Original Article

Effect of Bleaching Methods on Surface Roughness of Resin Impregnated Area of Tooth

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Abstract

The aim of this study was to evaluate the surface roughness of resin impregnated area of tooth before and after bleaching with different procedures. Buccal surfaces of extracted maxillary premolars were luted with orthodontic resin cement and polished until smooth with tungsten carbide bur. Initial surface roughness of the polished area was measured. Teeth were divided into 4 groups as follows: group 1) bleached with 10 % carbamide peroxide 8 hours per day (15 days), group 2) bleached with 20 % carbamide peroxide 8 hours per day (9 days), group 3) bleached with 40 % hydrogen peroxide 3 cycles (2 cycles at day 1 and 1 cycle at day 6) and group 4) bleached with 40 % hydrogen peroxide 2 cycles at day 1 and with 10 % carbamide peroxide 8 hours per day (9 days). Mean initial surface roughness between groups was not statistically different. After bleaching, the surface roughness was measured again. Data were analyzed using pair T-test and F-test one way ANOVA (p<0.05). Results revealed that all bleaching methods significantly increased the surface roughness of the resin impregnated areas. However, there was no significant difference between groups.

Keyword: Resin impregnated area, Surface roughness, Tooth bleaching

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Introduction

Tooth bleaching is a conservative treatment for whitening the tooth color. Currently, there are various approaches of tooth bleaching either in dental clinic or at patient's home with low and high concentrations of bleaching agents.^{1–3} Mechanism of vital tooth bleaching is based on hydrogen peroxide or chemical agents containing peroxides due to their high oxidizing capability. The process starts when hydrogen peroxide bursts into oxygen, water and

free radicals. These small radical molecules will diffuse through enamel and dentin to oxidize chromophores inside the tooth. Double bonds of the color molecules are broken into single bonds leading to brightened color.^{24,5} Carbamide peroxide is another tooth bleaching agent that decomposes to 30% hydrogen peroxide and 70% urea. Hence, 10% and 20% carbamide peroxide provide 3-3.5% and 6-7% of hydrogen peroxide, respectively.¹

Tooth surface morphology could be affected by bleaching process. Increasing surface roughness and decreasing surface hardness were reported in previous studies which used hydrogen peroxide and carbamide peroxide.¹⁻³ High concentration of bleaching agents induced more changes of the enamel surface.⁴ This surface alteration can lead to biofilm formation and tooth staining.⁶

In case of fixed orthodontic treatment, adhesive cement still remains on tooth surface after bracket is debonded. It will normally be polished out from the tooth surface, but impregnated cement layer approximately 30-50 microns is left in the enamel.⁷ Surface of this hybrid layer can be stained and appears as a spot on the tooth.^{8,9} Typically, this layer can finally be abraded out later by brushing. However, some orthodontic patients need to brighten their teeth immediately after the appliances are removed. This cement-enamel layer acts as a barrier, preventing the bleaching agent to diffuse inside the tooth.⁵⁻⁷ Bleaching on the resin impregnated area results in delayed color changes compared to sound tooth structure.¹⁰ Nantanapiboon and Maneenut¹⁰ used low- and high-concentration bleaching agents as well as several bleaching methods for resin impregnated tooth bleaching. They reported that prolongation of bleaching time was needed in resin impregnated site. The mismatched color between the resin impregnated area and the surrounding area was initially observed in the first week of bleaching and declined at the later weeks. Time used for blending the color of resin

impregnated area and the color of surrounding tooth depended on the bleaching agent concentration. The period of mismatched color was reduced by using high concentration of bleaching agents.

At present, there is no data about the effect of bleaching agent on resin impregnated area on tooth surface whether there will be less or more roughness after bleaching. Therefore, the aim of this study was to investigate whether different bleaching methods and different concentrations of bleaching agents could affect surface roughness of resin impregnated area of the tooth.

Materials and methods

Extracted human maxillary premolars were collected. Teeth were cleaned using dental scaler and polished with fine pumice slurry using a low-speed handpiece. They were inspected for signs of cracks, decay and restoration by stereomicroscope (Stereo Microscope SZ61, Olympus, Japan). Forty sound teeth were included into the experiment. The selected teeth were stored in 0.1% thymol solution at 37 degrees Celsius.

Teeth were mounted on acrylic resin blocks and individual silicone jig, 1 mm thick, was prepared. Circular hole, 6 mm in diameter, was made on the silicone jig at buccal surface of the tooth for locating the experimental area. Bleaching tray with 1 mm space at buccal aspect was also fabricated. Positioning of silicone jig and bleaching tray on each tooth could be repeated. (Fig. 1)

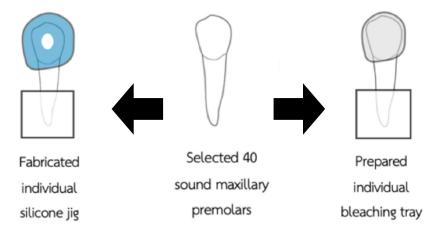


Figure 1 Silicone jig and bleaching tray preparation

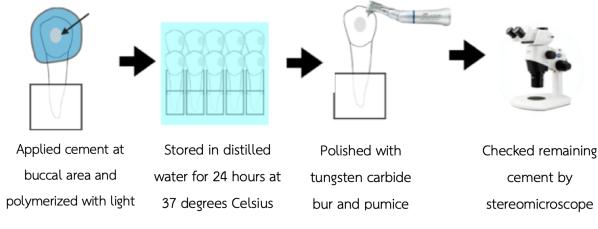


Figure 2 Resin Impregnated layer preparation

Resin Impregnated layer preparation (Fig. 2)

The silicone jig was placed on the tooth and buccal area in the hole was etched with 37% phosphoric acid for 30 seconds, rinsed with spray-water for 30 seconds, completely air-dried with air spray for 10 seconds, primed and luted with clear light-cured orthodontic adhesive cement (Transbond XT, 3M/Unitek, Monrovia, CA, USA) about 0.5 mm thick. The cement was pressed against tooth surface using a transparent cylindrical crystal, 6 mm in diameter and 2 mm in height. Light (1,100-1,330 mW/mm²) from light curing unit (DemiTM Plus, Kerr, USA) was applied through the crystal for 40 seconds. The cylindrical crystal and silicon jig were removed from the tooth and light from light curing unit was applied on cement for 20 seconds. Specimen was stored in distilled water at 37 degrees Celsius for 24 hours.

The on-surface cement polishing was carried out with a slow speed 30-fluted tungsten carbide bur (Shofu dental corporation, Japan) without water.¹¹ The bur was changed after polishing 5 specimens. The polishing was stopped when the cement was reduced to the same level of tooth surface and its smoothness was checked by dental explorer. The specimen was polished with nonfluoride pumice for 30 seconds and rinsed with 20 ml of distilled water. The resin impregnated area was confirmed using stereomicroscope (Stereo Microscope SZ61, Olympus, Japan) at X40 magnification. (Fig. 3)



Figure 3 Resin impregnated area

Initial surface roughness at the polished area was measured using a non-contact surface roughness tester (Infinite Focus SL, Alicona. Austria). Roughness average (Ra) value was calculated from 3 measurements. Surface texture measurement (Sa) value was also calculated. Teeth were divided into 4 groups, means initial surface roughness of which were not statistically different.

Bleaching protocols

Teeth in each group were subjected to 4 bleaching methods, according to the previous study results.¹⁰ (Table 1)

Table 1 Bleaching protocols

Group	Bleaching agent	Bleaching time	Teeth
1	Opalescence 10% carbamide peroxide, Ultradent, USA (Home-bleaching)	8 hours per day, 15 days	10
2	Opalescence 20% carbamide peroxide, Ultradent, USA (Home-bleaching)	8 hours per day, 9 days	10
3	Opalescence Boost 40% hydrogen peroxide, Ultradent, USA (In-office bleaching)	2 cycles at day 1 and repeated 1 cycle at day 6	10
4	Opalescence Boost 40% hydrogen peroxide, Ultradent, USA (In-office bleaching) and Opalescence 10% carbamide peroxide Ultradent, USA (Home-bleaching)	2 cycles of in-office bleaching at day 1, followed by 8-hour home bleaching per day for 9 days	10

For group 1 and 2, bleaching tray with gel was applied to the tooth for 8 hours per day for 15 days and 9 days, respectively. After 8-hour bleaching on each day, tray was removed, gel was rinsed out and tooth was stored in non-fluoride artificial saliva (Artificial saliva, Faculty of Dentistry, Chulalongkorn University, Thailand) at 37 degrees Celsius for 16 hours.

For group 3, the bleaching tray with gel was applied to the tooth for 2 cycles (20 minutes per cycle) at day 1.¹² The tray was removed, gel was rinsed out and the tooth was stored in non-fluoride artificial saliva at 37 degrees Celsius for 5 days. The other bleaching cycle was done at day 6. In group 4, 2 bleaching cycles (20 minutes per cycle) were done at day 1 followed by home bleaching for 8 hours per day for 9 days. After each bleaching session, the tray was removed, gel was rinsed out and the tooth was stored in non-fluoride artificial saliva at 37 degrees Celsius.

After bleaching procedure was done, the surface roughness of resin impregnated area in all groups was re-measured. The research proposal was approved by Ethics Committee of the Faculty of Dentistry, Chulalongkorn University. The study code was HREC-DCU2020-081.

Statistical Analysis

The data was analyzed by SPSS version 26.0 with a 95 % confidence interval to indicate the significant difference using paired *t*-test and F-test one way ANOVA.

Results

F-test one way ANOVA revealed that before bleaching, resin impregnated surface roughness (Ra and Sa values) of all groups were not significantly different (*p*>0.05) (Table 3). After bleaching, pair *T*-test revealed that surface roughness of all groups was significantly increased (Table 2).

However, F-test one way ANOVA showed that the increasing of roughness was not statistically different among groups (Table 3).

Surface roughness images revealed smooth resin impregnated surface of all groups before bleaching. The surfaces were rougher after bleaching (Fig. 4).

Material	Ra			Sa		
	Before bleaching	After bleaching	_ p - value	Before bleaching	After bleaching	_ p - value
	Mean (SD)	Mean (SD)		Mean (SD)	Mean (SD)	
Group 1	514.44 nm.	946.11 nm.	0.013*	985.89 nm ²	1526.54 nm ²	0.003*
10% Carbamide peroxide	(92.89)	(107.85)		(93.69)	(249.21)	

 Table 2
 Surface roughness, before and after bleaching

Table 2 Surface roughness, before and after bleaching (cont.)

Material		Ra	Sa			
	Before bleaching Mean (SD)	After bleaching Mean (SD)	_ p - value	Before bleaching Mean (SD)	After bleaching Mean (SD)	_ p - value
Group 2 20% Carbamide peroxide	532.16 nm. (61.07)	950.98 nm. (138.42)	0.025*	998.93 nm2 (89.27)	1602.47 nm ² (270.09)	0.041*
Group 3 40% Hydrogen peroxide	535.17 nm. (37.60)	937.75 nm. (187.67)	0.050*	1009.40 nm2 (76.46)	1492.93 nm ² (325.57)	0.047*
Group 4 40% Hydrogen peroxide follow by 10% Carbamide peroxide	514.19 nm. (60.77)	809.88 nm. (97.44)	0.011*	982.88 nm2 (88.30)	1384.48 nm ² (297.26)	0.031*

* Means of surface roughness before and after bleaching for each group

Table 3 Demonstrated the p-value of Ra and Sa before and after bleaching

	Ra		Sa		
	Before bleaching	After bleaching	Before bleaching	After bleaching	
	(p-value)	(p-value)	(p-value)	(p-value)	
Between groups	0.670	0.204	0.893	0.454	

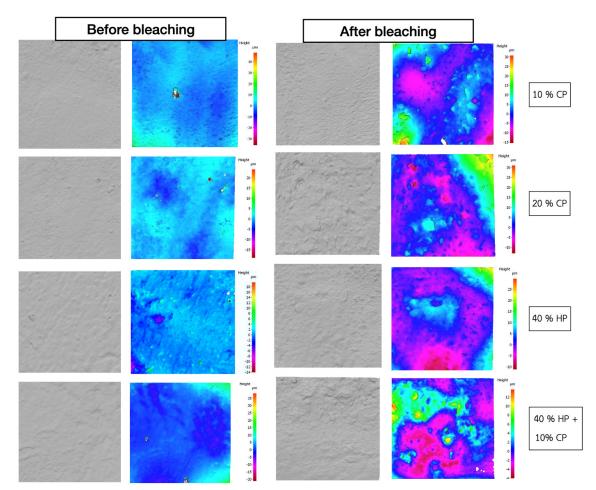


Figure 4 Represented images of resin impregnated surface area, before and after bleaching

Discussion

Using the non-contact surface roughness tester in this study could avoid the scratched surface of the specimen and could measure the roughness at the deep level. Apart from the scale of roughness average (Ra) and surface texture value (Sa) reported, 3D image of the surface could be generated. The principle of this tester is that laser light emits from the light source to the measured surface and reflects to the charge-coupled device camera to assess the roughness of the specimen.¹²

This present study found that using either 10% or 20% carbamide peroxide or 40% hydrogen peroxide or 40% hydrogen peroxide combined with 10% carbamide peroxide increased the roughness of resin impregnated surface. Without SEM picture of the polished surface, it could be speculated that after on-surface cement was polished to the level of enamel, the surface of this resin impregnated layer should consist of exposed enamel and resin. Orthodontic resin cement's components are similar to those of resin composite filling material. The matrix part comprises bisphenol A-glycidyl methacrylate (Bis-GMA), triethylene glycol dimethacrylate (TEGDMA) and camphorquinone. The fillers are inorganic silica 77 % by weight and also contain phosphorous hexafluoride.¹³ Reaction to bleaching of the surface may be similar to that of sound tooth structure and resin composite. Previous study revealed that the effect of both 10% CP and 40% HP increased the surface roughness of resin composite.¹⁴ This may be accounted for by that the peroxide containing compound decomposing from the bleaching agent oxidized the remaining double bonds of the polymer chains. According to the study of Durmer and colleagues¹⁵, they discovered that hydrogen peroxide reacted with not only the double bonds, but also the single bonds in resin composite polymer. This may cause resin matrix degradation and dissolution. This phenomenon may be increased in the present study due to the immersion of the tooth sample in artificial saliva for a period of time during bleaching. Water sorption of the resin impregnated layer could degrade the resin matrix.¹⁶ Moreover, free radicals derived from the peroxide compound affected at the resin matrix–filler interface resulting in the dislodgement of the filler. The stress occurred from the water absorption of resin composite was another factor for the dislodgement of resin the filler. On the other hand, Londono¹⁷ studied the effect of vital tooth bleaching on the solubility and roughness of dualcured and self–adhesive resin cement (Rely X ARC and Rely X Unicem) for crown cementation (Rely X ARC and Rely X Unicem) and found that the in-office bleaching agent, 38% hydrogen peroxide, and the home bleaching agent, 20% carbamide peroxide, did not affect the resin cement surface roughness. The authors explained that the self-cured resin cement would create the rigid and stable network so the oxidation reaction of bleaching agent could not proceed.

Bleaching agents have the capability to increase surface roughness of the intact enamel. Previous study¹⁸ revealed that higher concentration of the bleaching agent created more roughness after using 35% carbamide peroxide. The chemical agents in bleaching gel could lead to a loss of calcium and alter the Ca to P ratio on the tooth surface.¹⁹ Bleaching agents might also enlarge gapsbetween enamel prisms, creating invasive tract to the surface. However, the damages have not been detected macroscopically or clinically visible. In contrast, 10% carbamide peroxide agent does not affect the enamel surface.

Tooth bleaching agents normally have neutral pH.²⁰ The differences among them are only the chemicals and concentrations that are used for initiating the free radicals. Duration of the bleaching procedures in this present study was from the result of the previous study¹⁰, which was different for each group. According to the manufacturers' recommendation, the high concentration of bleaching agent, such as 40% hydrogen peroxide, has less tooth-contact time than the lower concentrations, i.e. 10% and 20% carbamide peroxide. The increase in roughness (Sa and Ra) of all groups in this study were not statistically significant (Table 3). This finding implies that the total amount of free radicals released from each group was comparable and

affects the surface roughness in the same rate. Nevertheless, if the contact time of all the groups had been the same, some differences would have occurred.

This study used the customized tray to carry the bleaching agent for the in-office bleaching, called "Sealed in office bleaching". This method was introduced to control the amount of peroxide in the oral situation²¹ as in this study to control the amount of gel for each tooth. Previous study showed that using a customized tray with 35% hydrogen peroxide in the in-office method did not affect the level of sensitivity reported by the patient during the procedures. However, this technique may increase the chances and intensity of tooth sensitivity for the first 24 hours after the completion of the procedure.²¹

Nowadays, many bleaching approaches have been provided to achieve the most effective way of tooth whitening. The combination between in-office and homebleaching methods (group 4) is recommended. Tooth whitening starts with in-office bleaching followed by continuous homebleaching. Previous studies found that the combination of both methods reduced the risk of tooth sensitivity and gingival irritation. Moreover, the final whitened tooth appeared faster than using single individual mean.²²

The present study did not evaluate the amount of peroxide penetrating into the tooth. The previous study by Benetti and colleagues.²³ revealed that the bleaching agents could enter the pulp through enamel and dentin after bleaching for 60 minutes. However, there was no report of penetration of bleaching agents through both resin composite and dentin into deeper area of the tooth. Since this resin impregnated layer is hybrid layer which has different properties from enamel and resin cement. Therefore, the outcome of bleaching on this layer may not be similar to that on the enamel or resin composite

Tooth bleaching process contributed to tooth surface changes²⁴⁻²⁶ and altered the surface characteristic of resin composite and glass ionomer cement.²⁷ From the present study, the surface roughness of resin impregnated area was increased after bleaching which may lead to more biofilm and bacterial accumulation. Previous studies showed

that the rougher surface of resin composite and resinmodified glass ionomer cement promoted cariogenic bacteria (i.e., *Streptococcus mutans* and *Streptococcus sanguinis*) biofilm adhesion after tooth bleaching. All of these bacteria are the pathogenic bacteria that may cause dental caries.^{14,28,29}

In a clinical situation, the use of high concentration of hydrogen peroxide may cause the patient discomfort or tooth sensitivity during bleaching procedure. This side effect seems to be higher in hydrogen peroxide treated teeth compared to carbamide peroxide treated teeth due to the ability of hydrogen peroxide in producing more free radicals.^{1,3-5} The result of the present study showed that there was no significant difference of roughness among groups of different methods used as shown in figure 4. Therefore, the use of a non-aggressive bleaching agent such as 10% carbamide peroxide seems to be more appropriate regarding tooth sensitivity. Moreover, it is recommended to re-polish the resin-impregnated surface after bleaching with fine and superfine polishing discs to decrease the roughness of the surface.

Conclusion

The surface roughness of resin impregnated area of tooth is influenced by bleaching procedures. All bleaching methods induce significantly increased roughness, but the final roughness is not significantly different between methods.

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Reference

1. Kwon SR, Wertz PW. Review of the mechanism of tooth whitening. *J Esthet Restor Dent* 2015;27(5):240-57. Kihn PW. Vital Tooth Whitening. *Dent Clin North Am* 2007;51(2):
 319-31, viii.

 Dahl JE, Pallesen U. Tooth bleaching - A critical review of the biological aspects. *Crit Rev Oral Biol Med* 2003;14(14):292-304.
 Goldberg M, Grootveld M, Lynch E. Undesirable and adverse effects of tooth-whitening products: A review. *Clin Oral Investig* 2010;14(1):1-10.

5. Tredwin CJ, Naik S, Lewis NJ, Scully Cbe C. Hydrogen peroxide tooth-whitening (bleaching) products: Review of adverse effects and safety issues. *Br Dent J* 2006.200(7):371-6.

6. Chen Y-H, Yang S, Hong D-W, Attin T, Yu H. Short-term effects of stain-causing beverages on tooth bleaching: A randomized controlled clinical trial. *J Dent* 2020;95:103318.

7. Kinch AP, Taylor H, Warltler R, Oliver RG, Newcombe RG. A clinical study of amount of adhesive remaining on enamel after debonding, comparing etch times of 15 and 60 seconds. *Am J Orthod Dentofac Orthop* 1989;95(5):415-21.

8. Hong YH, Lew KKK. Quantitative and qualitative assessment of enamel surface following five composite removal methods after bracket debonding. *Eur J Orthod* 1995;17(2):121-8.

9. Tüfekçi E, Merrill TE, Pintado MR, Beyer JP, Brantley WA, Campbell PM. Enamel loss associated with orthodontic adhesive removal on teeth with white spot lesions: An *in vitro* study. *Am J Orthod Dentofac Orthop* 2004;125(6):733-9.

 Nantanapiboon D, Maneenut C. Effect of bleaching methods on resin impregnated tooth. *J Dent Assoc Thail* 2019;69(1):53–9.
 Howell S, Weekes WT. An electron microscopic evaluation of the enamel surface subsequent to various debonding procedures. *Aust Dent J* 1990;35(3):245–52.

12. Paepegaey A-M, Barker ML, Bartlett DW, Mistry M, West NX, Hellin N, *et al.* Measuring enamel erosion: a comparative study of contact profilometry, non-contact profilometry and confocal laser scanning microscopy. *Dent Mater* 2013;29(12):1265–72.

13. Gama ACS, Moraes AG de V, Yamasaki LC, Loguercio AD, Carvalho CN, Bauer J. Properties of composite materials used for bracket bonding. *Braz Dent J* 2013;24(3):279–83.

14. Wongpraparatana I, Matangkasombut O, Thanyasrisung P, Panich M. Effect of vital tooth bleaching on surface roughness and streptococcal biofilm formation on direct tooth-colored restorative materials. *Oper Dent* 2018;43(1):51-59.

Durner J, Stojanovic M, Urcan E, Spahl W, Haertel U, Hickel R, *et al.* Effect of hydrogen peroxide on the three-dimensional polymer network in composites. *Dent Mater* 2011;27(6):573–80.
 Ortengren U, Wellendorf H, Karlsson S, Ruyter IE. Water sorption and solubility of dental composites and identification of

monomers released in an aqueous environment. *J Oral Rehabil* 2001;28(12):1106–15.

17. Londono J, Abreu A, Nelson S, Hernandez J, Torres C, Mettenburg D, *et al.* Effect of Vital Tooth Bleaching on Solubility and Roughness of Dental Cements. *J Prosthet Dent* 2009;102(3):148-54.

18. Moraes RR, Marimon JLM, Schneider LFJ, Correr Sobrinho L, Camacho GB, Bueno M. Carbamide peroxide bleaching agents: effects on surface roughness of enamel, composite and porcelain. *Clin Oral Investig* 2006;10(1):23–8.

19. Potocnik I, Kosec L, Gaspersic D. Effect of 10% carbamide peroxide bleaching gel on enamel microhardness, microstructure, and mineral content. *J Endod* 2000;26(4):203–6.

20. Basting RT, Amaral FLB, França FMG, Flório FM. Clinical comparative study of the effectiveness of and tooth sensitivity to 10% and 20% carbamide peroxide home-use and 35% and 38% hydrogen peroxide in-office bleaching materials containing desensitizing agents. *Oper Dent* 2012;37(5):464–73.

Correa AC, Santana TR, Nahsan FP, Loguercio AD, Faria-E-Silva
 AL. The Impact of a Customized Tray on In-Office Bleaching Tooth
 Sensitivity: A Randomized Clinical Trial. *Oper Dent* 2016;41(1):15–22.
 Reis A, Kossatz S, Martins GC, Loguercio AD. Efficacy of and
 effect on tooth sensitivity of in-office bleaching gel concentrations:
 A randomized clinical trial. *Oper Dent* 2013;38(4):386-93.

23. Benetti AR, Valera MC, Mancini MNG, Miranda CB, Balducci I. *In vitro* penetration of bleaching agents into the pulp chamber. *Int Endod J* 2004;37(2):120–4.

24. Basting RT, Rodrigues Júnior AL, Serra MC. The effect of 10% carbamide peroxide bleaching material on microhardness of sound and demineralized enamel and dentin in situ. *Oper Dent* 2001; 26(6):531-9.

25. Melo CF de M, Manfroi FB, Spohr AM. Microhardness and roughness of enamel bleached with 10% carbamide peroxide and brushed with different toothpastes: an in situ study. *J Int oral Heal* 2014;6(4):18-24.

26. Smidt A, Weller D, Roman I, Gedalia I. Effect of bleaching agents on microhardness and surface morphology of tooth enamel. *Am J Dent* 1998;11(1):83-85.

27. Turker ŞB, Biskin T. Effect of three bleaching agents on the surface properties of three different esthetic restorative materials. *J Prosthet Dent* 2003;89(5):466-73.

28. Marsh PD. Dental plaque as a microbial biofilm. *Caries Res* 2004;38(3):204–11.

29. Marsh PD. Dental plaque as a biofilm and a microbial community – implications for health and disease. *BMC Oral Health* 2006;6(1):S14.