

# Nonprescription Bleaching versus Home Bleaching with Professional Prescriptions: Which One is Safer? A Comprehensive Review of Color Changes and Their Side Effects on Human Enamel

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## Abstract

**Objectives** This study evaluates the efficacy and safety of the professionally prescribed and nonprescription over-the-counter (OTC) bleaching agents.

**Materials and Methods** Extracted human upper central incisors were prepared and stained with red wine for 14 days before being subjected to four different bleaching agents: professionally prescribed opalescence PF 15%, VOCO Perfect Bleach 10%, nonprescription OTC Crest 3D Whitestrips, and Whitelight Teeth Whitening System. Colorimetric measurement was performed with Vita Easyshade Handheld Spectrophotometer, enamel surface microhardness measured using Vickers Hardness machine, and surface roughness was evaluated with profilometer, before and after bleaching. Scanning electron microscope (SEM) evaluation and atomic force microscopy were conducted postbleaching.

**Statistical Analysis** The data were analyzed with *t*-test, two-way ANOVA, one-way ANOVA, and Turkey's test at a significance level of 5%.

**Results** All bleaching products have the same efficacy to whiten stained enamel. Opalescence PF 15% showed significant increase in the microhardness ( $92.69 \pm 68.316$ ). All groups demonstrated significant increase in surface roughness ( $p < 0.05$ ). SEM evaluation showed that Opalescence PF 15% resulted in same microscopic appearance as unbleached enamel, while VOCO Perfect Bleach 10%, Whitelight Teeth Whitening System and Crest 3D Whitestrips demonstrated mild to moderate irregularities and accentuated irregularities, respectively.

**Conclusion** Professionally prescribed bleaching agent of Opalescence PF 15% is effective in whitening the teeth, while the other bleaching products may be effective but also have deleterious effects on the enamel.

## Keywords

- ▶ bleaching
- ▶ home bleaching with professional prescriptions
- ▶ nonprescription bleaching
- ▶ efficacy of bleaching
- ▶ surface roughness of enamel
- ▶ microhardness of enamel

## Introduction

In recent years, with the increasing trend and desire for whiter teeth to enhance aesthetic appearance, tooth bleaching is becoming a popular choice of treatment among patients.<sup>1</sup> Current tooth bleaching regime ranges from professionally

applied in-office bleaching, professionally prescribed (PB) home bleaching, nonprescription over-the-counter (OTC) bleaching to the do-it-yourself (DIY) regime.<sup>2-4</sup>

Bleaching treatment is considered to be the most conservative, safe, effective, and minimally invasive method to

overcome external and internal tooth discoloration.<sup>5</sup> Professionally prescribed home bleaching agent has been proven to be efficient to whiten discolored teeth.<sup>6</sup> Professionally prescribed home bleaching is closely monitored by the dentist, thereby ensuring their safety and increasing the effectiveness of the bleaching procedure.<sup>2,7</sup>

The availability and easy access to non-prescription (OTC) bleaching products have increased their popularity. This regime is less time consuming, low cost, and avoids the need for an extra appointment with the dentist as compared with professionally prescribed home bleaching agent.<sup>8</sup> However, the safety issues of these bleaching agents remain controversial as the U.S. Food and Drug Administration (FDA) does not verify certain products. Unsupervised bleaching with nonprescription OTC bleaching agent may have potentially deleterious effects on general health and specifically dental health.<sup>2,7,9</sup>

Several studies were conducted to compare the effectiveness of nonprescription OTC bleaching agent, professionally prescribed home bleaching, in-office agent, and DIY product.<sup>4,10</sup> Studies carried out by Kwon et al and also Mielzareck et al found no significant difference in the enamel surface microhardness after bleaching with Opalescence PF which contains fluoride.<sup>4</sup> Based on manufacturer disclaimer, addition of fluoride in the Opalescence PF bleaching formulation ensures that enamel microhardness is enhanced and not affected. The study by Kwon et al found no significant difference in enamel microhardness, but Heshmat et al found that the incorporation of remineralizing agent in the bleaching product may enhance enamel microhardness.<sup>11</sup>

A study conducted by Kwon et al also observed no changes in the enamel surface roughness after being subject to professionally prescribed or nonprescription OTC bleaching agent.<sup>4</sup> In contrast, some other researchers found an increase in the enamel surface roughness after bleaching.<sup>12</sup> A study by Cavalli et al found a significant increase in the enamel surface roughness after being subjected to a high concentration of bleaching agent.

A clinical study comparing three different bleaching regimes found that bleaching efficacy correlates with the types of bleaching regime, in which in-office bleaching was found to be the most effective with minimal time required to whiten the teeth followed by professionally prescribed home bleaching and nonprescription OTC bleaching.<sup>13</sup> Subjective assessment of color changes of the teeth VITA shade guide was used in this study

This study was performed to make sense of the confusing findings pertaining to the effectiveness and safety of non-prescription OTC bleaching compared with the professionally prescribed home bleaching. The purpose of this study was to evaluate the efficacy and safety of the professionally prescribed home bleaching and the nonprescription OTC bleaching agents on the color changes, microhardness, surface roughness, and the surface morphology of the human natural tooth, with the null hypothesis that there were no significant differences between the two bleaching procedures.

## Materials and Methods

The conduct of this study fully conformed to the local practices, laws, and regulations. Ethical clearance was obtained from the institutional Human Research Ethic Committee, with the study protocol code JEPeM/15020047. Sample size calculation was carried out using PS software version 3.1.2 (Dupont and Plummer, 1998), with Type 1 error probability ( $\alpha$ ) of 0.05 and power of the study set at 80 for all parameters. For colorimetric evaluation, with mean difference ( $\delta$ ) of 7.4 and standard deviation ( $\sigma$ ) of 0.8,<sup>6</sup> two samples were required. For surface roughness, with mean difference ( $\delta$ ) of 0.08 Ra and standard deviation ( $\sigma$ ) of 0.05 Ra,<sup>14</sup> seven samples were needed, whereas for surface hardness, with mean difference ( $\delta$ ) of 19 VHN and standard deviation of 29 VHN,<sup>10</sup> 11 samples were needed. The highest sample size was used, and after considering 10 percent damage to the samples, 13 samples were needed for each group, with the total of 65.

Sixty-five intact, extracted human permanent maxillary central incisors were then collected and screened for surface cracks, caries, and fracture under magnification. The roots were then cut 1 mm below the cemento-enamel junction using hard tissue cutter (Exakt Apparatebau; 22851 Nordstedt, Germany). The buccal surface of the crown was cleaned with pumice and then flattened using a model trimmer (MAESTRA; 48150 Sondika Bilbao, Spain) to obtain an even surface of 14 × 8 mm ( $\pm 1$  mm). The coronal tooth structure was embedded in self-cured acrylic resin (Vertex, The Netherlands) with the flat surface facing the mold base. The exposed surface was polished using sandpaper (WS-FLEX 18, HERMES) with straight handpiece set at the same speed in increasing grit of 250, 500, and 2400  $\mu$ m.

The exposed enamel surface of all samples was etched with 35% phosphoric acid gel for 30 seconds and 15 seconds, respectively, and then stained with red wine (Chateau Du'Pond, Merlot 2008) in test tube for 14 days. The enamel samples were stored in humid environment for 7 days.<sup>15</sup>

Baseline colorimetric measurements were carried out on all 65 samples using Vita Easy Shade Advance 4.0 (VITA Zahnfabrik; H. Rauter GmbH & Co.KG, Germany). The samples were placed on a white paper to avoid disturbance in spectrophotometer reading from the surrounding. The probe tip was placed in the center perpendicular and in contact with the tooth surface. Then, the displayed results were recorded. The measurement recorded  $L^*$ ,  $a^*$ ,  $b^*$ , where  $L^*$  stands for luminosity dimensions or whiteness, ranging from 0 (pure black) to 100 (reference white),  $a^*$  for green-red contrast ( $-a^*$  = green and  $+a^*$  = red), and  $b^*$  for blue-yellow contrast ( $-b^*$  = blue and  $+b^*$  = yellow). Color change ( $\Delta E$ ) was calculated using formula:  $\Delta E = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$ . Positive  $\Delta L^*$  means the samples became whiter, while negative  $\Delta L^*$  means samples became darker.<sup>14,15</sup> The measurement was taken three times for each sample.

Baseline surface hardness measurement was conducted using microhardness tester (Vickers Hardness Tester, VM-50, Fuel Instrument & Engineers Pvt. Ltd. Kolhapur, Maharashtra

State, INDIA) for Vickers Hardness Number (VHN) with the use of sets of 100 g load and 20 seconds dwell time at room temperature (26°C). Three indentations on the top surface, not closer than 1 mm to the adjacent indentations or margin of the specimens, were made. The diamond indentation was read on the viewer and VHN was calculated.

Baseline surface roughness of all samples were measured using Profilometer Surf-Corder SE 1200 (Kosaka Laboratory Ltd., Tokyo, Japan) with measuring range of 520 µm vertically, 25 mm horizontally, vertical resolution of 0.008 µm, and measuring speed of 0.2 and 0.5 mm/s. Three different areas were selected on the tooth specimen randomly and measurements in different directions were recorded to evaluate the surface roughness value (Ra).

A pH meter (Hanna Instrument, pH 210 Microprocessor pH meter) was calibrated with the buffered potassium chloride (KCl) solutions (pH 4.01, 7.01 and 10.1). Two commercially and professionally prescribed (PB) bleaching agents, Opalescence PF 15 Ultradent Product Inc., South Jordan, Utah, USA) and VOCO Perfect Bleach, and two commercial, nonprescription OTC bleaching products, Whitelight Tooth Whitening Set and Crest 3D White (► **Table 1**), were placed in test tubes. The pH electrode was then immersed in each bleaching gel to allow uniform contact with the electrode tip for 20 minutes at room temperature (24°C) for pH measurement. Calibration and cleaning procedure of the electrode was performed before the pH measurement of each bleaching agent.<sup>16</sup>

All 65 tooth specimens were then randomly divided into five groups ( $n = 13$ ) according to bleaching procedures. For control group, no bleaching agent was applied. For PB Opal group (Opalescence PF 15%), PB Voco group (VOCO Perfect Bleach 10% gel), and nonprescription OTC WL group (Whitelight Tooth Whitening Set gel), the bleaching agents were applied on tooth specimens approximately 1 mm thick for 4 to 6 hours (PB Opal and PB Voco) and kept in humid atmosphere at 37°C. For nonprescription OTC WL, light activation was carried out using manufacturers' light source for 30 minutes. Thereafter, all bleaching gel was washed using plain water, and each sample was stored in distilled water. In the nonprescription OTC Crest group, the enamel surface of the specimens was covered with the bleaching strip (Crest 3D White Strip) for 30 minutes in humid atmosphere at 37°C. Then, the strip was removed, and the specimens were then stored in distilled water for 7 hours between treatments. Bleaching procedure for all group of bleaching agents was

conducted for 10 days consecutively. Bleaching regime for each group summarized in the ► **Table 1**.

All specimens were again subjected to colorimetric, microhardness, and surface roughness measurement after bleaching procedure.

Five samples, one from each group were randomly selected, gently air dried, dehydrated with alcohol, and gold coated under vacuum pressure using gold coating machine (LEICA EM SCD005) and then analyzed under scanning electron microscope (SEM; scanning electron microscope, Quanta FEG 459). Forty photomicrographs of serial SEM of the surface of each specimen at 5,000× and 10,000× from original magnification were obtained. The superficial morphology of enamel was examined and scores were recorded as follows: 0, enamel with smooth surface morphology; 1, enamel with slight irregularities; 2, enamel with moderate irregularities; 3, enamel with accentuated irregularities with loss of superficial structures.<sup>17</sup> The photomicrographs of SEM were evaluated in a double-blinded manner by two calibrated examiners. Intra- and inter-examiner agreements were tested using Cohen's Kappa test.<sup>18</sup>

Five samples, one for each group, were selected for atomic force microscopy (AFM) surface topography evaluation. This was done based on mean surface roughness (Ra) obtained from surface roughness evaluation by profilometer. All teeth specimens were analyzed at room temperature in dehydrated state. Images were obtained with a resolution of 512 X 512 pixels per image. Images 100 µm × 100 µm were taken with the AFM software Nanoscope IIIA (Software version 5.12r3, Veeco - Digital Instruments, Santa Barbara, CA) for each specimen. To quantify microroughness, the following methodology was adopted: first, conventional statistical roughness parameters, including Ra that is the arithmetic mean deviation of the surface; RMS that is the root-mean-square deviation of the surface; and Z range that defines the height between the highest peak and the deepest valley was determined.<sup>19,20</sup>

## Statistical Analysis

Data collected were analyzed using SPSS version 22.0. Color changes, microhardness, and surface roughness pre and post-bleaching were analyzed using paired *t*-test. Analysis of variance for repeated measurements (two-way ANOVA) was used to compare the color changes ( $\Delta E$ ), surface microhardness,

**Table 1** Composition, pH, and manufacturer regime of the bleaching protocols

Groups	Concentration and pH	pH	Application regimen
Control		7	Immersed in distilled water
PB Opal	15% CP	6.38	10 days application, 4–6 hours daily
PB Voco	16% CP	5.95	10 days application, 4–6 hours daily
OTC Crest	Not declared by manufacturer	4.60	10 days application, 30 minutes daily
OTC WL	Not declared by manufacturer	5.97	10 days application, 30 minutes with application of light activator

Abbreviations: OTC Crest, Crest 3D whitestrip; OTC WL, Whitelight tooth whitening set; PB Opal, Opalescence PF 15%; PB Voco, Voco perfect bleach 10%.

and surface roughness (Ra) between five groups. Mean value was statistically analyzed by one-way analysis of variance (ANOVA) and Turkey's test at the 5% level of significance.<sup>18,21</sup> The *p*-value set as significant at *p* < 0.05.

## Results

Paired *t*-test revealed that there were significant changes in the L\*, a\* values postbleaching in both control and all bleaching groups. Significant changes of the b\* values were only observed in the PB Voco and OTC Crest groups (►Table 2). One-way ANOVA test showed that there was a significant difference in the color changes when the comparison was made between groups with *p* < 0.001. Further posthoc Tukey analysis revealed that there were significant differences between all groups except for PB Opal and PB Voco, and PB Opal and OTC Crest with *p* = 0.682 and 0.052, respectively (►Table 3). PB Voco was the most efficient in bleaching the teeth, followed by PB Opal, OTC Crest, and OTC WL.

Paired *t*-test showed that there was a significant increase in the microhardness of enamel surface postbleaching only in PB Opal group with *p* < 0.001 (►Table 4). However, one-way

ANOVA test showed no significant difference in the microhardness changes pre and postbleaching between groups (*p* = 0.076)

Paired *t*-test showed significant increase in the surface roughness of the enamel postbleaching in all test groups with *p* < 0.001, respectively (►Table 5). One-way ANOVA test showed significant difference in the surface roughness changes after bleaching procedure between groups with *p* < 0.001. Further analysis with posthoc Tukey test revealed that there were significant differences between all groups except PB Opal and PB Voco, and PB Opal and OTC WL groups (►Table 6). The highest mean in the differences of the surface roughness was observed in OTC Crest 3D group followed by OTC WL, PB Opal, and PB Voco groups. These results were further supported by SEM photomicrographs illustrated in ►Fig. 1.

►Fig. 2 shows distribution of enamel surface morphology scores for each group under SEM investigation. The agreement achieved, *K* = 0.6125, indicated substantial agreement among the examiners.<sup>18</sup> Control group exhibits 50% smooth surface morphology mixed with slight and moderate surface irregularities. Both PB Opal and PB Voco group showed more than 75% slight irregularities. However, accentuated surface irregularities have been observed in PB Voco

**Table 2** Comparison of mean (SD) between pre- and postbleaching colorimetric assessment L\*, a\* and b\* values of enamel for all groups

Groups	n	Variables	Prebleaching mean (SD)	Postbleaching mean (SD)	Mean difference (95% CI)	T statistics (df)	p-Value
Control	13	L	37.32 (± 17.803)	34.26 (± 16.542)	3.05 (0.096, 6.011)	2.250 (12)	<b>0.044*</b>
		a	14.69 (± 2.754)	8.55 (± 3.109)	6.15 (3.100, 9.192)	4.396 (12)	<b>0.001*</b>
		b	17.79 (± 5.399)	15.58 (± 5.566)	2.22 (-0.522, 4.953)	1.763 (12)	0.103
(PB Opal)	13	L	43.05 (± 13.907)	82.42 (± 8.418)	-39.36 (-47.673, -31.050)	-10.319 (12)	<b>&lt; 0.001*</b>
		a	15.25 (± 2.824)	-0.01 (± 1.405)	15.25 (13.331, 17.177)	17.283 (12)	<b>&lt; 0.001*</b>
		b	22.88 (± 6.699)	21.55 (± 4.339)	1.34 (-2.064, 4.741)	0.857 (12)	0.408
PB Voco	13	L	37.00 (± 9.862)	77.82 (± 6.654)	-40.82 (-47.747, -33.899)	-12.847 (12)	<b>&lt; 0.001*</b>
		a	16.37 (± 1.677)	1.76 (± 1.652)	14.61 (13.051, 16.165)	20.443 (12)	<b>&lt; 0.001*</b>
		b	21.32 (± 6.904)	26.26 (± 5.493)	-4.94 (-9.200, -0.676)	-2.525 (12)	<b>0.027*</b>
OTC Crest	13	L	36.24 (± 9.800)	66.99 (± 7.615)	-30.75 (-36.029, -25.479)	-12.703 (12)	<b>&lt; 0.001*</b>
		a	16.62 (± 2.655)	2.23 (± 1.453)	13.38 (11.831, 14.938)	18.769 (12)	<b>&lt; 0.001*</b>
		b	19.04 (± 6.103)	24.17 (± 3.575)	-5.13 (-10.084, -0.178)	-2.257 (12)	<b>0.043*</b>
OTC WL	13	L	39.19 (± 14.106)	51.48 (± 7.743)	-12.29 (-21.526, -3.058)	-2.900 (12)	<b>0.013*</b>
		a	14.83 (± 4.241)	6.90 (± 2.941)	7.93 (5.028, 10.833)	5.954 (12)	<b>&lt; 0.001*</b>
		b	18.93 (± 5.762)	22.95 (± 4.497)	-4.02 (-8.106, 0.060)	-2.147 (12)	0.053

Abbreviations: OTC Crest, Crest 3D whitestrip; OTC WL, Whitelight tooth whitening set; PB Opal, Opalescence PF 15%; PB Voco, Voco perfect bleach 10%. Paired *t*-test.

\*Significant at *p* < 0.05.

**Table 3** Comparison of mean (SD) color changes ( $\Delta E$ ) postbleaching between different groups

Groups	n	$\Delta E$ Mean (SD)	MS	F statistics (df)	p-Value
Control	13	9.80 ( $\pm$ 4.706)	2880.618	29.83 (4)	<b>&lt;0.001*</b>
PB Opal	13	42.73 ( $\pm$ 13.629)			
PB Voco	13	44.32 ( $\pm$ 11.101)			
OTC Crest	13	35.09 ( $\pm$ 7.957)			
OTC WL	13	20.68 ( $\pm$ 9.404)			

Abbreviations: OTC Crest, Crest 3D whitestrip; OTC WL, Whitelight tooth whitening set; PB Opal, Opalescence PF 15%; PB Voco, Voco perfect bleach 10%.

\*One-way ANOVA test < significant at  $p < 0.05$ . Posthoc Tukey test: multiple comparisons show significant differences between control and all test groups: PB Voco and OTC Crest ( $p = 0.020$ ); PB Opal and OTC WL; PB Voco and OTC WL; OTC Crest and OTC WL with  $p < 0.001$ , respectively.

**Table 4** Comparison of mean (SD) enamel microhardness (VHN) pre and post bleaching for all groups

Groups	N	Prebleaching mean (SD) (VHN)	Postbleaching mean (SD) (VHN)	Mean difference (95% CI)	t statistics (df)	p-Value
Control	13	207.97 ( $\pm$ 54.915)	240.74 ( $\pm$ 73.235)	-32.77 (-75.013, 9.473)	-1.690 (12)	0.117
PB Opal	13	191.46 ( $\pm$ 22.750)	283.90 ( $\pm$ 68.764)	-92.44 (-133.944, -50.928)	-4.852 (12)	<b>&lt; 0.001*</b>
PB Voco	13	221.28 ( $\pm$ 57.936)	274.49 ( $\pm$ 63.142)	-53.21 (-109.335, 2.925)	-2.065 (12)	0.061
OTC Crest	13	230.46 ( $\pm$ 43.639)	241.59 ( $\pm$ 42.151)	-11.13 (-43.465, 21.211)	-0.750 (12)	0.468
OTC WL	13	234.28 ( $\pm$ 48.149)	224.41 ( $\pm$ 50.392)	9.87 (-25.106, 44.850)	0.615 (12)	0.550

Abbreviations: OTC Crest, Crest 3D whitestrip; OTC WL, Whitelight tooth whitening set; PB Opal, Opalescence PF 15%; PB Voco, Voco perfect bleach 10%. t test.

\*Significant at  $p < 0.05$ .

Comparison of mean (SD) microhardness changes (Ra) postbleaching between different groups

Groups	n	Differences mean (SD) (Ra)	MS	F statistics (df)	p-Value
Control	13	54.10 ( $\pm$ 53.654)	6362.052	2.232 (4)	0.076
PB Opal	13	92.69 ( $\pm$ 68.316)			
PB Voco	13	81.00 ( $\pm$ 67.724)			
OTC Crest	13	43.23 ( $\pm$ 31.192)			
OTC WL	13	46.18 ( $\pm$ 33.848)			

Abbreviations: OTC Crest, Crest 3D whitestrip; OTC WL, Whitelight tooth whitening set; PB Opal, Opalescence PF 15%; PB Voco, Voco perfect bleach 10%. One-way ANOVA.

**Table 5** Comparison of mean (SD) enamel surface roughness (Ra) pre and postbleaching for all groups

Groups	n	Prebleaching mean (SD) (Ra)	Postbleaching mean (SD) (Ra)	Mean difference (95% CI)	t-statistics (df)	p-Value
Control	13	0.15 ( $\pm$ 0.075)	0.19 ( $\pm$ 0.057)	-0.05 (-0.106, 0.011)	-1.765 (12)	0.103
PB Opal	13	0.09 ( $\pm$ 0.054)	0.34 ( $\pm$ 0.181)	-0.25 (-0.345, -0.150)	-5.536 (12)	<b>&lt; 0.001*</b>
PB Voco	13	0.10 ( $\pm$ 0.039)	0.34 ( $\pm$ 0.124)	-0.24 (-0.319, -0.157)	-6.374 (12)	<b>&lt; 0.001*</b>
OTC Crest	13	0.06 ( $\pm$ 0.012)	0.54 ( $\pm$ 0.118)	-0.48 (-0.549, -0.412)	-15.219 (12)	<b>&lt; 0.001*</b>
OTC WL	13	0.07 ( $\pm$ 0.023)	0.42 ( $\pm$ 0.155)	-0.35 (-0.443, -0.252)	-7.922 (12)	<b>&lt; 0.001*</b>

Abbreviations: OTC Crest, Crest 3D whitestrip; OTC WL, Whitelight tooth whitening set; PB Opal, Opalescence PF 15%; PB Voco, Voco perfect bleach 10%. Paired t-test.

\*Significant at  $p < 0.05$ .

which is absent in the PB Opal group. OTC Crest group contains more than 50% accentuated enamel surface irregularities mixed with moderate alterations. In the OTC WL group,

enamel surface was mostly represented with moderate alterations with small percentage of slight and accentuated surface irregularities.

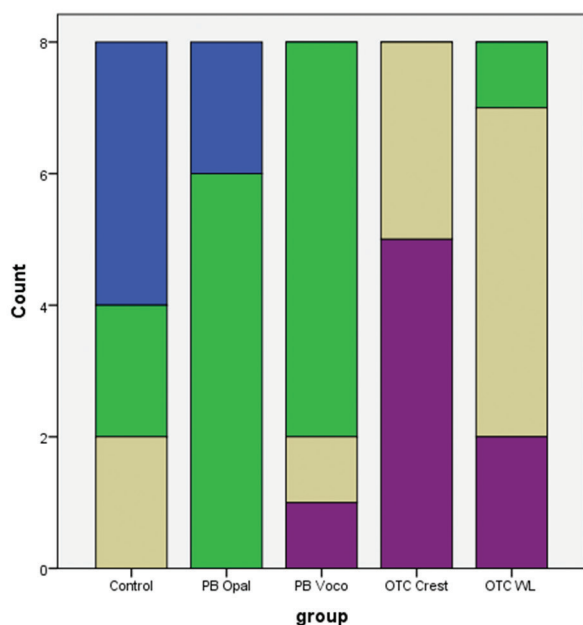


**Table 6** Comparison of mean (SD) surface roughness changes (Ra) postbleaching between groups

Groups	n	Differences mean (SD) (Ra)	MS	F statistics (df)	p-Value
Control	13	0.09 (± 0.061)	0.278	16.302 (4)	< 0.001*
PB Opal	13	0.25 (± 0.161)			
PB Voco	13	0.24 (± 0.135)			
OTC Crest	13	0.48 (± 0.114)			
OTC WL	13	0.35 (± 0.156)			

Abbreviations: OTC Crest, Crest 3D whitestrip; OTC WL, Whitelight tooth whitening set; PB Opal, Opalescence PF 15%; PB Voco, Voco perfect bleach 10%.

\*One-way ANOVA test < significant at  $p < 0.05$ . Posthoc Tukey test: multiple comparisons show significant differences between control and PB Opal ( $p = 0.003$ ); control and PB Voco ( $p = 0.004$ ); control and OTC Crest and OTC WL with  $p < 0.001$ , respectively; PB Opal and OTC Crest, PB Voco and OTC Crest with  $p < 0.001$ , respectively; PB Voco and OTC WL ( $p = 0.034$ ); OTC Crest and OTC WL ( $p = 0.013$ ).



**Fig. 1** Distribution of enamel surface morphology scores for each group ( $n = 65$ ).

► **Fig. 1** shows photomicrographs SEM at 5000x magnifications of control group (A), which demonstrates intact enamel with smooth surface morphology, while PB Voco (B and C) show enamel with slight irregularities. Group OTC Crest (D) show enamel with accentuated irregularities, increased in large and deep dimension of porosities with appearance of acid etch enamel, while OTC WL (E) show enamel with moderate irregularities with increased porosities and sign of depressions and erosions.

► **Fig. 3** shows AFM photomicrographs of all groups at 100  $\mu\text{m}$ . Enamel surface topography in all groups showed various ranges of irregularities. Enamel topography in PB Opal and PB Voco were comparable to the control group with less number of grooves and pits and smoother grooves and less depth. In OTC crest and OTC WL, enamel surface showed moderate to severe alteration with increase in roughness. Over the counter Crest enamel surface topography show shallow pits and grooves in the center but deeper and wider grooves in the periphery of the enamel surface. OTC WL group demonstrate presence of deep and wide groove in

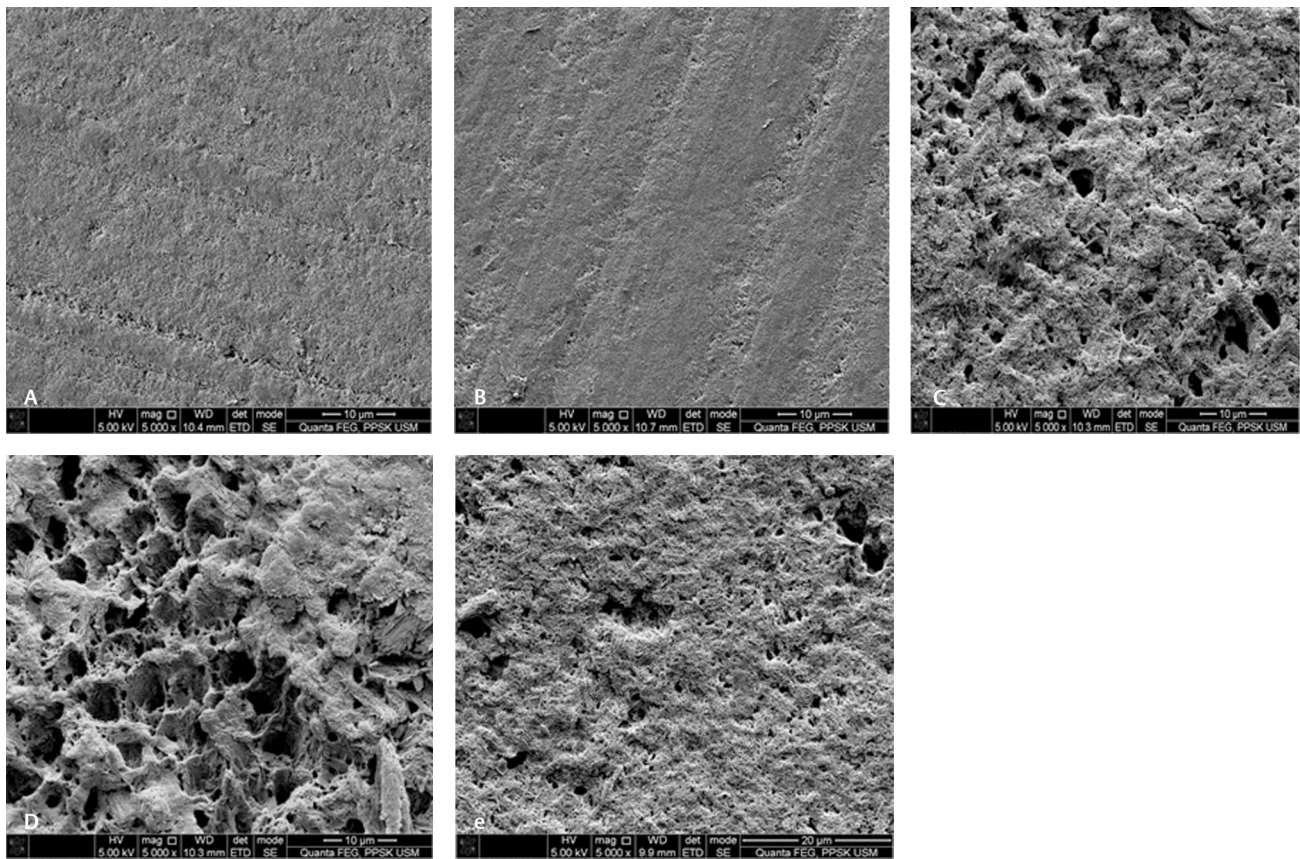
the center of the enamel surface with numerous small and shallow pits and grooves.

## Discussion

In this study, colorimetric measurement analysis showed that  $L^*$  values, which represent the brightness, increased significantly in all test groups postbleaching. The  $a^*$  values, which represent the green–red contrast, decreased significantly in all test groups postbleaching. This suggests that regardless of the bleaching regime or concentration of the bleaching agents, this study exhibits efficacy of tooth whitening, as was also found in several studies.<sup>6,8,10</sup> From the results, PB Voco showed similar bleaching efficacy with PB Opal, while PB Opal has the same bleaching efficacy with OTC Crest. Karadas and Duymus in 2015 also concluded that bleaching with strip-type or gel-type OTC products have the same efficacy in whitening the tooth compared with Opalescence PF 10%.<sup>8</sup> In this study, it was found that there was a significant decrease in the  $L^*$  values of the control group whose samples were not bleached, but only stored in distilled water. This suggests that the teeth stored in distilled water continued to become discolored or darkened over time. Whereas, in the other groups, the significant color changes were such that there was an increase of  $L^*$  value, which means the teeth became whiter.

Despite having the same efficacy with professionally prescribed bleaching agent, there is no long-term evidence of safety and bleaching stability with the nonprescription OTC products. The OTC products may give immediate effects to the color changes; however, it would be mandatory to monitor the long-term results and their detrimental effects on the enamel. The nonprescription OTC products active components' concentrations have not been declared by the manufacturer. Thus, concentration-related efficacy of different bleaching agents could not be concluded in this study.

Surface microhardness alteration is said to be associated with the loss and gain of minerals (balance in remineralization and demineralization) of the dental hard tissues<sup>21,22</sup> which is linearly associated with mineral content.<sup>23</sup> This study reveals significant increase in the enamel microhardness in the PB Opal group compared with the other groups. Normal value of human enamel microhardness was recorded as being in the range of 211.2 to 443.07(HVN).<sup>24</sup> In this study,



**Fig. 2** (A) Scanning electron microscope photomicrograph of control group demonstrates intact enamel with smooth surface morphology. (B) Scanning electron microscope photomicrograph of PB Opal Group demonstrates enamel with slight irregularities. (C) Scanning electron microscope photomicrograph of PB Voco Group demonstrates enamel with slight irregularities. (D) Scanning electron microscope photomicrograph of OTC Crest Group showed enamel with accentuated irregularities, and increase in large and deep dimension of porosities with appearance of acid etch enamel. (E) Scanning electron microscope photomicrograph of OTC WL Group showed enamel with moderate irregularities with increased porosities and sign of depressions and erosions.

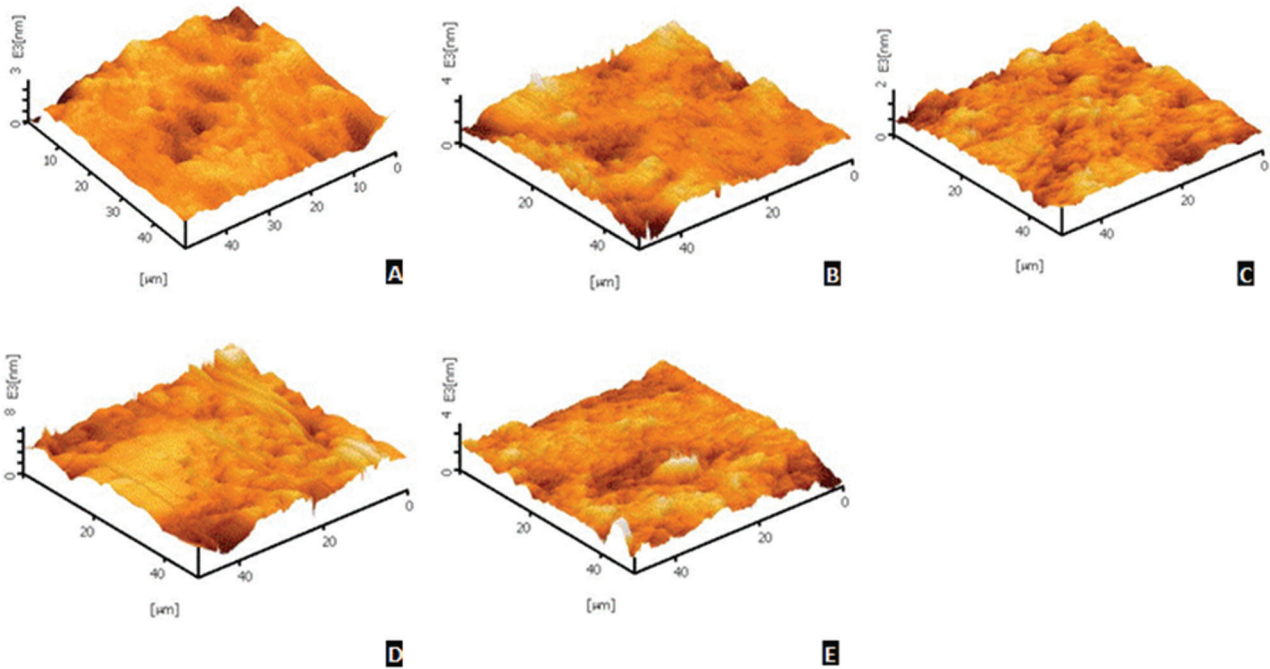
initial prebleaching enamel microhardness was found to be in the normal range except in the PB Opal group and control group. However, the result postbleaching showed significant increase in the enamel surface microhardness in the PB Opal group. This could be due to the fluoride incorporation in the bleaching agent<sup>1</sup> which causes less demineralization in the enamel surface.<sup>25</sup> PB Opal group contains PF (potassium nitrate and fluoride). The 0.11 percent ion fluoride in the opalescence PF formulation causes less hazardous effects on the enamel mineral content.<sup>26</sup>

Storage media used in this study was normal saline instead of normal or artificial saliva which has been found to provide protection and recovery to bleached enamel.<sup>27,28</sup> This not only eliminates the possible confounding factor of potential remineralization effect from saliva in the bleached enamel but also further strengthens the beneficial effect of fluoride addition in PB Opal.<sup>1,27-29</sup> The relevance of maintaining enamel surface hardness to the normal value after bleaching is to ensure the integrity of the outermost layer of the teeth is healthy, thus serving its main function in the oral cavity.

Interaction of the bleaching agent and the oral tissue is critically important as the usage of the bleaching agent may negatively affect the oral structure.<sup>30</sup> Altered enamel surface roughness has become a major problem in vital

bleaching. A few researchers have found that there are increases in the enamel surface roughness after the bleaching procedure.<sup>4,12,21,30-32</sup> This is due to the oxidizing process that occurs during bleaching treatment.<sup>21</sup> In this study, the profilometer readings showed significant increase in the enamel surface roughness in all experimental groups. This is in contrast with other studies which reported no changes in the enamel surface roughness after bleaching.<sup>32-34</sup> In this study, the surface roughness was seen when the enamel was bleached with the lower concentration of active agent, whereas Pinto and colleagues and Cavalli and colleagues found only significant increase in the enamel surface roughness with higher concentration carbamide peroxide or hydrogen peroxide. The increased enamel surface roughness postbleaching may be due to the low pH of the bleaching solutions. PH value contributes as an important factor in the reactions of the bleaching process; however, this low-pH bleaching solution leads to demineralization changes with erosion pattern, which is believed to occur irreversibly.<sup>35,36</sup> In this present study, OTC bleaching products give lower pH measurement (—Table 1) than the professionally prescribed bleaching agents which further leads to the erosive effect on enamel surface.<sup>4,37</sup> This validates the result of the highest surface roughness seen in group bleached with nonprescription OTC products

### Atomic Force Microscopy



**Fig. 3** (A) Atomic force microscopy photomicrographs of control group at 100  $\mu\text{m}$ . (B) Atomic force microscopy photomicrographs of PB Opal group at 100  $\mu\text{m}$ . (C) Atomic force microscopy photomicrographs of PB Voco group at 100  $\mu\text{m}$ . (D) Atomic force microscopy photomicrographs of OTC Crest group at 100  $\mu\text{m}$ . (E) Atomic Force Microscopy Photomicrographs of OTC WL group at 100  $\mu\text{m}$ .

(OTC Crest and OTC WL) compared with professionally prescribed bleaching agent. Increase in the enamel surface roughness of bleached enamel is a concern as rough enamel surface was found to attract more adherence of *Streptococcus mutans* (*S. mutans*) compared with unbleached surface.<sup>38,39</sup> Adhesion of *S. mutans* to the bleached enamel surface acts as an important contribution to the initiation and progression of dental caries and periodontal disease.<sup>39</sup>

The profilometer readings of this study are supported by the SEM and AFM data. SEM has been widely used for the qualitative evaluation of surface morphology of enamel and dentine following tooth bleaching. Various morphologic changes has been observed in previous studies, characterized by mild interprismatic dissolution, appearance of deep longitudinal cavities at the superficial part of enamel not affecting dentine, dissolution of enamel superficial area with opening of dentinal tubules, intermittent depression of various depths and diameters, dissolution of superficial enamel, and loss or eroded enamel prism core with retained periphery area, apparent cross-striation, and striae of retzius<sup>4,40-42</sup>.

It is known that enamel, when exposed to solutions with lower than pH 5.5, will begin to dissolve, appearing as demineralization. The pH of nonprescription OTC Crest bleaching agent in this study was lower than the critical pH of enamel (pH 4.60). Due to the lowest pH recorded for non-prescription OTC Crest group, it supported the result of accentuated morphological changes for this bleaching group. The PB Opal group, which had significant increase in surface hardness,

showed only mild surface morphology alteration in the SEM result. The increase in enamel surface microhardness reflects the uptake of minerals in the enamel; thus, less enamel surface alteration or demineralization upon observation through SEM.

AFM is capable of scanning objects up to atomic resolution with minimal sample preparation. AFM provides a three-dimensional surface profile with high resolutions. This technique has been widely used to characterize the erosion of enamel and dentine.<sup>43</sup> In the AFM result, the enamel topography in all groups shows various ranges of irregularities. Nonprescription OTC bleaching groups (OTC Crest and OTC WL) exhibit more numerous deeper peaks and valleys. This is supported by a previous study which suggests a more microporous layer on the enamel tissues.<sup>44</sup> However, enamel surface topography in PB Opal group showed fewer alterations compared with the other groups in our study. This could possibly be due to the presence of sodium fluoride incorporated in the bleaching agent which leads to remineralizing effects, as found by other researchers.<sup>20,45</sup>

Hydroxyapatite on tooth surface is undergoing a unique interaction with salivary components in the oral environment, which involves the interchange of various ions that regulate the remineralization and demineralization process.<sup>36</sup> The changes of human enamel after tooth bleaching in experimented *in vitro* study may not be clinically relevant, as it is difficult to replicate actual oral environment which varies from individual to individual.



## Conclusions

Within the limitation of this study, it can be concluded that

1. All bleaching agents showed significant color changes with an increase in the brightness and green-red contrast. Professional prescribed bleaching agents were more efficient in whitening teeth compared with the OTC bleaching products
2. PB Opal (Professionally Prescribed Opalescence PF 15%) showed significant increase in enamel microhardness compared with other bleaching agents used in our study.
3. All groups showed significant increase in enamel surface roughness
4. PB opal showed enamel surface morphology the same as unbleached enamel, while the other groups showed enamel morphological changes ranging from mild to accentuated
5. All groups showed a range of surface topography. Non-prescription OTC groups showed the roughest surface topography, characterized by increase in grooves and porosities.
6. Within the limitations of this present study, it can be concluded that professionally prescribed bleaching agent Opalescence PF 15% is effective in whitening the tooth with positive outcomes in maintaining enamel surface hardness *in vitro*, while the other bleaching products used in this study may be effective but have deleterious effects on the enamel.

### Key Messages

Care needs to be taken when using bleaching agents, especially with regard to nonprescription, OTC bleaching agent, as it can cause significant changes to enamel with repeated usage.

### Note

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### Conflict of Interest

None declared.

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