Demineralization Inhibition of Enamel at Restoration Margins of Vitremer, Z 100 and Activa BioActive Restorative Materials: An in Vitro Study

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Abstract

Objective: The purpose of the study was to examine the effectiveness of a new restorative material which releases fluoride and calcium, compared to resin-modified glass-ionomer cement and a non-fluoride releasing resin in preventing secondary caries development in simulated cariogenic condition.

Materials & Methods: Class V preparation were placed in molar teeth, the occlusal margin of enamel. Thirty permanent molars were randomly divided to 10 each in Vitremer as positive control group, Z100 as negative control group and Activa BioActive as an experimental group. All teeth had an acid-resistant varnish placed to within 1 mm of restoration margins and placed in artificial caries challenge solution (pH 4.4) for 4 days. Bucco-lingual sections of 100µm were obtained, photographed under polarized light microscopy and then demineralized areas adjacent to the restorations were measured and quantified.

Results: The mean (±S.D) of enamel demineralization (µm²) adjacent to the dental restorative materials were Vitremer: 439.10 (567.55), Z100: 1732.80 (143.58) and Activa BioActive: 1209.50 (72.91) respectively. Statistically significant difference was observed between the above groups (P<0.001). Tukey’s test demonstrated less enamel demineralization adjacent to Vitremer resin-modified glass ionomer cement compared to Activa BioActive and Z100 resin-based composite (P<0.05). Activa BioActive had significantly less adjacent enamel demineralization than Z100 resin-based composite (P<0.05).

Conclusion: Vitremer performed superiorly at inhibiting enamel demineralization at restoration margins, followed by Activa BioActive, and Z100.

Keywords: Calcium; Acid-resistant; Enamel; Tooth structure
Introduction

Recurrent caries or secondary caries, located at the interface between restorations and tooth structure are considered a critical contributing factor for the replacement of amalgam and resin-based composite restorations [1,2]. There is evidence that the incidence and severity of secondary caries is decreased around restorations that leach out fluoride. Several studies have determined the anticariogenic properties of restorations comprising glass ionomer cement [3-7].

A major concern with resins used in restorative dentistry is secondary caries inhibition, as they do not release fluoride, in addition to the risk of polymerization shrinkage which can lead to compromising restoration margins. Therefore, manufacturers have developed modern properties of dental restorative materials involving the use of bioactive materials and resin-based materials with incorporated glass filler, which have demonstrated an antimicrobial effect on oral bacteria that aids in decreasing adjacent tooth demineralization. This happens due to the ability of bioactive glass to release fluoride, calcium, and phosphate, which neutralize the local acidic environment in the oral cavity [8]. A definite benefit of these materials over glass ionomer is the increased compressive strength and subsequent wear properties. However, having this buffering effect and release of calcium and fluoride, the capacity of these materials in inhibiting demineralization at restoration margins is unknown. Thus, evaluation of demineralization process is required in order to establish the effect of new resin-based restorative materials in the prevention of secondary caries formation.

The purpose of this in vitro study was to examine the effectiveness of a new restorative material, which the company markets as a resin-modified glass ionomer cement, to inhibit adjacent enamel demineralization. The material contains and releases fluoride and calcium (Activa BioActive Restorative Pulpdent Corporation, Watertown, MA, USA) and was compared to a positive control of resin-modified glass-ionomer cement (Vitreme 3M, St. Paul, MN, USA), as well as a non-fluoride releasing resin (Z100 3M, St. Paul, MN, USA), which was used as a negative control in preventing secondary caries development in simulated cariogenic conditions. The hypothesis tested was that caries inhibition ability in enamel of restorations carried out with the different materials is affected differently by an artificial cariogenic solution.

Materials and Methods

Thirty extracted caries-free human permanent molars were collected from the University of Texas Health Science Center at San Antonio (UTHSCSA) dental clinics and were used for this in vitro study. Currently, UTHSCSA has a standing Institutional Review Board (IRB) acceptance of using human teeth for studies without individual IRB review for each study because extracted teeth were accumulated with no identifying features. Teeth were stored in 0.1% thymol solution until experimentation initiation to prevent dehydration and fungal growth. Teeth were cleaned using a rotating brush and prophylactic paste to remove any remnant biofilm. Class V preparation were made with a #330 carbide bur in a high speed hand piece on the buccal surfaces of each tooth. The preparation extended 2 mm axially, 5 mm mesiodistally and 2 mm occlusogingivally, the enamel margin having a 0.5 mm at 45-degree bevel.

After all the 30 preparations were completed, they were randomly distributed to one of the three restorative material groups. Firstly, ten teeth had a non-fluoridated resin restoration (Z100 3M, St. Paul, MN, USA) placed as a negative control. The preparations were etched with 37% phosphoric acid on the beveled enamel margins, followed by the placement of primer and adhesive, as recommended by the manufacturer. Z100 resin based composite was incrementally placed and light cured. Secondly, ten teeth had an experimental fluoride and calcium releasing bioactive restorative material (Activa BioActive Restorative, Pulpdent Corporation, Watertown, MA, USA) and thirdly, ten teeth had a resin-modified glass ionomer cement (Vitremer 3M, St. Paul, MN, USA) placed as a positive control. Materials were placed according to the manufacturer’s recommendations. All restorations were polished with Sof-Lex™ discs (3M ESPE, St. Paul, MN, USA).

All 30 teeth were coated with acid-protective varnish, leaving a minimum of 1 mm exposed adjacent to restoration margins. The teeth were then placed into an artificial caries challenge (pH 4.4) for four days [9]. At the end of the four days, teeth were sectioned buccolingually through the restored portion of the tooth using a Silverstone/Taylor hard tissue microtome (Scientific Fabrications, Colorado Springs, CO, USA). 100 µm sections were obtained from each tooth, then images were obtained using polarized light microscopy and imbibition media of water. After the images were saved, the demineralized areas adjacent to the enamel restoration margin were quantified using Image-Pro Insight software (Media Cybernetics, Rockville, MD, USA).

Statistical Analysis

Kruskal-Wallis One Way Analysis of Variance test was conducted for overall comparison between the groups. Subsequently, Tukey’s test was conducted for individual comparison between the materials. Statistical significance was set at $P<0.05$ for all the tests and was conducted using SigmaStat 3.5 software (Systat Software, Inc., San Jose California USA).

Results

The mean ($±$S.D) of enamel demineralization ($\mu$m$^2$) adjacent to the dental restorative materials were: Vitremer: 439.10 ($567.55$), Z 100: 1732.80 ($143.58$) and Activa BioActive: 1209.50 ($72.91$) (Table 1). The minimum measurement of the carious lesion was observed in the Vitremer group ($1062 \mu m^2$) and maximum was observed in the Z100 group ($1925 \mu m^2$) (Table 2). For overall comparison between the groups, Kruskal-Wallis One Way Analysis of Variance test was conducted, and it indicated a statistically significant difference between groups ($p<0.001$). For individual comparison between the materials, Tukey’s test was conducted, and it demonstrated significantly less enamel demineralization adjacent to Vitremer resin-modified glass ionomer cement compared to Activa BioActive and Z 100 resin-based composite ($P<0.05$). Similarly, Activa BioActive had significantly less adjacent enamel demineralization than Z 100 resin-based composite ($P<0.05$) (Figures 1-3).
Table 1: Descriptive statistics of Vitremer, Z100 and Activa BioActive materials used in this study.

<table>
<thead>
<tr>
<th>Material</th>
<th>Size</th>
<th>Missing</th>
<th>Mean</th>
<th>Std. Dev</th>
<th>Std. Error</th>
<th>C.I. of Mean</th>
</tr>
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<tbody>
<tr>
<td>Enamel Vitremer</td>
<td>10</td>
<td>0</td>
<td>439.100</td>
<td>567.551</td>
<td>179.475</td>
<td>406.001</td>
</tr>
<tr>
<td>Enamel Z100</td>
<td>10</td>
<td>0</td>
<td>1732.800</td>
<td>143.585</td>
<td>45.406</td>
<td>102.714</td>
</tr>
<tr>
<td>Enamel Activa BioActive</td>
<td>10</td>
<td>0</td>
<td>1209.500</td>
<td>72.910</td>
<td>23.056</td>
<td>52.157</td>
</tr>
</tbody>
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Table 2: Measurement of carious lesion area in squared micrometers (µm²).

<table>
<thead>
<tr>
<th>Enamel Vitremer</th>
<th>Enamel Z100</th>
<th>Enamel Activa BioActive</th>
</tr>
</thead>
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<tr>
<td>1063.0000</td>
<td>1658.0000</td>
<td>1130.0000</td>
</tr>
<tr>
<td>0.0000</td>
<td>1843.0000</td>
<td>1278.0000</td>
</tr>
<tr>
<td>1062.0000</td>
<td>1455.0000</td>
<td>1276.0000</td>
</tr>
<tr>
<td>0.0000</td>
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<td>1196.0000</td>
</tr>
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<td>0.0000</td>
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<td>1066.0000</td>
</tr>
<tr>
<td>1102.0000</td>
<td>1925.0000</td>
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</tr>
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</tr>
<tr>
<td>0.0000</td>
<td>1737.0000</td>
<td>1188.0000</td>
</tr>
</tbody>
</table>

Figure 1: The enamel (E) margin adjacent to a Vitremer (positive control) resin-modified glass ionomer cement restoration. Note the inhibition zone (IZ) where the lesion (L) is shallow at the restoration margin. RTI refers to restorations-tooth- interface.

Figure 2: The enamel (E) margin adjacent to Activa BioActive (experimental material). Note the lesion is deeper than the Vitremer and there is no inhibition zone associated with the lesion(L). RTI refers to restorations-tooth- interface.

Figure 3: The enamel (E) margin adjacent to a Z100 (negative control) resin based composite restoration. Note that there is a wall lesion (WL) forming and the caries lesion (L) is deeper than the lesion adjacent to the Activa Bioactive restoration. RTI refers to restorations-tooth- interface.

Discussion

The increase in patient demands for esthetic dental restorative materials makes these materials an essential part of a dentist’s tool kit. However, it is well known, as shown in the previous studies that caries is an active process faced by daily challenges of periods of demineralization followed by periods of remineralization in the oral cavity [10,11]. Therefore, it is critical to use a dental material that has the ability to reduce caries incidence and inhibit demineralization adjacent to the fluoride releasing restorations.

The search has been ongoing over many years for an ideal dental restoration and no material have ever been able to achieve the above said characteristics. Recently, a relatively new hybrid esthetic dental material called Activa BioActive restorative has been launched to the marketplace. It is claimed that this material combined both the clinical benefits of fluoride release, present in Glass Ionomer Cements (GICs), and the superior physical properties of resin-based composites to make it more bioactive than glass ionomer cements [12,13]. But it is unknown if the fluoride content released is sufficient enough to inhibit demineralization at restoration margins comparable to glass ionomer cements.

This in vitro study examined the demineralization inhibition potential at restoration margins of three different restorative materials. The results of this study indicated Vitremer resin-modified glass ionomer cement performed the best at inhibiting caries at enamel restoration margin. Similarly, previous research has shown that all of the fluoride-releasing materials demonstrated a statistically significant degree of protection of enamel from demineralization compared to the non-fluoride control materials [14]. This inhibition may be associated with resin-modified glass ionomer cement which has a continuous fluoride release [15]. In contrast to the findings of this study, Savarino et al reported that the fluoride-releasing materials were
unable to reduce the demineralization process at the enamel margin close to the restoration following immersion in demineralizing cariogenic solution. However, it is important to note in this study that the frequent change of the demineralizing solution every four hours during the day, exposed the enamel to a high level of acid challenge and hence reduced fluoridated material's ability to resist the cariogenic environment. Moreover, if this acid challenge were simulated in vivo, the patient would be considered a high risk patient and no fluoride-releasing restoration would be expected to withstand this strong acid challenge. Therefore, this type of restorations would not be indicated [16].

In our study, actual inhibition zones were found in six samples (60%) adjacent to the Vitremer resin-modified glass ionomer cement, while no inhibition zones were found in Z100 and Activa BioActive specimens. This observation is in agreement with the findings of a recently published article by Donly and Liu in which the dentin and enamel demineralization inhibition of a bioactive material (Centon N, Ivoclar, Amherst, NY, USA) was studied [17].

The reason behind these findings could be due the amount of fluoride release from the different materials used in the experiment. May and Donly [18], through an in vitro study found that Vitremer demonstrated greater fluoride release than Activa BioActive, while the Z100 demonstrated less fluoride release than Vitremer and Activa BioActive. The results of this study are in agreement with a previous study that evaluated another bioactive material [19]. It can be noted that six samples in the Vitremer group had zero value of adjacent demineralization (Table 2). This could be related to inhibition zones that were created due to fluoride release at the restoration/tooth interface, fluoride release being higher from resin-modified glass ionomer cements than the bioactive material and non-fluoride releasing resin. The 100 micron sections are very thin and only included the adjacent sound tooth structure in the inhibition zone, thus there was a zero value of demineralization. In addition, since the teeth used in this study come from a number of sources, different teeth could have different levels of fluoride exposure. Therefore, some teeth showed no lesion which could be due to higher external fluoride exposure, while others had less or no fluoride exposure and were more susceptible to demineralization. The outcomes of the differences from the mean when the sum of squares was calculated resulted in a higher variance than Vitremer and Activa BioActive, while the Z100 demonstrated less fluoride release [20]. This could be related to inhibition zones that were created due to fluoride release at the restoration/tooth interface, fluoride release being higher from resin-modified glass ionomer cements than the bioactive material and non-fluoride releasing resin. The 100 micron sections are very thin and only included the adjacent sound tooth structure in the inhibition zone, thus there was a zero value of demineralization. Any tooth that had zero inhibition zones was considered a high risk patient and no fluoride-releasing restoration would be expected to withstand this strong acid challenge. Therefore, this type of restorations would not be indicated [16].

In general, it became evident that resin-modified glass ionomer cement inhibited secondary caries at restoration margins making them a possible restorative alternative in a population with high caries risk, such as children or elderly patients [21-22]. However, studies on the bioactive materials and their effect on preventing caries are still limited. Various studies illustrated the capability of fluoride containing restorations to inhibit wall lesions and occasionally produce inhibition zones [17,23]. Previous research has indicated that resin with incorporated calcium fluoride nanoparticles demonstrated fluoride release [24]. Therefore, composites with incorporated nano-calcium fluoride have potential to reduce secondary caries at the restoration margins which is a frequent reason for replacement of restorations.

This study shows Activa BioActive to inhibit enamel demineralization. However, within the parameters of this in vitro study, resin-modified glass ionomer cement remains superior in the ability to inhibit demineralization at enamel margins. The main limitation of this research is that the study has been done in vitro and there may be different challenges in the oral cavity. Furthermore, little research has been previously conducted with this relatively new material, Activa BioActive. Therefore, additional research is recommended to gain further insight to determine that Activa BioActive restorative possesses bioactivity intraorally, which may inhibit adjacent tooth demineralization.

**Conclusion**

Vitremer performed superiorly in inhibiting enamel demineralization at restoration margins, followed by Activa BioActive, which showed better demineralization inhibition when compared to Z100. Further research is needed to evaluate the effectiveness of this new material in vivo.

**References**

13. Banon, R. Comparison of ACTIVATM BioACTIVE versus Compoemer for class II restorations in primary molars: A split mouth


