ABSTRACT
This in vitro study evaluated the demineralization around restorations class V made on the buccal and lingual surfaces of teeth when using different restorative materials. Thirty extracted teeth were randomly divided into 3 groups (n=10) according to the restorative material: Group I - Fuji II LC (GC America Inc., Alsip, Illinois, USA), Group II - Tetric (Ivoclar Vivadent AG, Schaan, Liechtenstein) and Group III - Chelon Fil (3M/ESPE., Seefeld, Germany). The teeth were submitted to a pH-cycling model associated to a thermocycling model. Sections were made and the specimens were analyzed under a polarized light microscopy as for the presence of demineralization. Measurements were performed and the results were subjected to statistical analysis using Anova and Tukey’s Test (α=0.05). Mean values of demineralization depth (µm) according to each positions showed that the demineralization was significantly reduced when Chelon Fil (Group III) was used for all depths, when compared to fluoridated resin materials. Also, it was verified that non-fluoridated resin material, composite resin Tetric, had the lowest inhibitory effect on the development of demineralization.

UNITERMS: Fluorides; Demineralization; Microscope.

INTRODUCTION
The development of marginal alterations and secondary caries around the margins of composite resin restorations has been documented and may be considered one of the major causes of restorative failures and replacements. To prevent secondary caries around restorations, key factors are marginal integrity of the restoration, durable adhesion, physical properties of restorative materials, and oral hygiene. However, replacement of the restorations due to secondary caries is still a continuing problem in restorative dentistry.

Fractures, insufficient marginal adaptation and excess of restorative material, failures that are frequently observed in restorations, result in microleakage. The opening permits penetration of oral fluids and microbiological agents along the interface between the dental tissue and the restoration, which may lead to secondary caries development.

Based on the current concept that the cariostatic effect of fluoride is enhanced in lower, yet permanent, concentrations in the oral environment, incorporating fluoride into restorative materials is of special interest. These fluoridated materials are potential sources of release of this element, therefore, expanding the spectrum of prevention in restorative dentistry. The use of fluoride to demineralize and remineralize early enamel carious lesions directly interferes with caries evolution. It is accepted that part of the fluoride present in restorative materials may be directly released onto the areas of risk, such as restoration margins, where secondary caries may develop. Thus, the use of fluoride may be considered an additional method of preventing caries.

Glass ionomer cement, initially described in the early 1970s, is regarded as the material of choice in cases, where cavity sealing and prevention of secondary caries are desirable, due to its properties of adhesion to dental structure and its high rate of fluoride release.

However, the predominance of resin composites among esthetic restorative materials is evident, especially due to its highly satisfactory esthetics and easy manipulation. The greatest difficulty with this material is the occurrence of secondary carious lesions adjacent to the restoration, which is observed less frequently in teeth restored with glass ionomer.

In an attempt to obtain an ideal material that would combine the good properties of both resin...
composites and glass ionomer cements, new materials have been developed, such as resin-modified glass ionomers and polyacid-modified resin composites.

Therefore, considering that employing fluoride-releasing restorative materials is important for inhibiting the occurrence of secondary carious lesions, especially in patients at high risk and those with high caries activity, knowledge of the behavior of such restorative materials, which contain fluoride, and the conventional resin composite, a comparative evaluation of the cariostatic action of these materials is required. Thus, the aim of this study was to evaluate the marginal demineralization around restorations in vitro using polarized light microscopy. The null hypothesis tested was that restorative material used did not have inhibitory effect on the development of demineralization in depths studied.

MATERIAL AND METHOD

Specimen Preparation and Restorative Procedures

Thirty extracted human third permanent molars were cleaned and stored in 2% formaldehyde solution pH 7.0 at room temperature.

Class V preparations were made in the middle third of both buccal and lingual surfaces of each tooth using a water-cooled high-speed handpiece and a # 16-F diamond bur (KG Sorensen Indústria e Comércio Ltda., Barueri, São Paulo, Brazil). The approximate dimensions of the sixty formed cavities were: 1.5 mm in depth and 1.2 mm in diameter. The bur was discharged after every five cavities. No bevel was made on the cavosurface angle. Restorative materials used in the study are shown in Table 1.

Previously to restoration placement, the teeth were individually immersed in deionized water and randomly divided into three study groups according to the restorative material used (Table 2).

<table>
<thead>
<tr>
<th>Groups</th>
<th>Specimen</th>
<th>Pre-treatment Procedure</th>
<th>Material</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Extracted</td>
<td>Flat Cut: Fuji II LC</td>
<td>Fuji II LC, GC American Inc., Alsip, Illinois, USA</td>
</tr>
<tr>
<td>II</td>
<td>Prepared</td>
<td>Flat Cut: CHelon Fil</td>
<td>CHelon Fil, 3M/ESPE, Seefeld, Germany</td>
</tr>
<tr>
<td>III</td>
<td>Prepared</td>
<td>Flat Cut: Tetric</td>
<td>Tetric, Ivoclar Vivadent AG, Schaan, Liechtenstein</td>
</tr>
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</table>

The restorative technique performed for all materials followed the manufacturers’ instructions. Teeth from Group I and II were restored using resin modified glass ionomer Fuji II LC (GC American Inc., Alsip, Illinois, USA) and glass ionomer CHelon Fil (3M/ESPE, Seefeld, Germany), respectively. Teeth restored with composite resin Tetric (Ivoclar Vivadent AG, Schaan, Liechtenstein) (Group II), were etched using 37% phosphoric acid (3M/ESPE Dental Products, St. Paul, USA) and received two applications of the adhesive system Prime & Bond (Dentsply Caulk, Milford, USA) before composite resin application using the incremental technique. No adhesive system was used in specimens from groups I and III.

Afterwards, specimens were polished using Sof-Lex Pop On (3M/ESPE Dental Products St. Paul, USA) aluminum oxide disks. All restored teeth were stored in a humid environment for 48 hours.

PH-cycling and Thermocycling

Round segments (4 mm in diameter) of adhesive tape (3M/ESPE Dental Products, St. Paul, MN, USA) were placed on the restorations. Then, all tooth surfaces were coated with acid-resistant varnish. A nylon wire was fixed at each tooth to facilitate its handling. The tape was removed from the tooth as soon as the varnish dried, leading to the exposure of the restoration as well as of 1 mm of dental tissue around the restorative material.

So, each group of teeth was immersed in 100 mL of synthetic acid demineralizing solution (2.0 mmol L⁻¹ Ca, 2.0 mmol L⁻¹ P, in 75 mmol L⁻¹ acetate buffer, pH 4.3) for 4 hours. The teeth were then immersed in 20 mL of remineralizing solution (1.5 mmol L⁻¹ Ca, 0.9 mmol L⁻¹ P, 0.1 buffer, pH 7.0) for 20 hours. All teeth were rinsed thoroughly with deionized water and dried with an absorbent paper before and after a demineralizing period. The continuous cycles of demineralization and remineralization were carried out for 28 days. During the in vitro demineralization/ remineralization cycling model, the teeth of each group were subjected to thermocycling for 100 cycles. A complete cycle consisted of 90 seconds at 37°C, 90 seconds at 55°C and 90 seconds at 5°C.

Optical Microscopy Analysis

After 28 days, the teeth were individually fixed in acrylic resin blocks and sectioned to a thickness of about 500 µm using a diamond sectioning disk in a metallographic cutter (Isomet 2000 – Buehler UK LTD, Lake Bluff, USA). The sections were then ground to a thickness of about 100 µm. After 48 hours of imbibitions in deionised water, the sections were examined and photographed using a polarized light microscopy (Axiophot – ZEISS DSM-940 A, Oberkochen, Germany).

Readings were performed at the R1 and R2 regions, around the occlusal and cervical margins of the restoration, along the interface between the dental tissue and the restoration (P1) and at 100 µm (P2), 200 µm (P3) and 300 µm (P4) from the interface.
between the dental tissue and the restoration. The
demineralization was measured according to its depth
(Figure 1).

Figure 1 - Illustration of positions P1, P2, P3, P4 regarding
caries development in the regions R1 and R2.

Data Analysis
The results obtained were submitted to one-
way analysis of variance (ANOVA) and Tukey test, at
a level of significance of α=0.05, to verify the inhibitory
effect on the development of demineralization around
class V restorations when using different restorative
materials.

RESULTS
After achievements of the original values, the
means depths of demineralization were calculated
(Figure 1) according to their respective groups and
positions regarding demineralization development
(Table 3).

Tukey test at the 5% level was used, as
presented in Tables 3 revealing that the interaction
between group III in all positions analyzed clearly
demonstrated a higher resistance to the development of
demineralization, since the interactions observed
presented the lowest mean depths of demineralization
on the enamel surface, for all positions investigated.
Groups I and III, which are fluoride release materials,
showed statistically similar results in P1 position.
However, this was not observed for positions 2,
3 and 4, which exhibited statistically different mean
depths of demineralization in these groups. Group II
had the lowest inhibitory effect on the development of
demineralization in all depths analyzed, while group
III presented the best outcomes in relation to
demineralization inhibition (Figure 2).

DISCUSSION
The control of carious lesions is mainly related
to the presence of fluoride in the oral cavity and may
not be considered as directly dependent on the amount
incorporated by the tooth, since the main
mechanism of action of fluoride is dynamic, inhibiting
the demineralization and enhancing remineralization.
Therefore, the constant presence of fluoride in the
oral cavity, in saliva or oral fluids, dental plaque
and enamel, may control or even inhibit the appearance
of secondary carious lesions, as well as lead to arrest
of existing lesions, preventing progression of incipient
lesions to formation of cavity.

It should be highlighted that some authors
adopted similar in vitro model of caries development,
presenting correlation with the onset and
progression of carious lesions in vivo, in situations
of high risk to caries. However, as to the cariogenic
challenge in the present study, besides utilization of
the pH cycling, thermal cycling was also conducted
to approach the real conditions of the oral cavity.

Thus, investigation by polarized light
microscopy revealed the values of depth of
demineralization in the dental enamel, considering the
factors group and position, whose means are
presented in Tables 3. Considering the values
presented in Table 1, representing the interaction
between P1 position (interface) with the other groups,
it was demonstrated that the material employed in
group III (Chelon Fil) presented a better performance
for control of demineralization, followed by the material
in group I (Fuji II LC) and finally the material used in
group II (Tetric) which exhibited the worst performance.

Similarly, the values found for the interactions
of groups with positions P2, P3 and P4, respectively,
demonstrated the same performance of the material
employed in group III (Chelon Fil) for control of
demineralization, as presented in Table 3. However,
the other groups revealed a tendency toward similar outcomes as to their cariostatic potential, proportional to their distance from the tooth/restoration interface. These findings further reinforce that the amount of fluoride present in the material, as well as its concentration and release, are important aspects to reduce the demineralization.

This leads to the assumption that the cariostatic property of Chelon Fil (group III) may be explained by the intensive fluoride release of this material, and the amount of fluoride released depends on its concentration in the material; in addition, ionic fluoride is present in this material, which favors its release.

These statements are corroborated by the mean values observed in this study, which demonstrated that the material Chelon Fil with high fluoride release and bonding to the tooth structure, presented a better performance for control of demineralization; these results observed (Figure 2) are in agreement with other studies, who also conducted investigations by polarized light microscopy and revealed the formation of mild demineralization, and that the establishment and progression of demineralization in these cases are reduced, possibly due to the precipitation of calcium and phosphate triggered by the high fluoride release of these materials. This may be explained by the intensive fluoride release of this material, which depends on its concentration in the material and especially on the presence of ionic fluoride, which enhances its release.

Considering the results achieved for group I (Fuji II LC), its performance was inferior to group III (Chelon Fil), thereby demonstrating that resin-modified glass ionomers also present an anticariogenic action, yet inferior when compared to the conventional glass ionomer. Therefore, the results obtained in this study for group II are in agreement with some studies, which achieved similar results, observing a significant reduction in the demineralization, assigned to the significant fluoride release of this material, because of the spontaneous acid-base reaction, which leaves free fluoride ions in the bulk to be released.

The performance presented by the material Tetric in group II demonstrated that composites are not effective in caries inhibition. Even though the fluoridated resins currently available present fluoride release, they do not maintain this pattern to favor a considerable incorporation of fluoride by the tooth or even reduce the mineral loss close to restorations; thus the present outcomes are in agreement with other authors.

Another factor, besides fluoride release, should be considered. The thermal cycling can produce interface degradation to the materials that use adhesive technique. This process can increase microleakage for some factors. The principal factor is the different on thermal expansion coefficient between restorative material and teeth. This difference can overload interface during thermal cycling and to take gaps

Some authors stated that enamel remineralization requires the presence of partially demineralized hydroxyapatite crystals and is dependent on the degree of saturation of the area, in agreement with the present findings. This also corroborates the results found for material Chelon Fil (group III) which may be related to its higher fluoride release compared to the other materials, presenting a more effective action in the process of inhibition and progression of demineralization. It should be highlighted that material presenting low fluoride release, as Fuji II LC, were not effective at distant sites, yet they were effective on the tooth/restoration interface, confirming that the efficiency for control of distant lesions requires topical fluoride application and utilization of fluoridated mouth rinses and dentifrices, which allow the constant presence of low concentrations of fluoride in the oral cavity, being much more effective than the fluoride release by materials.

These results demonstrated that control of demineralization depends on the ability of materials to release fluoride ions; however, the clinical individuality of each patient should be considered for indication and application of a material or restorative technique. Thus, the null hypothesis was rejected, therefore, further investigations are warranted to indicate the most adequate clinical approach; also, professionals should be aware and consider Dentistry as a science.

**CONCLUSION**

Based on the present study results, the resin modified glass ionomer Fuji II LC and glass ionomer cement Chelon Fil did show inhibitory effect on the development of demineralization. Moreover, the demineralization was significantly reduced when Chelon Fil (Group III) was used for all depths.

**RESUMO**

Este estudo in vitro avaliou a desmineralização ao redor de restaurações classe V realizadas nas superfícies vestibular e lingual dos dentes com diferentes materiais restauradores. Trinta dentes extraídos hígidos foram aleatoriamente divididos em 3 grupos de estudo de acordo com o material utilizado: Grupo I - Fuji II LC (GC America Inc., Alsip, Illinois, USA); Grupo II - Tetric (Ivoclar Vivadent AG, Schaan, Liechtenstein) e Grupo III - Chelon Fil (3M/ESPE., Seefeld, Germany). Os dentes foram submetidos a diferentes variações de pH associadas à termociclagem. Os espécimes foram então preparados e secionados para análise em microscopia óptica sob luz polarizada quanto à presença ou não de desmineralização. As
mensurações foram realizadas e os resultados foram submetidos à análise estatística utilizando o Anova e Teste de Tukey (á=0,05). Os valores médios da profundidade de desmineralização (µm) de acordo com cada posição analisada mostrou que o material ChelonFil (Grupo III) apresentou redução significativa de desmineralização em todas as profundidades, quando comparado com os materiais resinosos com fluoretos. Além disso, observamos que para o material resinoso sem fluoreto, como a resina Tetric, foram verificados os menores efeitos inibitórios no desenvolvimento de desmineralização.

UNITERMOS: Fluoretos; Desmineralização; Microscópio.

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