Evaluation of Microleakage of Self-Adhesive Composite Resin in Pits and Fissures of Extracted Premolar Teeth: An in Vitro Study

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Abstract

Background and objectives: Microleakage at the interface of tooth structure and fissure sealant plays a crucial role in the failure of the treatment. This in vitro study aimed to determine the microleakage of self-adhesive flowable composite resins as pit and fissure sealants.

Material and Methods: Sixty healthy extracted premolar teeth were randomly divided into the four groups (n=15), clinpro sealants (control), flowable composite resin with bonding agent (total-etch), flowable composite resin with a self-etch bonding agent, and self-adhesive composite resin. After thermocycling, the specimens were immersed in 2% methylene blue for 48 hours and then sectioned in the buccolingual direction. The microleakage was assessed by dye penetration using a stereomicroscope at 15X magnification. Data analysis was performed using SPSS version 18.0 and Mann-Whitney and Kruskal-Wallis tests.

Results: There was a statistically significant difference in microleakage score between the groups (P-value = 0.006). The lowest mean of the microleakage level was observed in the flowable composite group with total-etch bonding (group 2) at 1.26±0.96, followed by the Clinpro sealant group (group 1) at 1.62±1.20, and then the self-adhesive composite group (group 4) 1.85±1.00.

Conclusion: Based on the results of this study, the microleakage of self-adhesive composite resin revealed no significant difference with conventional fissure sealant; however, the microleakage of these composite resins was higher than flowable composite resin with a total-etch bonding agent.

Keywords: Dental Leakage [MeSH]; Pit and Fissure Sealants [MeSH], Composite Resins [MeSH], Bicuspid [MeSH]
Evaluation of microleakage of self-adhesive composite resin

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**Highlights**

- The results showed that there was a statistically significant difference between groups.
- The lowest mean of microleakage level was observed in the flowable composite group with total-etch bonding. The highest mean microleakage score was shown in the flowable composite group with self-etch bonding.

**Introduction**

The prevalence of dental caries in children with primary dentition and teenagers by the age of 15 is about 50% and 75% (1). Pits and fissures are one of the most susceptible places to caries because the anatomy of pits and fissures makes them act as a shelter for the causative microorganisms. Although they are inaccessible to plaque control, especially in children who cannot establish proper and regular plaque control and nutritional habits. Fissure sealants may prevent caries lesion development by creating a physical barrier between tooth structure and the oral environment (2, 3).

Various factors have contributed to the failure or success of this treatment. Some of these factors include: microleakage, sealant chipping, previous caries in the groove, and operator's technique (4). Research shows that a high percentage of recurrent caries and sealant loss occurs due to isolation loss that interferes with resin penetration in micropores of etched enamel structure, resulting in inappropriate adhesion and microleakage (5-7).

Microleakage is the seepage of bacteria, liquids, molecules, and ions through the tooth and restoration interface and plays an essential role in the longevity and clinical performance of bonded restorations (8, 9). Microleakage can cause complications such as increased saliva and microorganism penetration, secondary caries, marginal discoloration, pulp inflammation, and post-treatment sensitivity, which can compromise the clinical restoration's durability. Today, using an etchant, bonding systems, and the sealants releasing fluoride are some solutions to reduce the microleakage. Thus, prevention of caries will be more reliable.

Self-etch adhesives are a new generation of bonding agents that reduce clinical steps in applying adhesive, so the possibility of contamination by saliva is eliminated, especially in less cooperative children (10, 11). In 1996, the first generation of flowable composites was introduced. These composites had the advantages of reduced elastic modulus and enhanced flow. Introducing self-adhesive flowable composite resins led to a significant alteration in adhesive dentistry. Combining self-etch technology with self-adhesive composite resin makes them more user-friendly, patient-friendly, and less time-consuming. It also reduces the post-treatment sensitivity, which is favorable in pediatric dentistry (12). There is limited information on the microleakage of self-adhesive composite resins. Since previous studies did not make a consensus about microleakage of self-adhesive flowable composite resins compared to conventional composite resins, and there were few assays analyzing the use of this kind of composite resins as fissure sealants so, The aim of this in vitro study is the comparison between the microleakage of conventional and self-adhesive composite resins as pit and fissure sealants in permanent teeth.

**Materials and Methods**

**Samples preparation**

This experimental in-vitro study was conducted on 60 premolar teeth extracted for orthodontics after obtaining consent forms from patients. The teeth were free of caries, crack, restoration, and developmental defects and stored in distilled water, and then were disinfected in a 0.5% chloramine T for one week. Occlusal surfaces of teeth were brushed and cleaned with Pumice/water slurry and randomly divided into four groups (n=15).
The groups were as follows (Table 1):

- **Group 1**: pit and fissures were sealed with the Clinpro sealant with prior enamel etching (control group).
- **Group 2**: After applying etching and Single Bond, the pit and fissures were sealed with the flowable composite resin.
- **Group 3**: pit and fissures were sealed with the flowable composite resin after applying SE bond (without etching).
- **Group 4**: pit and fissures were sealed with the self-adhesive composite resin.

**Table 1.** The experimental groups

<table>
<thead>
<tr>
<th>Experimental groups</th>
<th>Intervention</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>Etchant+ Clinpro sealant</td>
</tr>
<tr>
<td>Group 2</td>
<td>Etchant+ Single Bond + Flowable composite resin</td>
</tr>
<tr>
<td>Group 3</td>
<td>Single Bond + Flowable composite resin</td>
</tr>
<tr>
<td>Group 4</td>
<td>Self-adhesive composite resin</td>
</tr>
</tbody>
</table>

**Interventions**

In group 1, the occlusal surface of the teeth was etched with 37% phosphoric acid (Condac37-Brazil) for 30 seconds, rinsed with water for 10-20 seconds, and gently dried for 10 seconds. The fissures were sealed with the Clinpro fissure sealant (3M-ESPE, St. Paul, USA) and cured by Cordless LED (a LITEX 696) Light Cure device for 20 seconds.

In group 2, the etching step was performed as mentioned in group 1. The bonding agent, Single Bond (3M-ESPE, St. Paul, USA), was applied in 2 layers with a micro brush. Each layer was dispersed by gentle airflow and light-cured for 20 seconds. Then, the fissures were sealed with the flowable composite Z350 (3M-ESPE, St. Paul, USA) and polymerized for 20 seconds.

In group 3, the SE Bond primer (Kurary, Japan) was first applied with a micro brush on the occlusal surface for 20 seconds and was dried with gentle airflow without washing. Then bonding agent (without etching) was applied, dried with gentle airflow, and cured for 10 seconds. The fissures were sealed with the self-adhesive composite (Vertise, Kerr, USA), which does not require acid etching and bonding protocols, was applied to the fissures by a micro brush for 20 seconds.

For simulating the temperature changes in the mouth environment, all the specimens were thermocycling (Vafaei-Iran) for 500 cycles between 55 °C and 5 °C with a dwell time of 30 seconds. For the dye penetration test; the root apices were sealed with epoxy resin. All the tooth surfaces were covered by two layers of nail polish, leaving 1 mm uncovered around the margins of the sealant. The sealed specimens were then immersed in 2% methylene blue (Merek KGaA-C.I.52015) for 48 hours at pH = 7. After 48 hours, the specimens were washed and sectioned from the CEJ, and the coronal area was mounted in transparent acrylic resin. The specimens were sectioned buccolingually using a diamond disk and evaluated under a stereomicroscope (Ztx-3E) at X15 magnification.

Two observers conducted the survey, and they agreed on the differences. The dye penetration under the sealant was scored as follows: 0 = No dye penetration, 1 = dye penetration limited to the outer half of the sealant, 2 = dye penetration extending to the inner half of the sealant, 3 = dye penetration extending to the underlying fissures.

**Data analysis**

Finally, the microleakage scores were analyzed by SPSS version 18 software. As the data did not meet the laws of normality distribution, pairwise comparisons were performed using the Mann-Whitney U test (p<0.05) for posthoc analysis. Kruskal-Wallis non-parametric test was used to compare the mean ± SD of microleakage scores in experimental groups.

**Results**

The study was performed on 60 healthy premolar teeth divided into four groups. microleakage...
scores based on dye penetration are shown in Table 2. One of the specimens in group 4 was excluded from the study because of a fracture during sectioning. The highest dye penetration score was observed in the flowable composite group with self-etch bonding (group 3). The specimens with no dye penetration mainly were seen in the clinpro sealant group (group 1).

The Kruskal-Wallis test analyzed the mean ± SD of composite microleakage in the study groups. The results showed that there was a statistically significant difference between the 4 groups (P-value = 0.006) (Table 3). The lowest mean of the microleakage level was observed in the flowable composite group with total-etch bonding (group 2) at 1.26±0.96, followed by the Clinpro sealant group (group 1) at 1.62±1.20, and then the self-adhesive composite group (group 4) 1.85±1.00. The highest mean microleakage score was shown in the flowable composite group with self-etch bonding (group 3) at 2.07±0.82.

The data did not meet the requirements of normality distribution; therefore, pairwise comparisons were performed using the Mann-Whitney U test (p<0.05).

Mann-Whitney statistical test was used for comparisons of the groups in pair. The results showed that the difference between the Clinpro sealant group (group 1) and the flowable composite resin group with self-etch bonding (group 3) (P-value = 0.047), between the flowable composite resin group with total-etch bonding (group 2) and the self-adhesive composite group (group 4) (P-value = 0.022) and between the flowable composite resin group with total-etch bonding (group 2) and the flowable composite group with self-etch bonding (group 3) (P-value = 0.001) were significant.

The mean microleakage score of specimens in each group are presented in Table 3.

Table 2. Microleakage scores based on dye penetration in 4 groups

<table>
<thead>
<tr>
<th>Groups</th>
<th>Microleakage score*</th>
<th>Sample size</th>
<th>Score 0</th>
<th>Score 1</th>
<th>Score 2</th>
<th>Score 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinpro sealant (control)</td>
<td></td>
<td>15</td>
<td>5(33.3)</td>
<td>0(0)</td>
<td>7(46.7)</td>
<td>3(20)</td>
</tr>
<tr>
<td>Flowable composite Z350+ Single Bond</td>
<td></td>
<td>15</td>
<td>4(26.7)</td>
<td>4(26.7)</td>
<td>6(40)</td>
<td>1(6.7)</td>
</tr>
<tr>
<td>(total-etch system)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flowable composite Z350+ SE Bond</td>
<td></td>
<td>15</td>
<td>0(0)</td>
<td>0(0)</td>
<td>8(53.3)</td>
<td>7(64.7)</td>
</tr>
<tr>
<td>(self- eteg system)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Self-adhesive composite (Vertise flow)</td>
<td></td>
<td>14</td>
<td>1(7.1)</td>
<td>1(7.1)</td>
<td>8(57.1)</td>
<td>4(28.6)</td>
</tr>
</tbody>
</table>

*: 0 = No dye penetration, 1 = dye penetration limited to outer half of the sealant, 2 = dye penetration extending to inner half of the sealant, 3 = dye penetration extending to the underlying fissures

Table 3. The mean microleakage level of 4 groups

<table>
<thead>
<tr>
<th>Groups</th>
<th>Microleakage score</th>
<th>Sample size</th>
<th>Mean ± SD</th>
<th>median</th>
<th>Min.</th>
<th>Max.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinpro sealant (control)</td>
<td></td>
<td>15</td>
<td>1.62±1.20</td>
<td>2</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>Flowable composite Z350+ Single Bond</td>
<td></td>
<td>15</td>
<td>1.26±0.96</td>
<td>1</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>(total-etch system)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flowable composite Z350+ SE Bond (self- eteg system)</td>
<td></td>
<td>15</td>
<td>2.46±0.51</td>
<td>2</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>Self-adhesive composite (Vertise flow)</td>
<td></td>
<td>14</td>
<td>2.07±0.82</td>
<td>2</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>60</td>
<td>1.85±1.00</td>
<td>2</td>
<td>0</td>
<td>3</td>
</tr>
</tbody>
</table>
Discussion

Microleakage can be caused by improper adhesion and lead to various complications such as pulp irritation and secondary caries (13).

In this study, the minor microleakage level was shown in the flowable composite group with total-etch adhesive followed by the Clinpro sealant, with no statistically significant difference. Etching of enamel surface by total-etch systems enhances surface energy by removal of the smear layer (14), creates a porous surface in the enamel, which cause the resin penetrate to the depth of 25–50 µm of enamel surface (15). After applying the flowable composite as a sealant and the polymerization of resin tags, a solid and durable bond is provided between the sealant and enamel. However, in clinical situations, the etching step can cause problems in the child's cooperation due to the bad taste of the acid; in addition, it is time-consuming and may cause anxiety (16). Besides, omitting the etching step can diminish the errors during etching process.

The reason for the worse performance of groups 3 and 4 in this study was a milder etching effect and lower resin tags depth because of less acidity of self-etch primers compared to phosphoric acid. In addition, the remaining material prevents the polymerization of monomers, which can affect the marginal seal and increase microleakage.

The results of this study are in accordance with the studies by Margvelashvili et al. (17) and Gorseta et al. (14). In the study of Margvelashvili et al., the shear bond strength and the microleakage of self-adhesive composite resin were higher than the conventional sealant but not statistically significant (17). It should be noted that, in their study, etchant was used before applying this type of composites. The Adper L-pop was used as adhesive, a strong self-etch adhesive, while the SE bond (self-etch adhesive) was used in the present study. In the study of Gorseta et al., the type of sealant was different.

Studies by Pitchika et al (18), Schuldt et al (15), were in contradiction with present study. The type of teeth used (third molar) and aging process and preparation of specimens (5 sections of each tooth) were some of the factors leading to increasing the accuracy of these studies. The result of Birlbauer et al. (3) study was in disagreement, either. They investigated three different formulations of self-etch primers in fissure sealant therapy. The microleakage of the control group using the conventional method was less than other groups.

It should be noted that this study was conducted in vitro, so isolation conditions were ideal. Therefore, the microleakage of the flow Z350 and Clinpro composite groups was minimal. In clinical situations, especially when working with children, child cooperation is one of the factors contributing to the success of fissure sealants. When adequate isolation is not possible, using self-adhesive composites can be a good option. In this new generation of composite resins, the three steps procedure of adhesion (etching, priming, and bonding) are simplified into one step. Eliminating the acid etching leads the application of these composites to be more patient-friendly, less time-consuming, and technique sensitive. The sour taste of acid during rinsing etchant could worsen the child's cooperation. Therefore, these composites can be a substitute. Also, the filler amount of Vertise flow is higher than Clinpro, causing more abrasion resistance.

One of the factors affecting the quality and durability of fissure sealant is fissure anatomy. In deeper fissures, the adaptation of resin is higher (17). One of the limitations of the present study was not examining the effect of fissure anatomy on sealants. Although, random dividing of the teeth into four groups eliminated this variable.

Another limitation of this study was the lack of the aging process. Over time, the sealing ability of the fissure sealant decreases. Therefore, performing aging in laboratory studies can simulate situations closer to the clinical conditions.

To uniform the conditions, no preparation was performed on enamel surface. In this study a primeless enamel was evaluated. Perhaps
removing this layer by refreshing enamel with fissurotomy burs will enhance the adhesion of these new composites. In addition, improving the formulation of these composites increases the clinical use of these materials. Generally, differences in the sample size, different methods, different type of dyes and teeth, the frequency of thermocycling and aging process, in most studies cause different results.

Conclusion

According to the limitations of the present study, the microleakage of self-adhesive composites was not significantly different from the conventional fissure sealant. However, the microleakage of these composites was higher than the flow composites with total-etch bonding.

Although the advantage of speeding up the pit and fissure sealing process is an efficient issue, especially in the treatment of children with low cooperation, our study shows that the higher microleakage of self-adhesive composite resins compared to flowable composite resins is an essential deterrent to the use of these products as sealants. We also concluded that further technological advancements are required to improve the sealing ability of self-adhesive composite resins.

Acknowledgement

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Consent for publication

The participant provided written informed consent.

Conflict of interest

The authors declare no conflict of interest, financial or otherwise.

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