 5 s. Means of the three indentations were calculated.

Data were analyzed statistically by two-way (treatment and time factors) analysis of variance, according to a split-plot design, and Tukey's test at 5% level of significance.

RESULTS

ANOVA revealed significant differences for treatments ($p<0.0001$), time ($p<0.0001$) and the interaction between factors ($p<0.0001$). Table 1 shows the effect of bleaching treatments and time on enamel surface microhardness. No significant differences ($p>0.05$) were observed among the groups before the bleaching treatments (baseline). Baseline data differed statistically from those of the other periods (7th, 14th and 21st days), except for the untreated control group. Statistically significant differences were also found between the untreated control group and the bleached groups at all evaluation periods, except for baseline.

DISCUSSION

The widespread use of bleaching agents for esthetic purposes has made necessary the choice for materials that promote minimal effects on enamel

Table 1. Surface microhardness according to the treatments and time (mean ± SD).

Treatment	Baseline	7th day	14th day	21st day (1 wk post-bleaching)
Control	259.9 ± 38.4 Aa	283.8 ± 39.1 Aa	268.9 ± 37.8 Aa	279.8 ± 42.4 Aa
10% CP	266.4 ± 42.3 Aa	114.5 ± 49.8 Bb	127.6 ± 52.7 Bb	130.5 ± 42.2 Bb
10% CP + 0.05% Ca^{++}	268.2 ± 39.2 Aa	111.9 ± 55.8 Bb	112.2 ± 46.2 Bb	117.9 ± 48.2 Bb
10% CP + 0.1% Ca^{++}	284.2 ± 40.7 Aa	139.3 ± 66.0 Bb	150.0 ± 70.2 Bb	145.4 ± 79.9 Bb
10% CP + 0.2% Ca^{++}	246.7 ± 41.8 Aa	118.1 ± 51.6 Bb	125.2 ± 51.0 Bb	119.4 ± 47.7 Bb
10% CP + 0.2% F^-	261.7 ± 42.0 Aa	119.7 ± 45.1 Bb	136.9 ± 54.9 Bb	134.2 ± 53.7 Bb
10% CP + 0.5% F^-	246.1 ± 37.9 Aa	120.3 ± 39.3 Bb	133.8 ± 35.8 Bb	137.7 ± 44.0 Bb

Means followed by distinct letters differ statistically at 5%. Uppercase letters show significant differences among treatments (columns). Lowercase letters show significant differences among times (lines). CP = carbamide peroxide.

surface. Studies have shown that carbamide peroxide is able to make changes on enamel surface texture, such as topographical alterations, decalcification and porosities (5,10,11). Although it has been shown that CP causes local microstructural and chemical changes, such as loss of calcium and phosphorus ions, enamel alterations are likely not significant clinically (6,12). It has been reported (13) that 10% carbamide peroxide bleaching had minimal effect on enamel surface morphology and that the effects did not worsen over time.

A consensus has not yet been reached about the effect of CP on enamel. Studies have agreed that this bleaching agent actually promotes changes on enamel surface, but these alterations can be either clinically significant or not. The addition of fluoride or calcium to bleaching gels could be a possible alternative to overcome the adverse effects of carbamide peroxide on enamel surface.

The results of this study showed that 10% carbamide peroxide bleaching produced changes on enamel surface, when the baseline microhardness values were compared to those recorded at the 7th and 14th days of treatment (Table 1). Smith et al. (14) also observed a decrease of initial microhardness after 10% carbamide peroxide treatments. Other studies found a decrease of microhardness of enamel exposed to a 10% carbamide peroxide whitening gel (Rembrandt, Oral-B), from 7th to 14th days and after four weeks (5,15).

Moreover, the addition of fluoride and calcium did not prevent enamel mineral loss at any concentration. However, this does not mean that these ions had no effect on enamel, although they were expected to have greater contribution in preventing mineral loss. The major importance of fluoride is to be present at low and constant concentrations in order to inhibit mineral loss and activate remineralization (16,17). It is likely that the presence of fluoride ions inhibited greater demineralization.

According to Burgmaier et al. (18), who evaluated fluoride uptake in bleached enamel, it is not feasible to improve fluoride uptake in enamel pretreated with 10% carbamide peroxide. While the bleaching substance is able to diffuse into the enamel, it can dissolve the newly bound fluoride ions. In this study, however, the ions were added to the bleaching gels and, at the same time, the hydrogen peroxide and fluoride or calcium could diffuse in enamel. Although the added ions were not able to avoid demineralization, the minerals might have inhibited the progression of demineralization. This can be confirmed by analyzing the effect of whitening gels containing fluoride or calcium on enamel during the cariogenic challenge.

The lack of effect of saliva on remineralization of enamel surface after bleaching treatments may possibly be attributed to the fact that the surface microhardness was evaluated in a short post-bleaching period (7 days). Freitas et al. (19) reported recovery of dentin mineral loss 14 days after the bleaching regimen.

Although tooth sensitivity has been described as a slight and transient discomfort, it is the most common side effect reported by patients during the bleaching treatment (1). Fluoride and calcium ions have also been used in some formulations as active ingredients of desensitizing agents to manage hypersensitive dentin (20). Therefore, another important role of the addition of these ions would be related to their effect on tooth sensitivity during tooth bleaching.

In conclusion, the findings of this study rejected the null hypothesis and showed that, in spite of the addition of calcium and fluoride ions, all bleaching treatments affected negatively the enamel surface microhardness. Carbamide peroxide bleaching gel containing fluoride or calcium did not avoid mineral loss resulting from the whitening procedures. However, the addition of these ions can be beneficial on prevention of greater mineral change.

RESUMO

Este estudo *in vitro* avaliou microdureza superficial do esmalte após o clareamento com peróxido de carbamida a 10% (PC) contendo cálcio ou flúor. Noventa e oito blocos dentais (5 x 5 mm^2) com a superfície do esmalte polido foram divididos em sete grupos experimentais (n=14): sem clareamento e armazenado em saliva artificial; 10% (PC); (3) 10% PC + 0,05% cálcio; 10% PC + 0,1% cálcio; 10% PC + 0,2% cálcio; 10% PC + 0,2% flúor; e 10% PC + 0,5% flúor. Os géis clareadores foram aplicados por 6 h durante 14 dias e após cada dia de tratamento, os espécimes foram armazenados em saliva artificial. A microdureza superficial foi mensurada antes (baseline), durante (7º dia), imediatamente após o clareamento (14º dia) e 1 semana após o fim do tratamento clareador. Os dados foram analisados pela ANOVA (2 fatores) e teste de Tukey (p<0.05). Os tratamentos clareadores reduziram significativamente a microdureza durante (7º dia), imediatamente após o clareamento (14º dia) e 1 semana após o seu término, quando comparados aos valores iniciais (baseline) e aos do grupo controle. Os achados deste estudo sugerem que, a despeito da adição de cálcio e flúor, todos os géis clareadores afetaram a microdureza superficial do esmalte.

ACKNOWLEDGEMENTS

The bleaching materials used in this study are not commercially available and were supplied by FGM Produtos Odontológicos. This study was supported by grant from CAPES, Brazil.

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Accepted February 4, 2004