©Operative Dentistry, 2013, **38-1,** 100-110

Effects of Two In-Office Bleaching Agents with Different pH on the Structure of Human Enamel: An *In Situ* and *In Vitro* Study

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Clinical Relevance

Salivary pellicle and adequate calcium and phosphate ions present in natural human saliva play pivotal roles in protecting the surface of human enamel during the process of in-office bleaching.

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DOI: 10.2341/11-173-L

SUMMARY

This study evaluated the effects of two in-office bleaching agents (Beyond and Opalescence Boost) with different pH on the structure and mechanical properties of human enamel *in vitro* and *in situ*. One hundred and eight enamel slabs were obtained from freshly extracted premolars. The specimens were randomly distributed into nine groups (n=12), and the human saliva (HS) in the volunteers' oral cavities was used to simulate the *in situ* condition: Beyond + HS, Opalescence Boost (O-Boost) + HS, Control + HS, Beyond + artificial saliva (AS), O-Boost + AS, Control + AS, Beyond + distilled water (DW), O-Boost + DW, and Control + DW. The bleaching treatments were performed on the first and eighth day,

and the total bleaching time was 90 minutes. Baseline and final surface roughness (RMS), surface morphology, microhardness, and fracture toughness (FT) were measured before the treatment and on the fifteenth day, respectively. Compared with control groups, surface alterations on enamel were found in the Beyond + AS and Beyond + DW groups under atomic force microscopy evaluation. Two-way analysis of variance and Tukey test revealed that the RMS showed significant intergroup differences for both storage condition and bleaching agent, whereas microhardness and FT revealed no significant alteration. The results indicated that in-office bleaching agents with low pH values could induce enamel morphology alterations under *in vitro* conditions. The presence of natural HS could eliminate the demineralization effect caused by low pH.

INTRODUCTION

Vital tooth bleaching has become a popular procedure for treating discolored teeth because of its simplicity and conservation property.[1] Currently, three fundamental approaches are adopted for vital tooth bleaching, which are at-home bleaching, in-office bleaching, and over-the-counter bleaching products.[2,3] Although at-home bleaching is effective and widely accepted, in-office bleaching still seems to be appropriate for the patient in some cases, such as severe or single tooth discoloration, lack of compliance, or desire for immediate whitening to recover confidence.[4]

The efficacy of in-office bleaching has been well documented, but a primary concern is that the enamel structure may be weakened by the bleaching agent.[5] Numerous studies were conducted to evaluate the safety of in-office bleaching agents on enamel. However, the results of these investigations usually conflict with each other. Some studies reported that in-office bleaching might have detrimental effects, such as alterations in surface morphology,[6,7] changes in chemical composition,[8,9] and a decrease in microhardness and fracture toughness (FT).[5,8,10,11] In contrast to these findings, there were also some investigations that held the opposite view to those observations above.[12,13] The great inconsistency in the outcome of those studies might be due to differences in study design[2,14] (type of storage condition, time of evaluation, different bleaching agents, time of application, bleaching agent pH, and so on). Among these factors, the pH of bleaching agents and storage conditions may have crucial influences on the results.

Generally, distinct bleaching agents have different pH values. Since acid bleaching agents may help to keep hydrogen peroxide stable and facilitate the bleaching process,[4] some in-office bleaching products containing highly concentrated hydrogen peroxide have a low pH. The possible adverse effects of in-office bleaching agents have been evaluated by previous studies. However, most of them focused on the concentration;[12,13,15,16] whether the pH value of in-office bleaching agents plays an important role in the bleaching treatment still remains unclear. In addition, storage condition is also an element that should be taken into account. Typically, studies on in-office bleaching use distilled water (DW) or artificial saliva (AS) as the storage condition. These models could partially influence the findings, but they had some limitations. Compared with *in vitro* studies, *in situ*/*in vivo* ones may better simulate clinical conditions and really reveal influences of bleaching agents on enamel.[2] Nevertheless, few investigations[11,13] about in-office bleaching have been done using *in situ*/*in vivo* methodologies, and none of them combined *in situ*/*in vivo* experiments with *in vitro* ones to compare the effects of different pH values and different storage conditions on human enamel.

Therefore, the primary purpose of the present study was to evaluate the effects of different pH values and different storage conditions on the structure and mechanical properties of dental enamel during the in-office bleaching process. This goal was achieved by the complementary use of atomic force microscopy (AFM), microhardness, and FT measurements. The null hypotheses in this study were that 1) the pH values of bleaching agents and storage conditions (*in vitro* and *in situ*) had no effect on the morphology alteration of human enamel during the in-office bleaching process and 2) the pH values of bleaching agents and storage condition (*in vitro* and *in situ*) had no effect on the mechanical properties of human enamel during the in-office bleaching process.

MATERIALS AND METHODS

Ethical Aspects and Volunteers

The protocol for this study was reviewed and approved by the Ethics Committee of the School and Hospital of Stomatology, Wuhan University. Four undergraduate dental students (2 men and 2 women, aged 20 to 22 years) who fulfilled the

inclusion criteria (absence of dental caries and/or periodontal disease, normal saliva flow, willing to perform bleaching treatment on the research schedule) without violating the exclusion criteria (restorations and prostheses in mouth, use of orthodontic appliances, dentin sensitivity, and smokers) were enrolled in the study after signing an informed consent form as volunteers.

Tooth Selection

The sample size was determined from the data of preliminary study in our research group. Assuming a standard type I error rate of α=0.05 and a standard type II error rate of "β=0.20 (power=0.80), a sample size of 12 enamel samples was calculated for each group to achieve more reliable results.

To achieve the calculated sample size, 54 freshly extracted orthodontic premolars were selected. All of them were examined under magnification (20×) to detect enamel cracks or fractures, carious, stains, and other defects. The teeth were cleaned thoroughly and stored in 0.2% thymol at 4°C until required.

Materials Preparation

The roots of stored teeth were separated from their crowns at the cementoenamel junction using a low-speed water-cooled diamond saw (Isomet, Buehler Ltd, Lake Bluff, IL, USA) under water cooling. Two dental slabs (4 mm × 3 mm × 2 mm) were obtained from the middle third of the buccal surface of each tooth and subjected to steam sterilization to avoid bacterial contamination. Each dental slab was placed in a polyvinyl chloride matrix and fixed with a colorless translucent acrylic resin, keeping the enamel surface unsealed for bleaching applications. The specimens were serially polished by means of 600-, 1000-, 1500-, and 2000-grit SiC papers with water as a cooler to obtain flat standardized enamel surfaces. Subsequently, they were polished with diamond spray (1 μm, 0.5 μm) and polishing cloths, followed by rinsing with running water to get rid of debris layers. At last, all samples were placed in an ultrasonic cleaning machine and immersed in DW for 5 minutes to remove residual particles and smear layers. Prior to the experiment, all prepared specimens were stored in AS, which was renewed every day in a 37°C incubator for 7 days to standardize the initial conditions. The AS[17] used in this study contained calcium and phosphate with a known concentration (50 mmol/L KCl, 1.5 mmol/L Ca, 0.9 mmol/L PO_4, 20 mmol/L tri-hydroxymethyl-aminomethane, pH=7.0).

For each volunteer, a full-arch maxillary impression was obtained and a stone cast mold made based on the impression. A 0.035-inch-thick soft bleaching tray (Soft-Tray Sheets, Ultradent Products Inc, South Jordan, UT, USA) was fabricated on the cast using a vacuum tray–forming machine (Ultraform, Ultradent Products Inc) and then modified with a palatal extension.

The two in-office bleaching agents used in the current study were Opalescence Boost (O-Boost, 38% hydrogen peroxide gel, Ultradent Products, South Jordan, UT, USA) and Beyond (35% hydrogen peroxide gel, Beyond Technology Corp, Santa Clara, CA, USA). The pH values of each bleaching agent were measured by a digital pH electrode (EASY-FERM PLUS 225, Hamilton, Bonaduz, Switzerland) three times, and then mean values were recorded as their final pH values. The information about the two in-office bleaching products is shown in Table 1.

Initial Measurements

AFM Detection—Initial surface roughness and surface morphology detections were performed using atomic force microscopy. This was conducted using a Shimanzu SPM-9500J3 (Shimadzu Corp, Kyoto, Japan). AFM software SPM-Offline (version 2.30) was used to obtain the surface roughness (RMS), which represents the average of the square height difference between surface peaks and valleys (root mean square of the heights). Meanwhile, three images (10 μm × 10 μm) of each specimen were obtained.

Microhardness Test—Vickers indentation microhardness baseline values were performed using a microhardness tester (HXD-1000TMC/LCD, Taiming Inc, Shanghai, China). Three indentations were made on each specimen with 100*g* for 15 seconds. The surface area of the flattened enamel was sufficient for each indentation without interfering with each other (Figure 1).

Fracture Toughness Test—Vickers indentations with a load of 9.8 N were performed to assess baseline FT values on each enamel surface (Figure 1). These indentations for FT and cracks were then recorded immediately under a light microscope with 400-fold magnification, which belonged to the microhardness tester (HXD-1000TMC/LCD, Taiming Inc. Shanghai, China).

The diagonal lengths of each indentation were measured for the evaluation of enamel FT. The hardness values were determined according to the following expression[18]:

Table 1: *In-Office Bleaching Agents Used in the Study*

Product	Composition, pH	Activation System	Application
Opalescence Boost	38% hydrogen peroxide, fluoride, potassium nitrate, thickening agent, gel, pH=7.52	Chemically activated	First and eighth day for 45 minutes each; total of 90 minutes
Beyond	35% hydrogen peroxide, H_2O, thickening agent, gel, pH=4.03	Powerful light-emitting diodes (LED) emit a high-intensive blue light (about 480-nm wavelength)	First and eighth day for 45 minutes each; total of 90 minutes

$$H = 0.47P/a^2$$

where H=hardness, a=half the diagonal of the indentation (m), and P=applied load (MN) (9.807×10^{-6} MN).

The lengths of the cracks were measured from optical micrographs using Image J software (version 1.41). For each indentation, a circle enclosing all associated cracks (both edge and side cracks) was drawn, and its radius, developed from the center of the indentation, was taken as the maximum crack length value of the corresponding indentation. FT was then computed by means of the following formula[19]:

$$K_{IC} = 0.016(E/H)^{1/2}P/c^{3/2}$$

where K_{IC}=fracture toughness ($N/\mu m^{3/2}$), E=Young's modulus (GPa), H=Vickers hardness (GPa), P=applied load (N), and c=the maximum crack length (μm) from the center of the indentation impression. The Young's modulus of human enamel was taken at 84.1 GPa.[20]

After initial measurements, specimens were randomly divided into nine groups, according to the bleaching agents and storage conditions (n=12): group Beyond + human saliva (HS), group O-Boost + HS, group Control + HS, group Beyond + AS, group O-Boost + AS, group Control + AS, group Beyond + DW, group O-Boost + DW, and group Control + DW (Table 2).

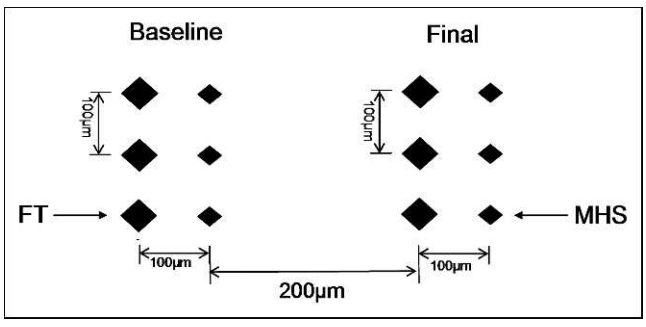

Figure 1. *Location of initial and final indentations performed for microhardness and fracture toughness test.*

Thirty-six dental slabs for *in situ* groups were removed from the polyvinyl chloride matrix by using probes and then were fixed on the palatal extension of the modified bleaching trays of four volunteers with light cure restorative material (3M ESPE, St Paul, ST, USA). Nine specimens arrayed in three columns for each volunteer: the first ones in each column were used as control specimens, and the second and third ones were the experimental specimens (Figure 2).

Once the *in situ* enamel specimens were fixed in the modified trays, they were cleaned with DW and then kept in the oral environment of volunteers for one day to form *in situ* pellicle to mimic the clinical situation. During the intraoral exposure period, the enamel slabs were cleaned every 12 hours with DW and a toothbrush without any dentifrice for a few seconds to mimic the daily oral hygiene conditions. Dentifrice was forbidden in an attempt to minimize its adverse abrasive effects on the pellicle and diminish the interference caused by other components of dentifrice, such as fluoride and desensitizing agents. Moreover, the trays were removed only for meals and stored in a 100% humidity environment to exclude the effects of food components on pellicle development.

To have a comparable model, enamel specimens for AS and DW were also exposed to the corresponding storage environment until the bleaching treatment started.

Bleaching Procedure

After one day's storage, the bleaching procedure started on the first day. Based on a usual clinical scenario, two in-office visits and three bleaching episodes for each visit were performed in this study. For the first visit, all specimens were taken out of the storage environment and dried by compressed

Table 2: *Bleaching Agent and Storage Condition in Each Group*

Storage Condition	Bleaching Agent		
	Beyond	O-Boost	Control
HS (in situ)	Beyond + HS	O-Boost + HS	Control + HS
AS (in vitro)	Beyond + AS	O-Boost + AS	Control + AS
DW (in vitro)	Beyond + DW	O-Boost + DW	Control + DW

Abbreviations: AS, artificial saliva; DW, distilled water; HS, human saliva; O-Boost, Opalescence Boost.

air for 15 seconds. Then the two bleaching agents were applied on the specimens according to the standard bleaching procedure extraorally as follows: 1) positioned the head of the matched light (Beyond Technology) upon the teeth surfaces for Beyond groups; 2) painted the specimen surfaces with corresponding whitening gels to form a 2-mm-thick layer; 3) started the timer and turned on the light at the same time, with the first bleaching episode lasting for 15 minutes at room temperature; 4) removed the bleaching agents carefully with soft cotton pellets and then repeated steps 1, 2, and 3 twice; 5) removed the bleaching gel carefully with soft cotton pellets under running DW; and 6) dried the surfaces of samples and stored *in vitro* groups in AS or DW. For the *in situ* groups, the modified bleaching trays were replaced in the oral cavity of the four volunteers as previously described. Then, the first in-office visit ended.

After seven days' storage, the second in-office visit started on the eighth day. We repeated all the procedures above and then kept samples in the corresponding storage environment for another seven days' storage.

Final Measurements

On the fifteenth day, the *in situ* samples were replaced into the polyvinyl chloride matrix, and all samples were then retested for final measurements of surface roughness RMS, surface morphology, microhardness, and FT. The experimental design is summarized in Figure 3.

The percentage of microhardness loss (PML) was calculated using the following calculation: $PML(\%) = (VHN_{(B)} - VHN_{(F)})/VHN_{(B)}$, where $VHN_{(B)}$ was the average of the baseline microhardness values and $VHN_{(F)}$ was the average of final microhardness values. The variation of RMS (ΔRMS) was expressed as follows: $\Delta RMS = RMS_{(F)} - RMS_{(B)}$, where $RMS_{(B)}$ was the average of the baseline surface roughness RMS values and $RMS_{(F)}$ was the average of final surface roughness RMS values. Similarly, the variation of FT (ΔFT) was expressed as follows: $\Delta FT = FT_{(F)} - FT_{(B)}$, where $FT_{(B)}$ was the average of the baseline FT values and $FT_{(F)}$ was the average of final FT values.

Statistical Analysis

Statistical analysis was performed by SPSS 16.0 for Windows. All statistical analyses were carried out at a significance level of 0.05. Mean values of RMS, microhardness, and FT of samples in the experiment were expressed as means ± SD. Variations for baseline and final values in each group were analyzed by analysis of variance (ANOVA). Since the storage condition and the bleaching agent were investigated as two main factors, a two-way ANOVA test and an additional Tukey *post hoc* analysis was used to analyze the effects of the two main factors on ΔRMS, PML, and ΔFT of the enamel.

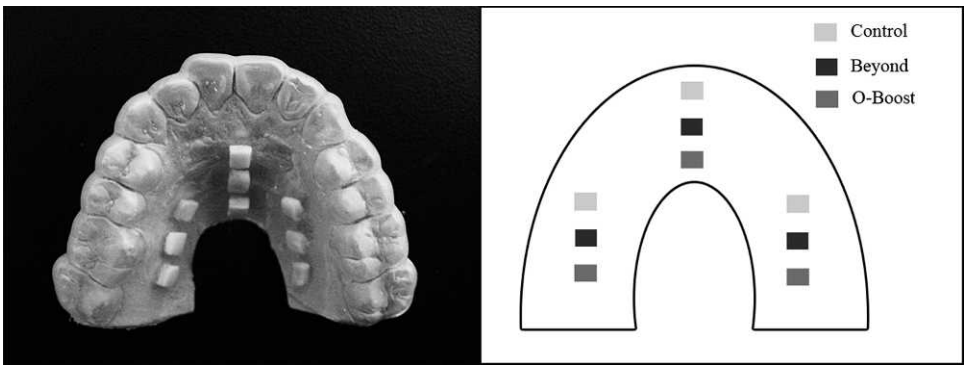

Figure 2. *Distribution of the experimental and control samples for the* in situ *condition.*

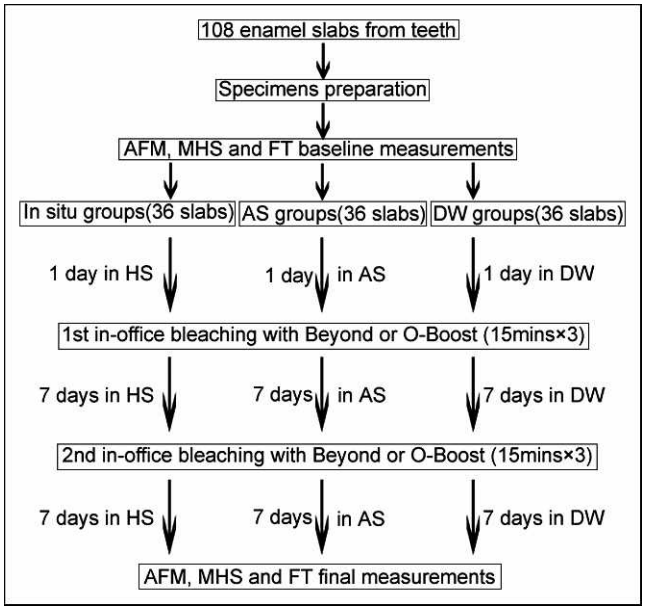

Figure 3. *Flow diagram of the experimental process in the study.*

RESULTS

AFM Detection

The morphological alterations of enamel surface were obtained by means of AFM. Figure 4 illustrates the means and standard deviations of enamel surface roughness RMS in each group. After treatment, the RMS of group Beyond + DW and group Beyond + AS increased significantly ($p<0.001$, $p<0.001$, respectively). No significant increase of RMS was found in any other groups ($p>0.05$).

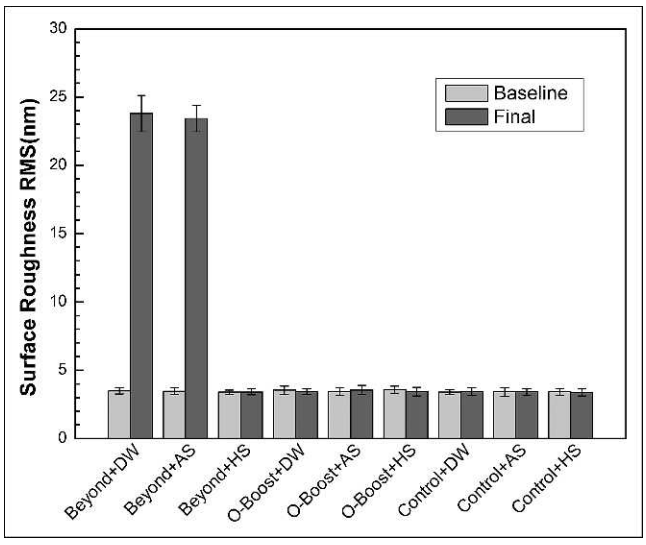

Figure 4. *RMS values following bleaching for each group. Compared with baseline RMS values, significantly higher final RMS values are shown in group Beyond + AS and group Beyond + DW.*

Table 3: *Two-Way Analysis of Variance of ΔRMS*

Source	Dependent Variable	*df*	Mean Square	*F*	*p* Value
Storage condition (A)	ΔRMS	2	551.545	1.221E3	0.000
Bleaching agent (B)	ΔRMS	2	2171.126	4.805E3	0.000
A × B	ΔRMS	4	534.185	1.182E3	0.000

Two-way ANOVA on RMS revealed statistically significant intergroup differences ($p<0.001$) for both factors and their interaction ($p<0.001$; Table 3). The additional Tukey *post hoc* test was used to clarify differences among the groups. For the factor of storage condition, the Tukey test revealed significant differences between the *in situ* condition and DW or between the *in situ* condition and AS ($p<0.001$, $p<0.001$, respectively). No significant difference was found between DW and AS ($p=0.960$). For the factor of bleaching agent, the Tukey test demonstrated significant differences between Beyond and O-Boost or between Beyond and Control ($p<0.001$, $p<0.001$, respectively). No significant difference was revealed between O-Boost and Beyond ($p=0.957$).

Representative AFM images are shown in Figures 5 and 6, and these images were in accordance with RMS data. It could be found that the grooves on the enamel surfaces became deeper and wider in the group Beyond + DW and the group Beyond + AS, and morphology alterations were more evident in the group Beyond + DW. Even some enamel rods and narrow interrod structures were seen in this group (Figure 5). To the contrary, no obvious alteration was found on enamel surfaces in the group Beyond + HS (Figure 5) and the surfaces of specimens treated by O-Boost (Figure 6). These patterns of microscopic enamel surfaces appeared to be relatively flat surfaces with some irregular and shallow grooves, resulting from the various polishing treatments.

Microhardness Test

Figure 7 provides the means and standard deviations of Vickers microhardness in each group. There was no difference between baseline and final values in all groups ($p>0.05$). Two-way ANOVA on PML revealed no statistically significant differences among the groups for both main factors ($p>0.05$) and their interactions ($p>0.05$; Table 4).

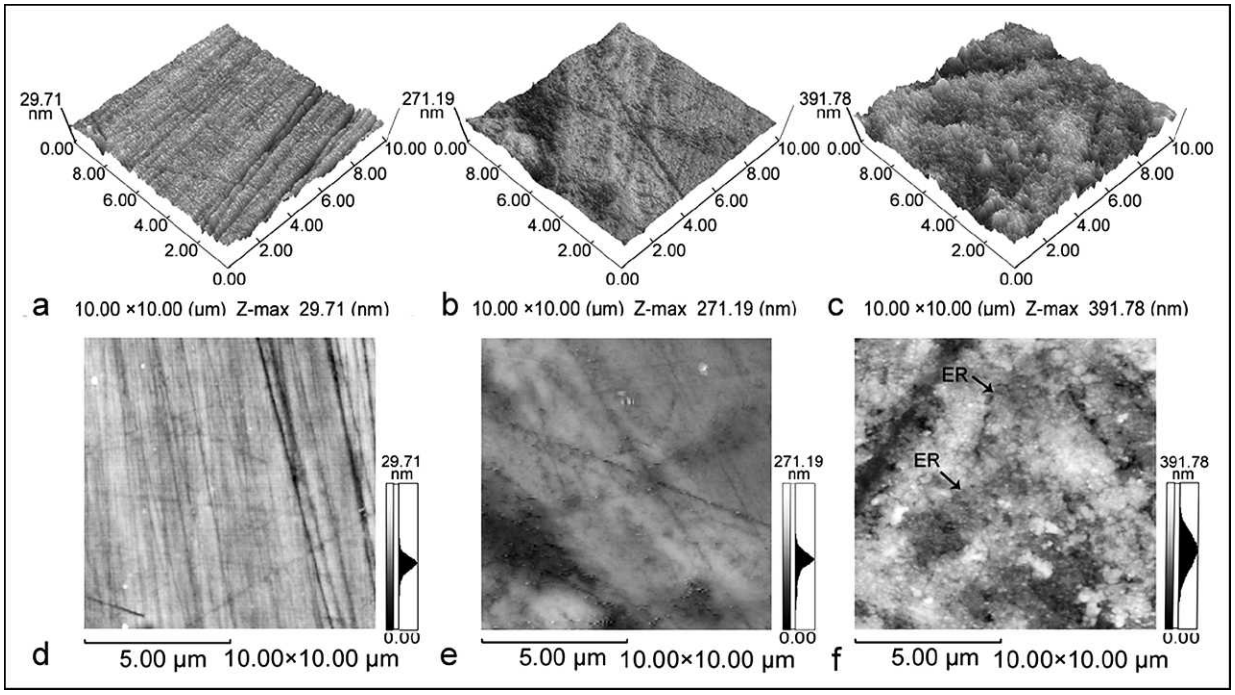

Figure 5. *Atomic force microscopy (AFM) images of specimens in the Beyond groups with three-dimensional (upper) and corresponding two-dimensional (lower) observations. (a, d) Group Beyond + HS. (b, e) Group Beyond + AS. (c, f) Group Beyond + DW. The enamel rod was depicted as ER.*

FT Test

FT values were calculated according to the aforementioned formula. Figure 8 denotes the means and standard deviations of FT in each group. After treatment, no significant decreases were found in all groups when comparing the baseline and final values ($p>0.05$). Two-way ANOVA on ΔFT exhibited

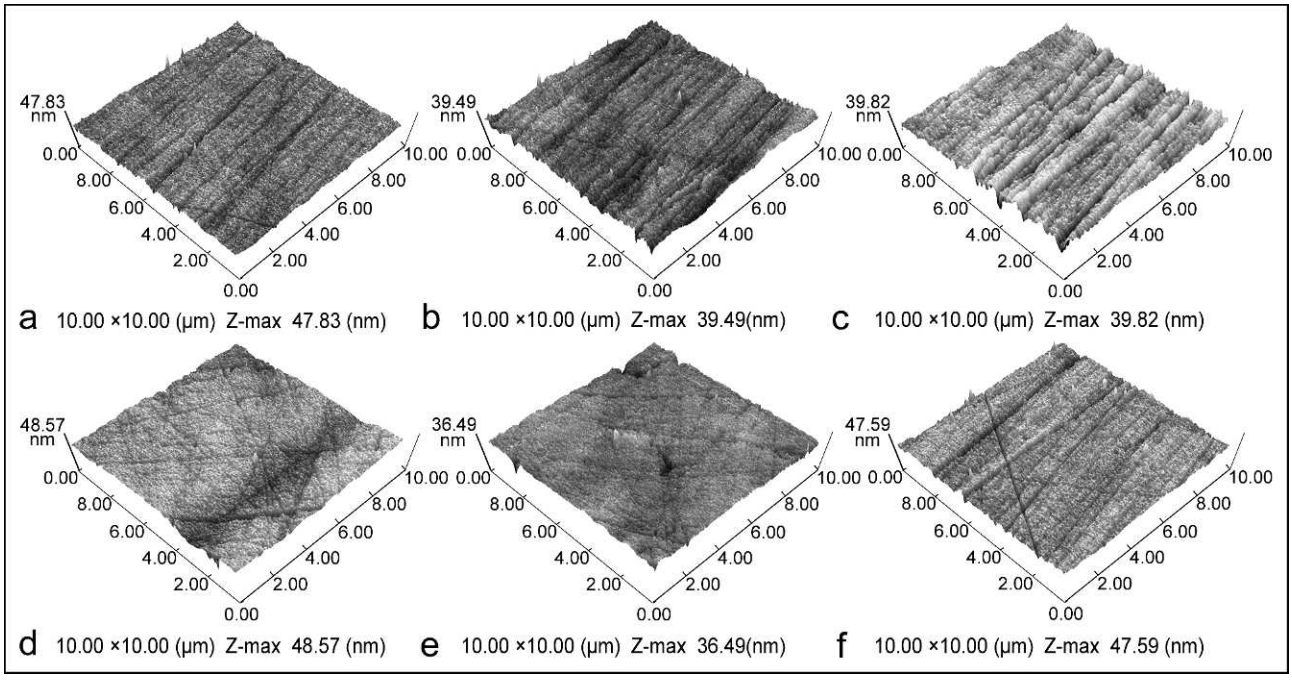

Figure 6. *Atomic force microscopy (AFM) images of specimens in O-Boost and control groups with three-dimensional observations. (a) Group O-Boost + HS. (b) Group O-Boost + AS. (c) Group O-Boost + DW. (d) Group Control + HS. (e) Group Control + AS., (f) group Control + DW.*

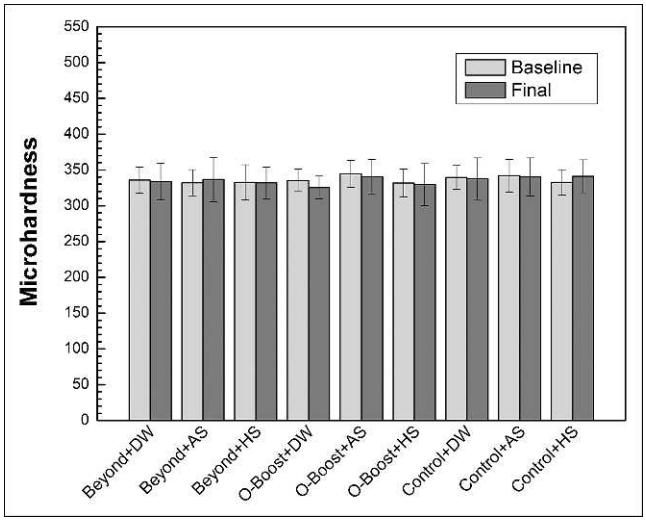

Figure 7. *Microhardness (100 g) values following bleaching for each group. No significant difference was found between the baseline and final values in each group.*

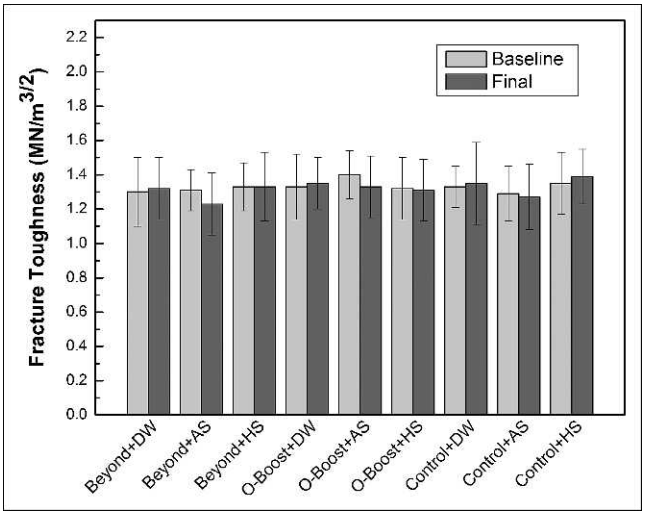

Figure 8. *Fracture toughness values following bleaching for each group. No significant difference was found between baseline and final values in each group.*

no statistically significant differences among the groups for both main factors ($p>0.05$) and their interactions ($p>0.05$; Table 5).

DISCUSSION

The results of this study indicated that bleaching agents with low pH values could induce alterations of enamel surfaces under the *in vitro* situation, while no change was observed in bleached specimens that were exposed to *in situ* conditions. Moreover, the pH values of bleaching agents and storage condition (*in vitro* and *in situ*) had no effect on the mechanical properties of human enamel after bleaching. Therefore, the first null hypothesis was partially rejected, and the second null hypothesis was accepted.

Group Beyond + DW and group Beyond + AS revealed obvious changes on enamel surfaces when compared with control groups in this study. On the contrary, little morphological modification of the enamel surface was found with the application of O-Boost in group O-Boost + DW and group O-Boost + AS. RMS results showed agreement with the morphology observations. It was claimed that the oxidative effect, pH, and composition of the bleaching agents could induce possible side effects during tooth bleaching.[5] In the current study, O-Boost and Beyond had a similar concentration of HP, but O-Boost did not result in surface alterations of specimens in DW or AS. Thus, under the same condition, the morphological alterations in Beyond groups could not be attributed to the oxidative effect of bleaching agents. Moreover, previous studies[11,21] reported that thickened agents could promote the decline of microhardness in enamel; however, it was assumed that the low pH value contained in thickened agents may be the reason for the demineralization.[21] Therefore, the pH value might be the main factor for consideration.

In the present study, the pH was 7.52 in O-Boost and 4.03 in Beyond. It has been reported that

Table 4: *Two-Way Analysis of Variance of PML*

Source	Dependent Variable	*df*	Mean Square	*F*	*p* Value
Storage condition (A)	PML	2	0.005	1.043	0.355
Bleaching agent (B)	PML	2	0.006	1.243	0.292
A × B	PML	4	0.002	0.330	0.857

Table 5: *Two-Way Analysis of Variance of ΔFT*

Source	Dependent Variable	*df*	Mean Square	*F*	*p* Value
Storage condition (A)	ΔFT	2	0.075	1.227	0.297
Bleaching agent (B)	ΔFT	2	0.005	0.077	0.926
A × B	ΔFT	4	0.009	0.149	0.963

enamel demineralization occurs when the pH falls below 5.2.[2,22,23] Thus, under the *in vitro* condition, the lower pH value may cause the demineralization of enamel surfaces. This viewpoint was supported by our previous study.[5] In that study, alkaline hydroxyapatite (HA) effectively minimized the demineralization of enamel surfaces caused by an acidic composition containing 30% HP by elevating the pH value of the HP solution and forming a protective layer for lessening the direct contact of acid HP to enamel.

By comparing AFM images obtained following treatment between group Beyond + AS and group Beyond + DW (Figure 5), typical enamel prisms and interprisms were observed in only the latter one. It indicated that some degrees of remineralization probably happened on the enamel surface in AS. This might be due to the high level of calcium and phosphate components contained in AS. These components could interact with enamel surfaces and precipitate spontaneously during the storage episode,[24,25] inducing remineralization of an existing enamel lesion.

It was of great value to note that little change was detected on the enamel surface in group Beyond + HS in this study, indicating that HS better counteracted the adverse effects of low pH compared with AS or DW. This result showed great accordance with some previous studies.[13,26]Apart from the similar calcium, phosphate, and fluoride ions in AS, HS had two other advantages that could account for its superior protective ability compared with AS.[27] One advantage was that HS with organic components might promote the formation of a pellicle layer,[2,14] which was a potentially protective surface layer that consisted mainly of salivary proteins and covered the underlying enamel surface in the oral environment.[28,29] Previous experiments[30,31] found that the *in situ*–formed short-term salivary pellicle, even within 3 minutes, could protect the enamel surface to a certain extent against demineralization. In the current study, the *in situ* samples were placed in the corresponding HS 24 hours prior to bleaching to mimic the clinical condition. Therefore, it was speculated that the salivary pellicle had formed enough thickness to offer effective protection against bleaching agents with low pH values. Another advantage of HS was that bicarbonate and phosphate-buffering systems in HS could hinder the decline of pH values and played an important role in the remineralization process when pH was within physiological limits.[26,32,33]

It was worth noting that none of the bleaching/storage regimens had any effect on the mechanical properties of the enamel specimens. This result was in contrast to several previous studies.[9,11,16,34,35] We suggested that different bleaching agents and the relatively short bleaching procedure in the current study might be the reasons for the mechanical results. These factors resulted in very shallow demineralization of the enamel surface. As the AFM images showed, the most deleterious effect was confined to less than 400 nm under the enamel surface. These relative small variations in the outermost layer of enamel were not sufficient to influence whole mechanical properties of enamel.

One limitation of the present study design should be noted. Under the clinical situation, bleaching agents are typically applied on the labial or buccal surface of the anterior teeth, whereas in the current study, we placed the enamel blocks on the palate. That was mainly attributed to the consideration of comfort and esthetic effects for the volunteers. Moreover, some previous *in situ* studies[36,37] chose the palatal model to simulate the condition of the human oral cavity and successfully proved its superiority over the *in vitro* strategy. It was possible that the action of the tongue may have removed superficial bleached enamel; however, the action of the lips may show a similar effect.

This study examined the possible enamel alterations in terms of morphology and mechanical properties. Although no morphological or mechanical alterations resulted from bleaching treatments under *in situ* conditions, these two agents might induce other alterations in enamel structure that were not apparent in this study. After all, the neutral HP agents still hold oxidizing ability for whitening efficiency. This will be an interesting direction for us to pursue in a future study.

CONCLUSION

Within the limitations of the present study, the following conclusions were drawn: In-office bleaching agents with low pH values could induce enamel morphology alterations under *in vitro* conditions. The presence of natural HS could eliminate the demineralization effect caused by low pH.

Acknowledgements

All of the authors would like to sincerely appreciate the contribution of volunteers in this study who were undergraduate dental students of the School and Hospital of Stomatology, Wuhan University, China. This work was supported by

the Youth Chenguang Project of Science and Technology of Wuhan City (No. 200950431186), Natural Science Foundation of China (No. 81071190), the National Key Technology R&D Program of China during the 11th Five-Year Plan (No.2007-BAI18B05), and the Fundamental Research Funds for the Central Universities in 2010.

Conflict of Interest Declaration

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

(Accepted 20 September 2011)

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