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# Biodegradation and Abrasive Wear of Nano Restorative Materials

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**Clinical Relevance**

Some nanofilled restorative materials may provide superior resistance to biomechanical degradation.

## SUMMARY

**The purpose of this study was to evaluate the biomechanical degradation of two nanofilled restorative materials (a resin-modified glass ionomer, Ketac N100 and a composite, Filtek Z350), compared with conventional materials (Vitremer and TPH Spectrum). Twenty specimens obtained from each material were divided into two storage groups (n=10): relative humidity (control) and *Streptococcus mutans* biofilm (biodegradation). After 7 days of storage, roughness values (Ra) and micrographs by scanning electron microscopy (SEM) were obtained. In a second experimental phase, the specimens previously subjected to biodegradation were fixed to the tooth-brushing device and abraded via toothbrushes, using dentifrice slurry (mechanical degradation). Next, these specimens were washed, dried, and reassessed by roughness and SEM. The data were submitted to repeated measures three-way analysis of variance (ANOVA) and Tukey tests ($p<0.05$). There was statistically significant interaction among factors: material, storage (humidity/biofilm), and abrasion (before/after). After biodegradation (*S mutans* biofilm storage), Ketac N100 presented the highest Ra values. Concerning bio plus mechanical challenge, TPH Spectrum, Ketac N100, and Vitremer presented the undesirable roughening of their surfaces, while the nano composite Filtek Z350 exhibited the best resistance to cumulative challenges proposed. The degraded aspect after biodegra-**

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DOI: 10.2341/10-221-L

**dation and the exposure of fillers after mechanical degradation were visualized in micrographs. This study demonstrated that the nanotechnology incorporated in restorative materials, as in composite resin and resin-modified glass ionomer, was important for the superior resistance to biomechanical degradation.**

## INTRODUCTION

Developed from concepts of molecular engineering, nanotechnology has enabled the structure of materials to be manipulated, to provide significant improvements in electrical, chemical, mechanical and optical properties, and to develop materials with new features.[1,2] In dentistry, nanoscale fillers or nanofillers correspond to primary particles about 40 nm or 0.04 μm in size. However, the material innovations are not exactly related to particle size, but to the possibility of increasing the nanofiller load in restorative materials,[2] leading to better mechanical behavior of materials.[3] Some studies have shown that nanocomposites presented higher surface hardness values and lower brushing abrasive wear than microfilled and hybrid composites.[2,4] In addition, resin-based nanocomposites offer high translucency, high polish, and polish retention, with a vast range of shade and opacity options.[5] Thus, the manufacturers indicate nanocomposites for both posterior and anterior restorations, since these materials show high mechanical properties and superior esthetics.

Recently, a new resin-modified glass ionomer cement (RMGIC) has been introduced for operative dentistry. Ketac N100 light polymerizing nanoionomer (3M ESPE) includes fluoroaluminosilicate glass, nanofillers, and nanofiller "clusters" combined to improve mechanical properties, such as three-body wear resistance (3M ESPE Internal Data).[6] In addition, this material contains 2-hydroxyethyl methacrylate (HEMA), bisphenol glycidyl methacrylate (Bis-GMA), and triethylene glycol dimethacrylate (TEGDMA) as resin monomers, different from the known RMGICs. It would be interesting to compare this material with a traditional resin-modified glass-ionomer, as Vitremer, and with a nano composite, as Filtek Z350, which has similar filler characteristics and greater diversity of resin monomers. Thus, it could establish if the nanoionomer shows a behavior similar to ionomeric and composite classes, predicting its mechanical and chemical properties.

Although it is possible to obtain improvement in the microstructure and surface morphology with the incorporation of nanofillers in restorative materials, one should consider that the restorative materials are constantly subject to thermal, mechanical, and chemical challenges in the oral cavity. The chemical challenges can be caused by acids produced by cariogenic biofilm,[7] acidic diet,[8,9] and salivary enzymes,[10,11] leading to softening and increased surface roughness of resin-based materials.[11–13] When brushed with dentifrice daily, these damaged surfaces gradually would lose softened material (matrix and filler), causing loss of contour, change of color, and roughening of the restoration surface again, influencing its esthetic and clinical longevity.[14] Nevertheless, little is actually known about the cumulative effects of an acidogenic biofilm and tooth-brushing abrasion on the surface characteristics of nanofilled restorative materials.

Therefore, the aim of this study was to evaluate *in vitro* the surface roughness and micromorphology of nano restorative materials, a nano-ionomer, and a nanocomposite, subjected to *Streptococcus mutans* biofilm degradation (biodegradation) and three-body abrasion (mechanical degradation), when compared with other resin-modified glass ionomer cements and composites.

## MATERIALS AND METHODS

### Specimen Preparation

Twenty specimens of each resin-modified glass ionomer cement and composite tested (described in Table 1) were fabricated using sterilized Teflon molds (5 mm in diameter; 2 mm deep) according to the manufacturer's instructions, under aseptic conditions. The materials were manipulated, placed in the mold by one operator, covered, and pressed flat with a sterilized glass slide. All specimens were polymerized with a curing light unit (Elipar Trilight, 3M ESPE, St Paul, MN, USA), according to the manufacturer's instructions, after the intensity of the light-curing unit was checked with a curing light meter (Hilux Dental Curing Light Meter, Benlioğlu Dental Inc., Demetron, Ankara, Turkey). After this, all disks were stored in 100% relative humidity at 37°C for 24 hours, and the polishing steps were not performed to avoid surface contamination. Initially, the specimens were distributed into two groups (n=10): the control group and biodegradation group. The control group was maintained in 100% relative humidity at 37°C for 7 days, while the other group was submitted to biodegradation for the same period of time.

Table 1: *Materials Used in This Study*

| Materials | Composition | Mean Filler Size[a] | Manufacturer (Batch) |
|---|---|---|---|
| Filtek Z350 | Zirconia/silica cluster filler, nonagglomerated silica filler, Bis-GMA, Bis-EMA, UDMA, TEGDMA | 0.6-1.4 μm (cluster)<br>5–20 nm (nanofiller) | 3M/ESPE, St Paul, MN, USA (8NU) |
| TPH Spectrum | Ba-Al-borosilicate glass, coloidal silica, Bis-GMA, Bis-EMA, TEGDMA | 0.8 μm | Dentsply Ind E Com Ltda, Petropolis, RJ, Brazil (L797977) |
| Ketac N100 | Paste A: silane treated glass, silane treated zirconia oxide silica, polyethylene glycol dimethacrylate, silane treated silica, HEMA, Bis-GMA, TEGDMA paste B: silane treated ceramic, silane treated silica, copolymer of acrylic and itaconic acids, HEMA | 1 μm (cluster)<br>5–25 nm (nanofiller)<br>< 3.0 μm (glass) | 3M/ESPE, St Paul, MN, USA (M3M3) |
| Vitremer | Powder: fluoroaluminosilicate glass, redox system; liquid: aqueous solution of a modified polyalkenoic acid, HEMA | 3.0 μm | 3M/ESPE, St Paul, MN, USA, (P:6LP/ L:6FH) |

[a] *Manufacturers' information.*
*Abbreviations: Bis-EMA, ethoxylated bisphenol-A dimethacrylate; Bis-GMA, bisphenol glycidyl methacrylate; HEMA, 2-hydroxyethyl methacrylate; TEGDMA, triethylene glycol dimethacrylate; UDMA, urethane dimethacrylate.*

## Biofilm Growth

*S mutans* strain UA159 was obtained from the culture of the Department of Microbiology and Immunology, Piracicaba Dental School, University of Campinas. To prepare the inoculum, *S mutans* was first grown on Mitis Salivarius agar (Difco Laboratories, Sparks MD, MI, USA) plates at 37°C for 48 hours in an environment supplemented with 10% $CO_2$. Subsequently, single colonies were inoculated into 5 mL of brain heart infusion (BHI) broth (Difco Laboratories) and incubated at 37°C for 18 hours. The biodegradation group specimens were exposed under static conditions to 25 μL of *S mutans* inoculum adjusted to an optical density of 0.6 at 550 nm (approximately $8 \times 10^{11}$ CFU/mL).

After 2 hours at room temperature, the nonadhering cells were removed by washing two times with 0.9% NaCl solution (saline). After this, a single material disk was placed in each well of polystyrene plates (Nunc™ multidish 96-well, Sigma, Saint Louis, MO, USA) with 2 mL of sterile, fresh BHI broth with addition of 1% sucrose (wt/vol). The bacterial accumulation occurred at 37°C in an environment supplemented with 10% $CO_2$, developing 7-day-old biofilms. The medium was renewed at 48-hour intervals. The purity of the cultures in the media was verified every day using Gram staining and plating samples. At the end of the experimental period, specimens were ultrasonically washed for 10 minutes and analyzed for surface roughness.

## Surface Roughness Measurements

Before the biodegraded disks were submitted to abrasion, both groups (control and biodegradation) were analyzed using a Surfcorder SE1700 surface roughness-measuring instrument (Kosaka Corp, Tokyo, Japan). Three readings from each specimen were taken. Additional specimens from the biodegradation group were taken to compare unbrushed surfaces with abraded surfaces by scanning electron microscopy (SEM) later.

## Three-body Abrasion Test

The tooth-brushing test was conducted at 250 cycles/min, for 30,000 cycles with a 200-g load on the biodegradation group specimens only. Colgate Total dentifrice (Colgate Palmolive Ind e Com Ltda, SB Campo, São Paulo, Brazil) diluted in distilled water (1:2) was used as an abrasive third body. Next, samples were washed in an ultrasonic bath for 10 minutes and gently dried. Three final surface roughness readings were taken from each specimen from this group (biomechanical degradation) in the opposite direction to that of the tooth-brushing movement.

Table 2: *Surface Roughness Values (μm) (Mean and Standard Deviation in Parentheses) of Restorative Materials Submitted to Relative Humidity (Control), Biodegradation, or Biomechanical Degradation (Biodegradation + Abrasion)†*

| Group | Materials | | | |
|---|---|---|---|---|
| | Filtek Z350 | TPH Spectrum | Ketac N100 | Vitremer |
| Control | 0.10 (0.02)$^{Ba}$ | 0.0.08 (0.02)$^{*Ba}$ | 0.19 (0.08)$^{*ABb}$ | 0.24 (0.20)$^{*Aa}$ |
| Biodegradation | 0.08 (0.02)$^{\#Ca}$ | 0.0.08 (0.05)$^{\#Ca}$ | 0.57 (0.12)$^{Aa}$ | 0.36 (0.08)$^{\#Ba}$ |
| Biomechanical degradation | 0.24 (0.05)$^{B}$ | 0.0.29 (0.03)$^{B}$ | 0.46 (0.09)$^{B}$ | 1.01 (0.46)$^{A}$ |

*† Uppercase letters indicate comparison among materials (horizontal) within each group. Lowercase letters demonstrate comparison between storage groups (control × biodegradation). Symbols represent the differences between biomechanical degradation and control groups (asterisks) / biodegradation group (hash). Groups denoted by the same letter/symbol represent no significant difference (p>0.05).*

### Surface Morphology Assessment

After the experimental period, three representative specimens of each group (control, biodegradation, and biomechanical degradation) were rinsed, dried, and mounted on a holder using double-sided adhesive carbon tape in order to illustrate the effect of tooth-brushing and biodegradation on the material surfaces. The samples were sputter-coated with gold under vacuum (Balzers-SCD 050 sputter coater, Balzers, Liechtenstein) and examined with a Model JEOL JSM 5600 LV scanning electron microscope (Tokyo, Japan) operating at 1000× magnification.

### Statistical Analysis

First, the data were evaluated to check the equality of variances and normal distribution. Then, the data were submitted to repeated measures three-way analysis of variance (ANOVA) and Tukey tests with a significance limit of 5%, since the specimens used for the abrasion test were the same ones used previously for the biodegradation procedure (biomechanical degradation).

## RESULTS

Surface roughness values of all materials tested are described in Table 2. There was significant difference among materials studied ($p<0.0001$), between storage conditions (humidity/biofilm; $p<0.0001$) and between tooth-brushing effects (before/after; $p<0.0001$). Moreover, a significant interaction was observed among the three factors: materials, storage, and abrasion (before/after) ($p<0.0001$).

When different storage conditions were compared for each material before abrasion, the *S mutans* biofilm provided degradation, ie, significantly higher roughness values for Ketac N100 specimens. The other materials tested presented similar values between the control (humidity) and biodegradation groups. However, the cumulative effect of biodegradation plus abrasion roughened the specimens of all materials, except the Filtek Z350 surface. Under these conditions (biomechanical degradation), the roughness values of TPH Spectrum and Vitremer almost tripled, while the nano-ionomer became about two times rougher. With regard to the effects of abrasion on the biodegradation group surface, only Ketac N100 showed similar roughness values before and after brushing. The other materials presented higher values after abrasion than before it.

When the materials were compared within the control group (relative humidity), both composites presented similar roughness values. Ketac N100 values showed no statistical difference from those of composites and Vitremer, while the latter material presented the roughest surface. When biodegraded materials were compared before abrasion, the composites retained the smoothest surfaces, followed by Vitremer, then Ketac N100. Moreover, after biomechanical degradation, Vitremer showed higher roughness values than the other materials.

The scanning electron micrographs in Figure 1 show details of the surface morphology of the studied materials, distributed in rows (different materials) and columns (different conditions). In the control group images, a smooth surface layer with undetectable fillers was observed for all materials (Figure 1a,d,g,j). Only Vitremer presented a large number of porosities on its surface, probably caused by the incorporation of air bubbles during conventional powder/liquid mixing (Figure 1j). After bacteria-surface interaction (biodegradation group), changes in the surface texture were very evident for resin-

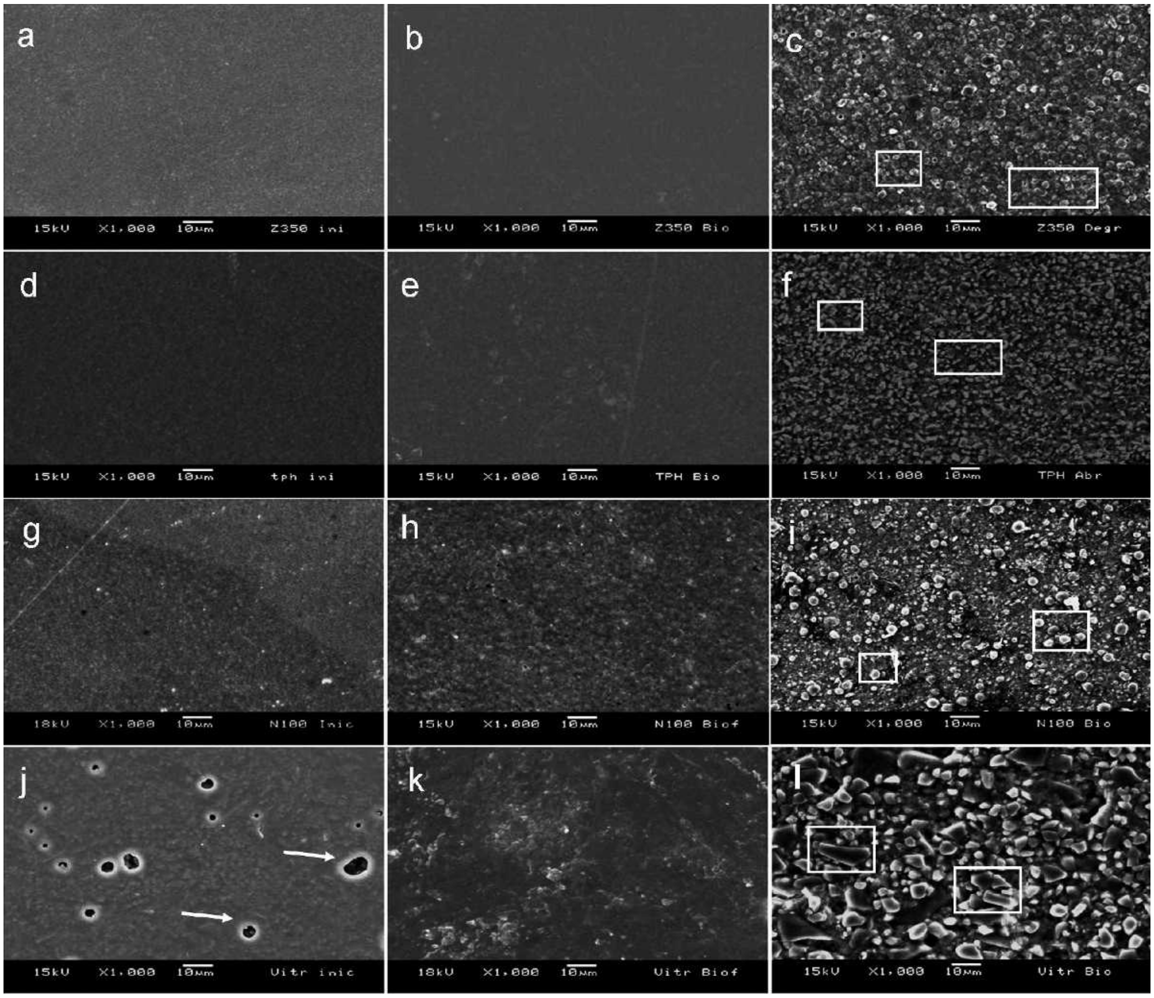

Figure 1. *Scanning electron micrographs of Filtek Z350 (a, b, c), TPH Spectrum (d, e, f), Ketac N100 (g, h, i), and Vitremer (j, k, l), at an original magnification of 1000×. The first column shows the relative humidity storage groups (a, d, g, j), with porosities (small spherical and irregular shapes) indicated by arrows. The second column represents the S mutans biofilm storage groups (b, e, h, k), with a severe degraded aspect of the matrix pointed out by marking it with circles. The third column corresponds to biofilm storage plus abrasion groups (c, f, i, l), with many exposed particles at the surface of materials (squares).*

modified ionomer samples, particularly for the nanoionomer, which presented a degraded aspect of the matrix (Figure 1h). After the abrasion test of biodegraded specimens (biomechanical degradation group), all materials showed a discernible loss of organic matrix, leading to irregular surfaces and protruding filler particles (Figure 1c,f,i,l). The difference in particle shapes and sizes among the studied materials (nano × conventionals) was clearly visible.

## DISCUSSION

Corrosive wear or biomechanical degradation results from the joint action of chemical and mechanical forces and is associated with the mechanical removal of degraded layers that form on the surface of a material by reaction with its environment.[15] Since corrosive wear is a continuous process occurring during the lifetime of the restoration, degradation characteristics of restorative materials are related to their long-term clinical performance. While resin-based materials undergo the cleavage of polymer chains to form oligomers and monomers, the ionomeric cements present a complex process of absorption, disintegration, and outward transportation of ions.[16] The present study evaluated the biomechanical degradation resistance of two composites and two resin-modified glass ionomers with important differences in their chemical composition, as discussed below.

Initially, the interaction of the studied materials with a *S mutans* biofilm promoted the biodegradation process. The organic acids produced by bacterial metabolism can change the environment pH,[17] which started at about 7.3 and fell to about 4.0 for composites and nano-ionomer and 4.5 for Vitremer during the experimental period (7 days). According to Sarkar[15], corrosive wear begins with water absorption that diffuses internally through the resin matrix, filler interfaces, pores, and other defects, accelerated by low pH. Thus, the biodegradation rates of different restorative materials depend greatly on their hydrolytic stability, which is related mainly to resin matrix composition and polymerization reactions in this study. It is important to remember that the present study did not use any method of surface finishing in order avoid contamination of the aseptic surface of the specimens, since all available sterilization methods may affect the structure and properties of the restorative materials.[18,19] Therefore, the outermost surface subjected to the biodegradation process was a resin-rich surface layer (Figure 1a,d,g,j), due to organic polymer migration to the surface of the material.[20] Furthermore, this superficial layer remains only partly polymerized due to the oxygen inhibition of polymerization, producing inferior surface properties of glass ionomer[21] and composite resins.[22]

In this study, resin-modified glass ionomer and composite resins behaved differently with regard to their surface roughness and morphology as a result of the 7-day biodegradation period. The ethoxylated version of the Bis-GMA (Bis-EMA - ethoxylated bisphenol-A dimethacrylate) existing in the composition of Filtek Z350 and TPH Spectrum matrices probably contributed to their hydrolytic and biochemical stability due to the hydrophobicity of this monomer.[12] Whereas the nanofilled resin-modified glass ionomer, Ketac N100, significantly presented the highest roughness value in comparison with the other materials subjected to biodegradation, it was the only material with higher roughness values when compared with the control group (humidity). A reasonable explanation for this severe biodegradation, also observed in the micrographs of Ketac N100 specimens (Figure 1h), is the other resin monomers in addition to 2-hydroxyethyl methacrylate (HEMA) present in this material, such as Bis-GMA and TEGDMA. Some studies have shown that in the presence of water, Bis-GMA/HEMA undergoes microphase separation, the hydrophilic tertiary amine and hydrophobic camphoroquinone tend to exist in the hydrophilic HEMA phase and hydrophobic Bis-GMA phase, respectively.[23,24] This decreases the chance of their coming into contact; therefore, fewer radicals will be generated and a lower degree of conversion could be found in the Ketac N100 matrix than in Vitremer matrix. Moreover, Vitremer shows a third polymerization setting reaction that ensures that any HEMA not polymerized by irradiation will set.[25] Clinically, the removal of the outermost surface by finishing-polishing procedures would tend to make the nanofilled RMGICs more resistant to biodegradation, and therefore, more esthetically stable restorative materials.[21,22,26]

With regard to relative humidity storage, Vitremer presented the highest surface roughness value in comparison with the other materials. Composites and Ketac N100 exhibit an external hydrophobic film,[20] arranged by different monomers, with different molecular structure and chemical characteristics (Bis-GMA, TEGDMA, Bis-EMA, UDMA, among others). The hydroxyl groups of HEMA give a hydrophilic property to the polymer matrix of Vitremer.[27] At 100% relative humidity, the water vapor can adsorb to the Vitremer surface through hydrogen bridges with the hydroxyl of HEMA[28] and promote a roughening of its superficial layer. Thus, the protection of Vitremer surface with varnishes, adhesives systems, or petroleum jelly is fundamental in order to avoid premature contact with water and the filling of small surface voids and defects, reducing the uptake of stains, the loss of calcium and aluminum ions, surface erosion, and loss of translucency.[29] Moreover, Ketac N100 is a paste/paste ionomer, while Vitremer requires the conventional powder/liquid mixing which promotes the incorporation of air bubbles (Figure 1j).

The wear resistance can be ascribed to several factors, such as the size, hardness, and percentage of surface area occupied by filler particles and the filler/matrix interaction,[13] as well as the degree of conversion of the polymer resin matrix.[29] The selective abrasion of the resin matrix and exposure of filler particles were observed for all materials studied (Figure 1c,f,i,l). It is known that there is a difference between filler and matrix hardness in resin-based materials,[30] mainly when this surface is a resin-rich layer, partially polymerized (oxygen inhibition) and softened by the biodegradation process. With regard to three-body wear (toothbrushing), it is necessary to establish two evaluations: the effect of wear alone (biodegradation × biomechanical degradation groups) and the cumulative effect of biofilm plus abrasion (control × biomechanical degradation groups).

When previously biodegraded, only Ketac N100 specimens presented no increase in roughness values after tooth-brushing abrasion; only Filtek Z350 specimens retained similar roughness values before and after biomechanical degradation. In the micrographs the removal by abrasion of the superficial exposed layer of all materials was observed as soon as it reached a critical degree of softening as a result of the biodegradation process. However, the surface roughness value of Ketac N100 was already high after this process due to its matrix composition, as related above. Therefore, there was probably no statistical difference between the abrasion values obtained from the biodegraded specimens of Ketac N100. Further clinical studies are necessary to confirm the effectiveness of this recent nano-ionomer as a restorative material able to withstand all adverse conditions of the oral environment, as well to inhibit the growth of bacteria and caries progression by means of fluoride release.

The biomechanical degradation resistance of nanocomposite Filtek Z350 is basically related to its chemical composition. With regard to filler particles, this material is formulated using a combination of nano-sized particles with the nanocluster formulations.[5] The higher filler loading with smaller particle size provides a reduction in the interstitial spacing, which effectively protects the softer matrix, reduces the incidence of filler exfoliation, and enhances the overall resistance of the material to abrasion.[31] When the nanocomposite undergoes toothbrush abrasion, only nano-sized particles are plucked away, leaving the surfaces with defects smaller than the wavelength of light.[5] The larger and irregular filler particles of Vitremer (Figure 1l) may be easier to "pluck out" as whole filler particles from the resin matrix, which could then act as an additional abrasive agent once detached from the surface and held against the specimen.[32] Thus, Vitremer was the roughest material after biomechanical degradation.

With regard to the resin system of Filtek Z350, the greater part of TEGDMA was replaced with a blend of UDMA (urethane dimethacrylate) and Bis-EMA.[33] The low strength of TEGDMA-rich resin mixtures could be caused by low crosslink density and cyclization, since TEGDMA is a small and flexible molecule.[34] In addition, the absence of a phenol ring in the monomer chain of UDMA leads to higher flexibility and toughness in comparison with Bis-GMA, making the UDMA resins more reactive, with higher conversion and crosslink density than the Bis-GMA polymers. Thus, Filtek Z350 showed the best biomechanical degradation resistance, since TPH Spectrum does not contain the nanotechnology and UDMA in its matrix composition. Vitremer presented the highest roughness values after the cumulative changes, probably due to larger and irregular filler visualized in the micrographs of its specimens (Figure 1l).

## CONCLUSION

The nanofilled composite Filtek Z350 exhibited the best resistance to cumulative challenges (biofilm plus tooth-brushing abrasion), since both tests promoted the exposure of its regular and small particles. Although Ketac N100 contains the nanotechnology, its outermost matrix was fragile under the biodegradation process, suggesting the requirement of finishing-polishing procedures after restoration treatment. Differently, Vitremer presented a satisfactory resistance to biodegradation alone, but the exposure of its particles after abrasion promoted the highest roughness values in this study. It would be interesting to select carefully the restorative material for intra-oral sites where there are frequent accumulation of dental biofilm and brushing abrasion, giving special attention to the shape and size of fillers, since these will certainly be exposed and will determine the surface characteristics of tooth-colored restoratives.

### Acknowledgements

Financial support received from CAPES (Coordenação de Aperfeiçoamento Profissional de Nível Superior) is gratefully acknowledged. The authors wish to thank 3M ESPE and Dentsply for providing the materials and Department of Diagnostic Oral, Microbiology and Immunology Area, Piracicaba Dental School, University of Campinas, Piracicaba, SP.

*(Accepted 11 September 2010)*

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