Programa de Pós-Graduação S*tricto Sensu* em Odontologia – Nível Mestrado
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BIANCA MEDEIROS

EFEITO DA DEGRADAÇÃO BIOLÓGICA SEGUIDA DA ESCOVAÇÃO SOBRE PROPRIEDADES DE SUPERFÍCIE DE MATERIAIS RESTAURADORES ESTÉTICOS

EVALUATION OF THE SURFACE PROPERTIES OF AESTHETIC RESTORATIVE MATERIALS EXPOSED TO BIOLOGICAL DEGRADATION FOLLOWED BY TOOTHBRUSHING

UNIVERSIDADE ESTADUAL DO OESTE DO PARANÁ – UNIOESTE
CENTRO DE CIÊNCIAS BIOLÓGICAS E DA SAÚDE
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CASCAVEL – PR
2015

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Dissertação apresentada ao Programa de Pós-Graduação *Stricto Sensu* em Odontologia – Nível Mestrado, do Centro de Ciências Biológicas e da Saúde, da Universidade Estadual do Oeste do Paraná, para obtenção do título de Mestre em Odontologia.

Orientadora: Profa. Dra. Fabiana Scarparo Naufel

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A Comissão Julgadora dos trabalhos de Defesa da Dissertação de Mestrado, em sessão pública, realizada em 04 de janeiro de 2015, considerou a candidata Bianca Medeiros aprovada.

Prof.ª Dr.ª Fabiana Scarparo Naufel

Prof.ª Dr.ª Vera Lucia Schmitt

Prof.ª Dr.ª Alessandra Reis

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RESUMO

Materiais restauradores estéticos devem resistir às adversidades do meio bucal, sofrendo mínima degradação. Este estudo avaliou a rugosidade (Ra), dureza Knoop (KHN) e alteração de cor (ΔE) de materiais restauradores estéticos submetidos ao contato com biofilme de *Streptococcus mutans* associado a abrasão gerada pela escovação. Foram confeccionados 10 discos (8 mm de diâmetro por 2 mm de espessura) de cada material (Filtek Z350, resina composta nanoparticulada; Empress Direct, resina composta nanohíbrida e IPS e.Max, cerâmica). Os discos de cerâmica receberam aplicação de glaze. Os compósitos foram armazenados em umidade relativa 100%, por 24 h, após, realizou-se o polimento dos compósitos com discos abrasivos sequenciais. Então, todos os espécimes foram armazenados em umidade relativa 100% a 37ºC por 24 h, sendo assim, avaliadas as propriedades de superfície iniciais. Os espécimes foram esterilizados em óxido de etileno; e submetidos a degradação biológica através da inoculação de 25 µL de *Streptococcus mutans* com densidade ótica (DO) padronizada, mantido por 2 h sobre os discos. Os espécimes foram imersos em meio brain heart infusion (BHI) com 1% de sacarose, o qual foi trocado a cada 48 h. Após 7 dias, todos os discos foram lavados em ultra-som, e avaliadas as propriedades de superfície novamente. Por fim, os espécimes foram submetidos a degradação mecânica, sendo fixos a um dispositivo de escovação e desgastados através das cerdas dentais, via dentifrício, e, após esse processo, foram mensuradas, mais uma vez, as propriedades de superfície. Os dados foram analisados pelos testes Proc-Mixed e Tukey ($\alpha = 0,05$). Inicialmente e.Max apresentou maior Ra e KHN; após a degradação biológica os compósitos tiveram aumento de Ra, porém, a KHN não se alterou; já posteriormente a degradação mecânica, Empress teve sua Ra diminuída e a KHN aumentou para o Z350, e todos tiveram aumento da luminosidade. Os resultados permitem concluir que, quando expostos ao biofilme de *S. mutans* e à abrasão por escovação, a cerâmica sofre mínima degradação e os compósitos sofrem degradação variável, dependendo da sua composição.

PALAVRAS CHAVES

Biofilmes; Propriedades de superfície; Materiais dentários.

ABSTRACT

Aesthetic restorative materials must withstand the adversities of the oral environment, suffering minimal degradation. This study evaluated the roughness (Ra), Knoop hardness (KHN) and change of color (ΔE) of esthetic restorative materials subjected to contact with biofilm of *Streptococcus mutans* associated with abrasion generated by brushing. 10 disks (8 mm diameter; 2 mm deep) of each material were prepared (Filtek Z350, composite nanoparticle; Empress Direct, composite nanohybrid and IPS e.Max, ceramic). The ceramic disks received application of glaze. The composites were stored in 100% relative humidity, after 24 h, the composites were polished with sequential abrasive discs. Then, all specimens were stored in 100% relative humidity at 37°C for 24 hours, and so, evaluated the properties of initial surface. The specimens were sterilized with ethylene oxide; and after undergoing biological degradation, by 25 µL *Streptococcus mutans* inoculum with optical density (OD) standard, maintained by 2 h on discs. The specimens were immersed in brain-heart infusion (BHI) medium with 1% sucrose, which was changed every 48 h. After 7 days, all disks were washed in ultrasound, and evaluated the surface properties again. Finally, the specimens were subjected to a mechanical degradation, being fixed to a device of brushing and worn through for dental bristles, with toothpaste, and after this process, were measured, once again, the surface properties. Data were analyzed by Proc-Mixed and Tukey ($\alpha = 0.05$). Initially e.Max showed higher Ra and KHN; after biological degradation, the composites showed increased Ra, but KHN has not changed; later the mechanical degradation Empress had its Ra decreased and KHN increased to Z350, and all had lightness increased. The results suggest that when exposed to *Streptococcus mutans* biofilm and toothbrush abrasion, ceramic undergoes minimal degradation; the nanoparticulate composite promoted increased roughness and composites exhibit variable degradation, depending on the composition of the material.

KEYWORDS

Biofilms; Surface properties; Dental materials.

LIST OF ABBREVIATIONS

º	Graus
ºC	Graus Celsius
>	Greater
µL	MicroLiter
µm	Micrometer
%	Percent
ΔE	Variation of color
BHI	Brain-heart infusion
CFU/mL	Colony forming unit/miliLiter
CO_2	Carbon dioxide
H	Hours
KHN	Knoop hardness
L*	Lightness
Min	Minutes
mL	MiliLiter
mm	Milimiter
mN	MiliNewton
Nm	Nanometer
OD	Optical density
Ra	Surface roughness
S	seconds
Wt	Weight
Wt/vol	Weight/Volume

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SUMÁRIO

1 INTRODUCTION

All restorative materials are susceptible to degradation. Degradation of biomaterials may be caused by low pH due to cariogenic biofilm, consumption of acidic drinks or foodstuffs, and toothbrushes.[1-4]

Toothbrushing is the most used and efficient mechanical method for removing dental biofilm from all accessible tooth surfaces.[5] Published studies have shown that this method may cause tooth and resin composite abrasion.[1,6] This degradation process may lead to several drawbacks, such as an increase in wear and surface roughness, softening, and a decrease in hardness of dental materials.[7-9] Over time, intra-oral degradation also interferes with the fracture strength of the material, culminating in lower durability of the restoration in the long term.[10] Surface texture, gloss, and color are also included among the important characteristics that determine the aesthetic effect of these composite resin restorations, and they are also influenced by the intra-oral surroundings.[11,12]

It's important know about this process, because the search for dental esthetics has been one of the main reasons why the patient seeks the dentist. Thus, the need for tooth-colored fillings has increased more, rendering the use of metal restorations and dental amalgam fillings or cast metal, dwindling, unlike the use of aesthetic materials such as composite resin and ceramic which have been increasingly used.[13] The most aesthetic materials to restore the function of damaged teeth are composite resins and ceramics.

Ceramics are considered the most inert of all dental materials used for restorations, composed of elements metal (aluminum, calcium, lithium, magnesium, potassium, sodium, lanthanum, tin, titanium and zirconium) and substances not metal (silicon, boron, fluorine and oxygen) and characterized two phases: a crystalline phase surrounded by a layer vitreous.[14] So far, little information about surface degradation followed by biofilm is available in the literature. Some studies have evaluated the interaction between biofilm and ceramic, but they verified only the biofilm characteristics instead of the biodegradation produced on material surfaces.[15,16]

Resin-based composites are currently the most-used material in the field of restorative dentistry. Basically, these materials are composed of three chemically different components: a polymeric matrix of dimethacrylate monomers, filler particles (dispersed phase), and an organosilane which is a coupling agent that bonds the fillers to the polymeric matrix.[17]

In this context, nanotechnology, consisting of nanofillers, has emerged in the dental market.[18] This technology came with the intention of improving the electrical, chemical,

mechanical, and optical properties of restorative materials with advantages such as less toothbrush abrasion, greater hardness, and better translucency, polish, gloss, and opacity options being used for restorations of anterior and posterior teeth; as a result, studies have been done to prove these characteristics.[2,19-21]

A nanohybrid composite IPS Empress Direct promises similar aesthetics to those of ceramics beyond the advantages of easy handling of composite resin (Ivoclar Vivadent).

Therefore, it becomes interesting to compare the IPS Empress Direct composite with a 100% nanoparticulate resin such as Z350, as well as with the ceramic (IPS e.Max), considering the material that undergoes minimal degradation.

Thus, the aim of this study was to test the hypothesis that aesthetic restorative materials submitted to *Streptococcus mutans* biofilm associated with brushing abrasion would differ in surface stability to degradation, depending on their composition.

2 MATERIALS AND METHODS

2.1 Specimen Preparation

10 specimens of each biomaterial tested (described in Table 1) were fabricated using silicon molds (Express 3M ESPE, St. Paul, Minn, USA) of 8 mm in diameter and 2 mm deep (Fig. 1), with the exception of the ceramic. The materials were placed in the mold for the incremental technique by one operator and covered by polyester strips with a glass slide under a load of 454 g for 45 s to obtain a flat surface. All specimens were polymerized with a curing light unit (Elipar Freelight, 3M ESPE, St. Paul, MN, USA) for 40 s (Fig. 2). The light intensity of the curing device was checked with a curing light meter (Hilux Dental Curing Light Meter, Benlioglu Dental Inc., Demetron, Ankara, Turkey). After storage for 24 h in 100% relative humidity at 37°C, the composites were polished with sequential abrasive discs (Soflex Pop-On, 3M ESPE, St. Paul, MN, USA).

For the ceramic, specimens were fabricated with the same dimensions of the composites, in a prosthetic laboratory by using the pressing process in an oven (Programat P500–Ivoclar Vivadent, Schaan, Liechtenstein) and received glaze application.

Then, all specimens were stored in 100% relative humidity at 37°C for 24 h for evaluation of the baseline properties.

Table 1. Tested materials

Material	Composition	Color	Batch #
Filtek Z350 XT (3M ESPE)	Bis-GMA (1-10 wt%); UDMA (1-10 wt%); TEGDMA (< 5 wt%); Bis-EMA (1-10 wt%); PEGMA (< 5 wt%) Silica, zirconia, zirconia/silica (78.5 wt%)	A3E	1124300109
IPS Empress Direct (Ivoclar Vivadent)	UDMA (10-<20 wt%); TEGDMA (3-<5 wt%); Bis-GMA (2.5-<3 wt%) Barium glass, ytterbium trifluoride, mixed oxide, silicon dioxide and copolymer (77.5-79 wt%) Additives, catalysts, stabilizers and pigments (<1.0 wt%)	A3E	N32078
IPS e.Max (Ivoclar Vivadent)	SiO_2, Li_2O, K_2O, MgO, ZnO, Al_2O_3, P_2O_5 and others oxides	A3E	P82207

2.2 Surface Roughness Measurements

Surface roughness (Ra) measurements were measured in a rugosimeter (Surfcorder SE 1700, Kosaka, Tokyo, Japan). At a constant speed of 0.5 mm/s with a load of 0.7 mN (Fig. 3). The cut-off value was set at 0.25 mm to maximize filtration of surface waviness. Ra values for each specimen were taken across the diameter over a standard length of 1.25 mm. The mean surface roughness values (μm) of the specimens were obtained from three successive measurements of the center of each disk, in different directions (45°). A calibration was done periodically to check the performance of the surface roughness-measuring instrument.

2.3 Hardness Measurements

Three Knoop hardness (KHN) indentations were made on the specimen surface under a load of 50 g for a 10 s (HMV-2, Shimadzu, Tokyo, Japan) (Fig. 4). The Knoop hardness number for each specimen was recorded as the average of the three readings.

2.4 Color Measurement

The readings were performed using a spectrophotometer (CM-700d, Konica Minolta, Osaka, Japan). Initially its calibrated ambient light in a light cabin, (GTI Mini Matcher MM1e,

GTI Graphic Technology Inc., Newburgh, NY, USA), the specimens were positioned in a sample carrier to obtain the baseline readings (Fig. 5). The parameters L*, a*, and b* from the color space, referred to as CIELAB (L*, a*, b*) was recorded. L* indicates lightness (L + = lightness and L - = darkness), the a* coordinate represents the red/green range (a* + = redness and a* - = greenness), and the b* coordinate represents the yellow/blue range (b* + = yellowness and b* - = blueness). The L*a*b* system allows the numeric definition of a color as well as the difference between two colors using the following formula: $\Delta E = [(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2]1/2$. The data acquisition was performed by a microcomputer using On Color QC Lite software (Konica Minolta, Osaka, Japan).

2.5 Biofilm Growth – Biological degradation

After surface roughness, hardness, and color measurements, the specimens were sterilized for 4 h in ethylene oxide chamber (Ferlex, São Paulo, SP, Brazil). A *Streptococcus mutans* (UA 159) strain was obtained from the culture of the Department of Microbiology and Immunology, Piracicaba Dental School, University of Campinas. To prepare the inoculums, *S. mutans* was first grown on mitis-salivaris agar plates (Difco Laboratories, Sparks, MI, USA) at 37°C for 24 h in an environment supplemented with 5% CO_2. Subsequently, single colonies were inoculated into 5 mL of brain-heart infusion (BHI) broth (Difco Laboratories, Detroit, MI, USA) and incubated at 37º C for 24 h. The specimens were exposed under static conditions to 25 µL of *S. mutans* inoculums adjusted to an optical density of 0.6 at 550 nm (approximately $8x10^{11}$ CFU/mL) (Fig. 6), after 2 hours at room temperature, the non-adhering cells were removed by washing twice with 0.9% NaCl solution (saline).

A single material disk was placed in each well of polystyrene plates (Nunc multidish 48-well, Sigma, St. Louis, MO, USA) with 2 mL of sterile, fresh BHI broth with the addition of 1% sucrose (wt/vol). The bacterial accumulation occurred at 37ºC in an environment supplemented with 5% CO_2, developing 7-day-old biofilms. The medium was renewed at every 48 h. At the end of the experimental period, specimens were ultrasonically (UNIQUE 1400, Indaiatuba, SP, Brazil) washed for 10 minutes; and after that the measurements were repeated.

2.6 Three-body abrasion test – Mechanical degradadation

After biological degradation, the toothbrushing test was conducted at 250 cycles/min for 30,000 cycles with a 200 g load. The Oral B Pró Saúde dentifrice (Procter & Gamble, São Paulo, SP, Brazil) was diluted in distilled water (1:2) and used as an abrasive third body (Fig. 7). Specimens were washed in an ultrasonic bath for 10 min and gently dried with absorbent

paper. Then, three final surface roughness readings were taken from each specimen in the opposite direction to that of the toothbrushing movement; Knoop hardness and color were also evaluated as previously reported.

2.7 Statistical Analysis

The equality of variances and normal distribution of the data were verified using Shapiro-Wilk test, than the results were submitted to Proc-Mixed and Tukey's tests ($\alpha = 5\%$), as the specimens used for the mechanical degradation were the same ones used previously for the biological biodegradation procedure. Also, the correlation between all the studied variables was evaluated.

3 RESULTS

Table 2. Means (standard deviations) of surface roughness (Ra) (µm) for the different experimental conditions.

Materials	Baseline	Biological biodegradation	Mechanical degradation
Z350	0.26 (0.09) Bb	1.51 (1.08) Ab	1.48 (0.70) Aab
Empress	0.24 (0.07) Bb	2.71 (0.43) Aa	0.86 (0.34) Bb
e.Max	2.60 (0.71) ABa	3.26 (0.98) Aa	2.20 (0.79) Ba

Means followed by different capital letters in the same line and small letters in the same column were significantly different ($p < 0.05$).

There was significant difference among materials studied ($p < 0.0001$) and between the degradation methods (baseline/biological biodegradation/mechanical degradation; $p < 0.0001$).

From the materials evaluated, e.Max showed the highest roughness at baseline, and the composite resins Empress direct and Z350 showed very low and similar roughness.

After biological degradation both composite resins showed significantly increased roughness, and e.Max remained similar to baseline. And compared to material, Z350 showed the lowest roughness.

After mechanical degradation the roughness reduced to baseline values to Empress direct and e.Max the Z350 remained similar after biological degradadation (presented in Table 2).

Table 3. Means (standard deviations) of the Knoop hardness (KHN) for the different experimental conditions.

Materials	Baseline	Biological biodegradation	Mechanical degradation
Z350	62.1 (24.0) Bb	51.6 (15.38) Bc	82.438 (17.8) Ab
Empress	82.2 (15.8) Ab	80.4 (13.5) Ab	106.2 (16.7) Ab
e.Max	811.7 (139.9) Aa	656.8 (105.6) Aa	757.8 (151.1) Aa

Means followed by different capital letters in the same line and small letters in the same column were significantly different ($p < 0.05$).

Initially, the composite resins Empress direct and Z350 showed very low and similar hardness.

After biological degradation there was no statistical difference between all materials, though Z350 presented the lowest values.

After mechanical degradation resulted in hardness increases for Z350 and Empress remained similar to biological degradation.

In all conditions, e.Max showed the highest hardness values and no change was observed in ceramic during three times. (presented in Table 3).

Table 4. Means (standard deviations) of color change (ΔE) for the different experimental conditions.

Material	Biological biodegradation	Mechanical degradation
Z350	2.8 (1.0) a	1.9 (0.5) a
Empress	2.1 (0.5) b	1.2 (0.4) a
e.Max	3.0 (0.6) a	1.7 (1.1) a

Groups denoted by the different letter represent significant difference ($p<0.05$).

After biodegradation, the Empress composite showed the lowest ΔE. There was no statistical difference between the composites after the mechanical degradation (presented in Table 4).

Table 5. Means (standard deviation) of lightness (L*) for the different experimental conditions.

Material	Baseline	Biological biodegradation	Mechanical degradation
Z350	72.96 (0.23) Ba	72.78 (0.69) Ba	73.50 (0.29) Aa
Empress	68.59 (0.53) Ba	71.81 (0.71) Bb	72.50 (0.51) Ab
e.Max	72.60 (0.53) Ba	73.18 (0.55) Ba	73.30 (0.51) Aab

Means followed by different capital letters in the same line and small letters in the same column were significantly different ($p < 0.05$).

The lightness of the composites and ceramic was affected after mechanical degradation, produced an increase in lightness compared to the two methods (baseline and biological degradation) (presented in Table 5).

No significant correlation was observed between any pair of the properties evaluated ($p > 0.05$).

4 DISCUSSION

Aesthetic restorative materials are prone to a gradual degradation process in the oral cavity due to pH changes (chemical or bacterial action), temperature, chewing, and brushing, depending on the composition of the restorative material. [19,20,22-24]

The results of this study showed that the composites showed similar average roughness after polishing. After biological degradation the composites with different roughness variations, that depend on the degree of conversion and hydrolytic stability of the polymer matrix.[20,21] According to Sarkar, these changes are due to absorption and diffusion of water and organic acids from the bacterial metabolism, internal resin matrix, interfaces between the inorganic particles, pores, and other defects.[25] The greatest increase in the roughness of the composite Empress compared to Z350 can be attributed to the fact that the second one is 100% nanoparticulate, with less interstitial spacing of the matrix, which decreases its hydrolysis, in addition, we should mention the presence of Bis-EMA, hydrophobic monomer, which favors the hydrolytic stability.[17,19,21,26,27]

But the hardness remained statistically similar to the initial results; the presence of TEGDMA monomer is justified in both composites which increases the degree of conversion, reducing leach and softening.[14,27] Hardness, becomes an important parameter to measure the performance of materials in the oral environment, correlating to the resistance to compression and abrasion, and indirectly reflects the rate of polymerization of the material.[8]

Materials with decreased hardness has reduced longevity and may require early replacement of the restoration.[8,28]

The hardness of the Z350 composite was lower than the Empress in the three degradation methods, this can be attributed to differences in size and distribution of the charged particles of these materials;[9,29] after biological biodegradation, the difference was statistically significant and, beyond the aforementioned factor, it can be speculated that this is the association of the consequent hydrolysis of the polymeric matrix to the inorganic framework differences of the studied composites.[29-31]

The nanoparticulate composite may be prone to absorbing liquids because of the greater contact area load-matrix, and this interface is more susceptible to fluid accumulation in the bacterial biofilm, or alternatively, the spaces resulting from the presence of imperfect engagement of charged particles in the polymeric matrix. Spaces or “microvoids” in the polymeric matrix can increase retention of acids and thereby increase the degradation of Z350.[29]

In the oral cavity, the deleterious effects of biodegradation are generally associated with toothbrush abrasion, for the abrasiveness of dentifrice along with the toothbrush may promote the displacement of charged particles, which is directly proportional to the size of these effects.[1,2,32]

Empress composite showed decreased roughness after mechanical degradation, returning to average values statistically equivalent to the initial, this may be due to the movement of larger particles, which weakens the softened matrix and enhances the abraded mass of the polymer.[17,19,33] Z350 composite remained similar roughness to that observed after biological degradation, with interstitial space in the polymeric matrix, showed a higher abrasion resistance. Although no statistically significant differences were observed in Empress, both composites exhibited greater-than-initial roughness, which can be attributed to the effect of the bristles of the toothbrush. [33]

The hardness of the composites increased in both, and this increase was significant for Z350, resulting in similar hardness to the initial phase. This can be attributed to the process of maturation or late polymerization of the composite[34].

The color stability and lightness, important properties of aesthetic restorative materials, are influenced by various factors such as the composition of the inorganic portion, diet, habits, or even the organic matrix. The sensitivity of the human eye to detect color variation translates to ΔE> 3.3; thus the color changes were imperceptible to human sensitivity.[28,35] However, analysis of the CIELAB color scale coordinates (L *, a *, and b *) showed significant changes in the values of L *.

After the abrading process there were increases in the lightness for all materials studied, probably due to scratches caused by the abrasive process, resulting in less smooth surfaces, where the lightness is the ability of the material to reflect direct light and is closely related to the surface characteristics of the material, ranging from light (100) to dark (0).[12,33,36] Because the specimens were not exposed to any coloring agent to be standardized and there was standardization of the thickness of the specimens, the optical changes occurring reflect physical and chemical reactions: i) internal—such as hydrolysis or ii) surface—such as increased roughness, as these affect the lightness by changes in the refractive index and reflection, respectively.[35]

So far the ceramic, after polish showed higher roughness, which is due to surface irregularities of the resulting glazing process. After biological biodegradation, there was no significant variation of surface roughness of the ceramic, which may be due to the stability of the material, as it's considered the most inert dental material[14.] These results are in agreement with the study of Padovani et al.[4] The final roughness in ceramic was comparable to the original, in agreement with studies evaluating resistance to toothbrush abrasion.[12,16,37]

But in all degradation methods showed higher hardness than the composites studied, which is due to its glassy character, as in the sintering process there is coalescence of the particles and higher solid density.[4,14]

Restorative materials are constantly exposed to mechanical weathering and the biological and chemical environment of the mouth; they may affect to a greater or lesser extent the surface properties of restorative materials, and these are fundamental for aesthetics and clinical longevity of restorations. Under the experimental conditions described, the results showed that the degradation process associated with toothbrush abrasion promoted increased roughness of nanoparticulate composite; the nanohybrid composite exhibited less variations in roughness and hardness that of the nanoparticle; and ceramic materials were more stable and resistant to degradation in the oral environment.

5 REFERENCES

1. Voltarelli FR, Santos-Daroz CB, Alves MC, Cavalcanti AN, Marchi GM. Effect of chemical degradation followed by toothbrushing on the surface roughness of restorative composites. *J. Appl. Oral Sci.* 2010;18(6):585-590.

2. de Paula AB, Fúcio SBP, Ambrosano GMB, Alonso RCB, Sardi JCO, Puppin-Rontani RM. Biodegradation and abrasive wear of nano restorative materials. *Oper. dent.* 2011;36(6):670-677.

3. Flausino JS, Soares PBF, Carvalho VF, Magalhães D, da Silva WM, Costa HL, Soares CJ. Biofilm formation on different materials for tooth restoration: analysis of surface characteristics. *J Mater Sci.* 2014:49(1):6820–6829.

4. Padovani GC, Fúcio SBP, Ambrosano GMB, Sinhoreti MAC, Puppin-Rontani RM. *In situ* surface biodegradation of restorative materials. *Oper. dent.* 2014;39(2):in press.

5. Srivastava N, Vasishat A, Gupta G, Rana V. A Comparative evaluation of efficacy of different teaching methods of tooth brushing in children contributors. *Oral Hyg Health.* 2013;1(3):1-4.

6. Tellefsen G, Liljeborg A, Johannsen A, Johannsen G. The role of the toothbrush in the abrasion process. *Int. J. Dent. Hyg.* 2011;9(4):284-290.

7. Bomfim da Silva MA, Fardin AB, de Vasconcellos RCC, Santos LM, Tonholo J, da Silva JGS, et al. Analysis of roughness and surface hardness of a dental composite using atomic force microscopy and microhardness testing. Microsc. Microanal. 2011;17(3):446-451.

8. Sousa Barbosa RP, Pereira-Cenci T, Missio da Silva W, Coelho-de-Souza FH, Demarco FF, Cenci MS. Effect of cariogenic biofilm challenge on the surface hardness of direct restorative materials in situ. J. dent. 2012;40(1):359-363.

9. Moreira da Silva E, de Sá Rodrigues CUF, Dias DA, da Silva S, Amaral CM, Guimarães JGA. Clinical effect of toothbrushingmouthrinse-cycling on surface roughness and topography of nanofilled, microfilled, and microhybrid resin composites. *Oper. dent.* 2014;39(5):521-529.

10. Wei Y, Silikas N, Zhang Z, Wattsb DC. Hygroscopic dimensional changes of self-adhering and new resin-matrix composites during water sorption/desorption cycles. *Dent. mater.* 2011;27(1):259-266.

11. Sarkis E. Color change of some aesthetic dental materials. Effect of immersion solutions and finishing of their surfaces. *Saudi Dent J*. 2012;24(1):85-89.

12. Roselino LMR, Cruvinel DR, Chinelatti MA, Pires-de-Souza FCP. Effect of brushing and accelerated ageing on color stability and surface roughness of composites. *J. dent.* 2013;41(Suppl5):e54-61.

13. Correa MB, Peres MA, Peres KG, Horta BL, Barros AD, Demarco FF. Amalgam or composite resin? Factors influencing the choice of restorative material. *J. dent.* 2012;40:703-710.

14. Anusavice KJ. Degradability of dental ceramics. Adv Dent Res. 1992;6(1):82-89.

15. Azevedo SM, Kantorski KZ, Valandro LF, Bottino MA, Pavanelli CA. Effect of brushing with conventional versus whitening dentifrices on surface roughness and biofilm formation of dental ceramics. *Gen Dent.* 2012;60(3):e123-30.

16. Rashid H. The effect of surface roughness on ceramics used in dentistry: A review of literature. *Eur J Dent***.** 2014;8(4):571-9

17. Ferracane JL. Resin composite – State of the art. Dent. mater. 2011;27(1):29-38.

18. Ozak ST, Ozkan P. Nanotechnology and dentistry. *Eur J Dent.* 2013;7(1):145-151.

19. Carvalho FG, Sampaio CS, Fúcio SBP, Carlo HL, Correr-Sobrinho L, Puppin-Rontani RM. Effect of chemical and mechanical degradation on surface roughness of three glass ionomers and a nanofilled resin composite. *Oper. dent.* 2012;37(5):509-517.

20. Fúcio SBP, Paula AB, Carvalho FG, Feitosa VP, Ambrosano GMB, Puppin-Rontani RM. Biomechanical degradation of the nano-filled resin-modified glass-ionomer surface. *Am. j. dent.* 2012;25(6):315-320.

21. de Paula AB, Fúcio SBP, Alonso RCB, Ambrosano GMB, Puppin-Rontani RM. Influence of chemical degradation on the surface properties of nano restorative materials. *Oper. dent.* 2014;39(3):e109-e117

22. Smith R, Oliver C, Williams DF. The enzymatic degradation of polymers *in vitro*. *J. biomed. mater. res.* 1987;21(8):991-1003.

23. Heintze SD, Forjanica M, Ohmitib K, Roussonb V. Surface deterioration of dental materials after simulated toothbrushing in relation to brushing time and load. *Dent. Mater.* 2010;26(1):306-319.

24. Park JW, Song CW, Jung JH, Ahn SJ, Ferracane JL. The Effects of Surface Roughness of Composite Resin on Biofilm Formation of *Streptococcus mutans* in the Presence of Saliva. *Oper. dent.* 2012;37(5):532-539.

25. Sarkar NK. Internal corrosion in dental composite wear: its significance and simulation. *J. biomed. mater. res.* 2000;53(4):371-380

26. Hengtrakool C, Kukiattrakoon B, Kedjarune-Leggat U. Gradual surface degradation of restorative materials by acidic agents. *Gen Dent.* 2011;59(2):e50-e62

27. Cornelio RB, Wikant A, Mjosund H, Kopperud HM, Hassum J, Gedd UW, Örtengren UT. The influence of bis-EMA vs bis GMA on the degree of conversion and water susceptibility of experimental composite materials. *Acta Odontologica Scandinavica*. 2014;72(6):440-447.

28. Garcia LFR, Mundin FM, Pires de Souza FCP, Rontani RMP, Consani S. Effect of artificial accelerated aging on the optical properties and monomeric conversion of composites used after expiration date. *Gen Dent.* 2013;58(6):e262-267.

29. Moreira da Silva E, Gonçalves L, Guimarães JGA, Poskus LT, Fellows CE. The diffusion kinetics of a nanofilled and a midifilled resin composite immersed in distilled water, artificial saliva, and lactic acid. *Clin. oral invest.* 2011;15(1):393-401.

30. Ilie N, Hickel R. Resin composite restorative materials**.** *Aust. dent. j.*2011;56(Suppl1):59-66.

31. Melander J, Dunn WP, Link MP, Wang Y, Xu C, Walker MP. Comparison of flexural properties and surface roughness of nanohybrid and microhybrid dental composites. *Gen Dent.* 2011;59(5):342-347.

32. Moreira da Silva E, Dória J, Rodrigues da Silva JJ, Santos GV, Guimarães JGA, Poskus LT. Longitudinal evaluation of simulated toothbrushing on the roughness and optical stability of microfilled, microhybrid and nanofilled resin-based composites. *J. dent.* 2013;41(1):1081-1090.

33. Suzuki T, Kyoizumi Araki Y, Finger WJ, Kanehira M. Toothbrush abrasion of resin composites with different filler concepts. *World J Dent*. 2012;3(2):184-193.

34. Alrahla A, Silikas N, Watts DC. Post-cure depth of cure of bulk fill dental resin-composites*. Dent. mater.* 2014;30(1):149-154.

35. de Oliveira DCRS, Souza-Júnior EJ, Coppini EK, Paulillo LAMS. Color stability and polymerization behavior of direct esthetic restorations. *Journal esthet. restor. dent.* 2014;26(4):288-95.

36. Takahashi R, Jin J, Nikaido T, Tagami J, Hickel R, Kunzelmann KH. Surface characterization of current composites after toothbrush abrasion. *Dent. mater. j.* 2013;32(1):75-82.

37. Castro HL, Pereira PC, Feitosa AS, Valera MC, Araujo MAM, Araujo RM. Influence of brushing on a machined lithium disilicate-based ceramic: assessment of color maintenance and surface roughness. *Rev. Fac. Odontol.* 2014;19(1):83-87.

APPENDIX

Figure 1. Tested materials

1A 1B 1C

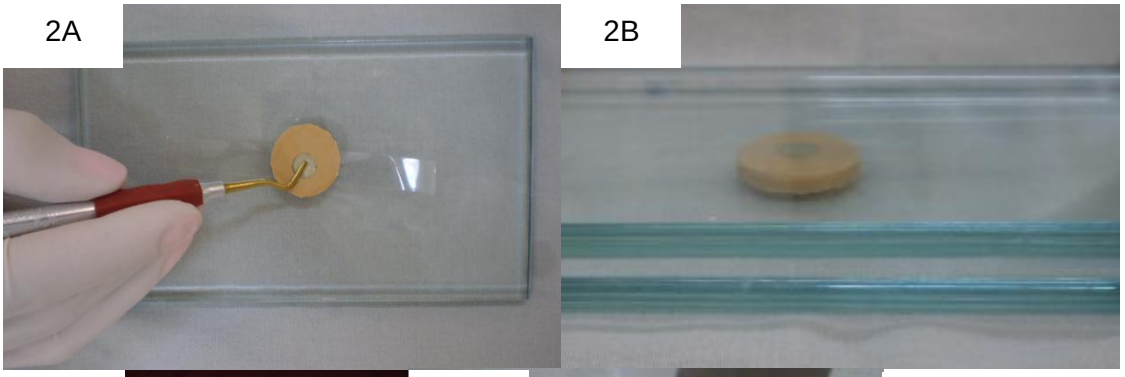
1A – Filtek Z350 (3M/ESPE)

1B – IPS Empress Direct (Ivoclar/Vivadent)

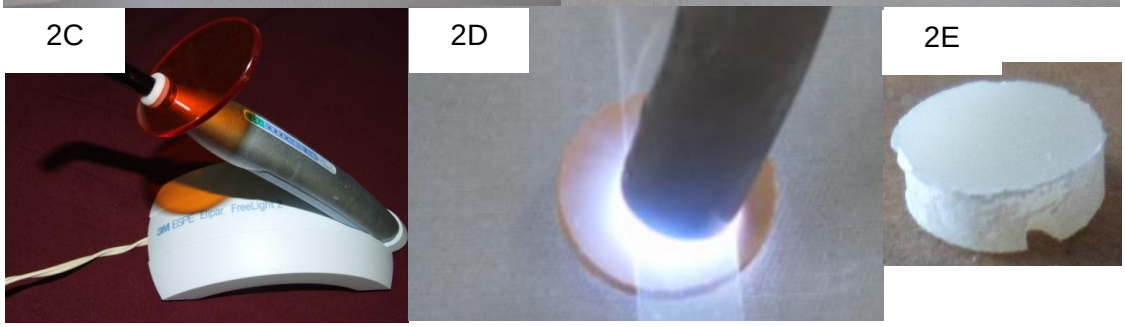
1C – IPS e.Max Ceram (Ivoclar/Vivadent)

Figure 2. Composites samples preparation

2A 2B



2C 2D 2E



2A – Composites samples preparation in silicone matrix

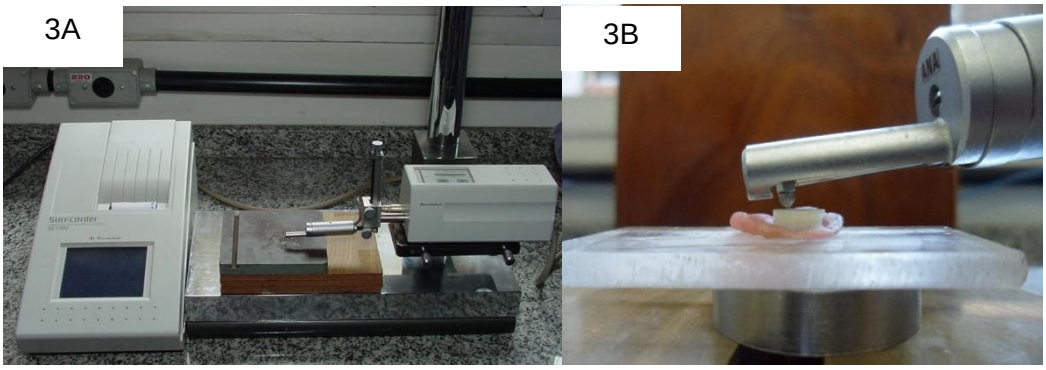
2B – Use of the polyester matrix and glass plate

2C – Curing light used

2D – Photopolymerization of specimens

2E – Perforation with drill to identify upper and lower surface

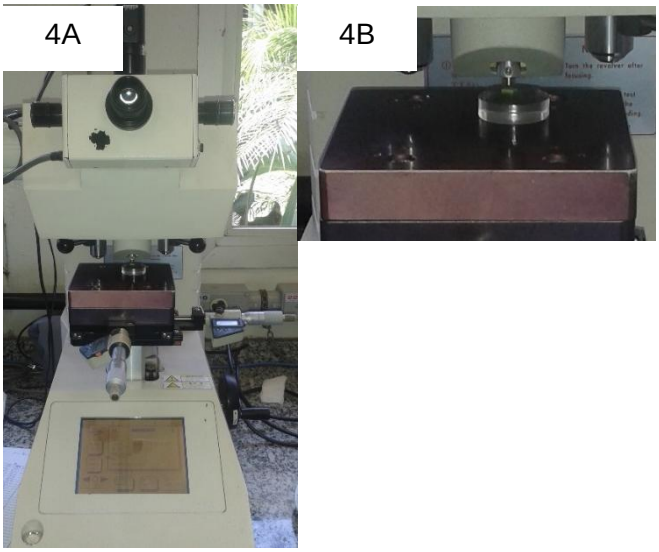
Figure 3. Rugosimeter – Surfcorder SE 1700, Kosaka, Tokyo, Japan



3A – Rugosimeter

3B – Roughness measurement

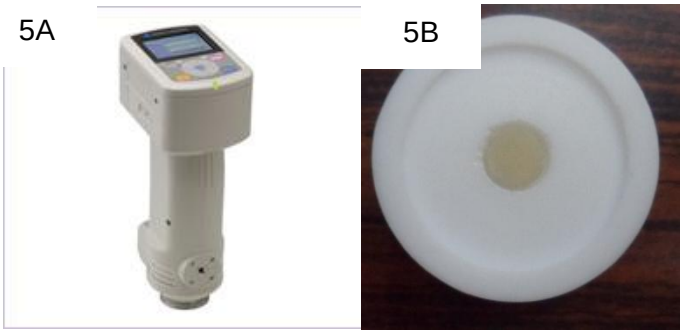
Figure 4. Durometer – HMV-2, Shimadzu, Tokyo, Japan



4A – Durometer

4B – Hardness measurement

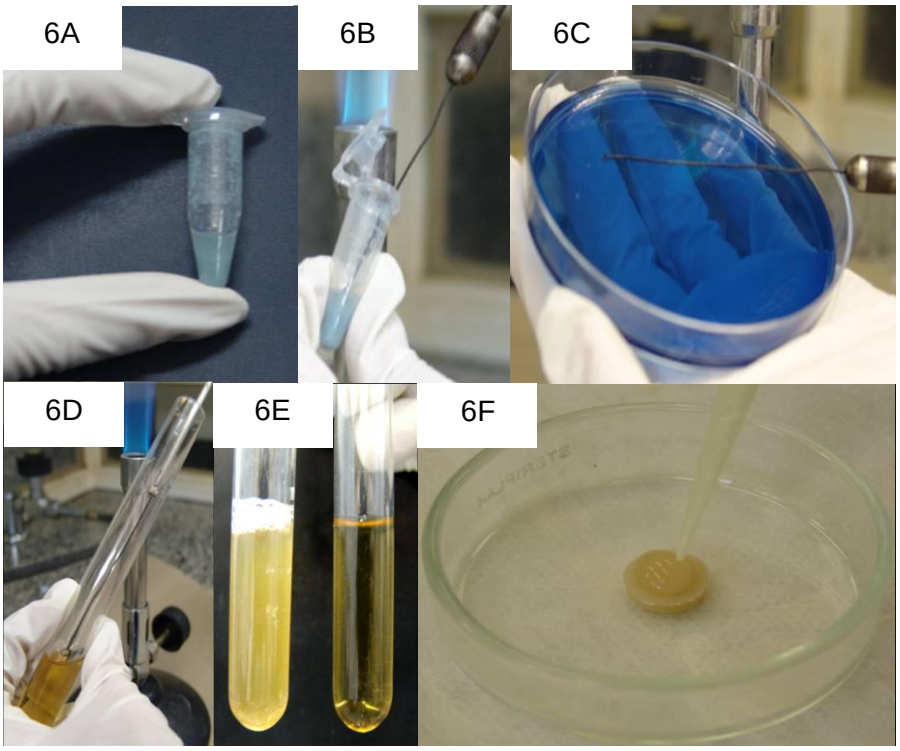
Figure 5. Spectrophotometer – CM-700d, Konica Minolta, Osaka, Japan



5A – Spectrophometer

5B – Sample carrier

Figure 6. Biofilm growth – Biological degradation



6A – Strain of *S. mutans* UA 159 frozen in Eppendorf

6B – Removal inoculum with handle

6C – Sowing inoculum in plate with miti-salivaris agar

6D – Inoculation of colonies on BHI broth

6E – Assay tubes with different turbidity after 24 hours

6F – 25 µL inoculum on the disk surface for initial adhesion of cells

Figure 7. Three-body abrasion test – Mechanical degradation

ANEXX

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