Effect of Different Enamel Preparation Methods on Microleakage of Fissure Sealant: An *In Vitro* Study

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Abstract

Objective: Lack of a dental material with optimal adhesion and sealability is an important challenge in modern dentistry leading to marginal leakage. There are controversies on the necessity of enamel preparation in pit and fissure sealant therapy and its effect on decreasing the microleakage; therefore, the present *in vitro* study aimed to assess the amount of microleakage with and without enamel preparation.

Methods: In this experimental study, 30 sound premolars assigned suitable for sealant application, were chosen and randomly divided into two groups. Sealant was applied to all teeth with the same conventional technique. In group A, fissure sealant was applied without enamel preparation while in group B, sealant was applied after fissurotomy with bur. The teeth were thermocycled and microleakage was measured using silver nitrate as leakage tracer. The teeth were then cut into three bucco-lingual sections and examined under a stereomicroscope with 32× magnification. The amount of dye penetration into the sealant was recorded in microns and the degree of microleakage was classified into four degrees of 0, 1, 2 and 3. T-test was applied for the comparison of data between the two groups.

Results: In total,20% of specimens in group B (fissurotomy) had degree 1 and 80% had degree 0microleakage and no specimen had degrees 2 and 3 microleakage, while in group A (no preparation), 20% had degree 1, 33.3% had degree 2 and 46.7% had degree 3microleakage. No specimen had degree 0 microleakage. Therefore, placement of sealant with enamel preparation significantly decreased microleakage ($p<0.001$).

Conclusion: In view of the findings of this investigation, it seems that enamel preparation reduces marginal leakage in pit and fissure sealant therapy.

Key words: Fissure sealant, Microleakage, Pit, Tooth preparation.

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Introduction:

Microleakage is defined as the passage of bacteria, fluids, molecules and ions between the tooth and the over lying fissure sealant. It is an important concern in restorative dentistry (1) because of the related clinical complications such as secondary carious lesions, pulpal pathologies, post-operative pain and tooth hypersensitivity and consequent failure of the restorative procedure (2). Leakage in bonded restorations is very important since presence of microleakage may decrease the bond strength or even lead to debonding of the restoration (3). Sealability of the fissure sealant highly depends on the integrity of its interface with the underlying tooth structure; which per se, depends on the type of surface preparation performed by the clinician. Thus, success assessment must not be limited to the physical, chemical or biological properties of sealant materials alone and surface preparation must be taken into account as well. Surface cleaning and preparation play an important role in success of fissure sealant therapy. Debris trapped in the fissures, air bubbles and the geometrical shape of fissures may all limit the penetration of sealant. Conventionally, the teeth are etched with etchants to cause enamel porosity and enhance resin penetration. However, the ability of etchant to completely remove debris from the fissures is a matter of debate (4). Enamel preparation is performed to increase retention and prevent microleakage (5). In order to achieve an optimal seal, enamel needs to be dry and uncontaminated. Several methods have been developed for this purpose such as conventional prophylaxis using rubber cups with/without pumice paste, invasive techniques like fissurotomy with bur (4) and more recent methods like air abrasion, pumice or aluminum oxide abrasion and hard tissue laser (6). Literature review on this topic revealed contradictory results as some researchers have shown that this technique can reduce microleakage (7-10) while others believe otherwise (11-14). Thus, the current study aimed to assess the effect of enamel preparation via fissurotomy on fissure sealant microleakage in permanent premolar teeth.

Methods:

Thirty sound premolar teeth with no caries, restorations, discolorations, fractures, cracks or obvious hypoplasia extracted for orthodontic purposes within the previous6monthswere selected (4, 5). Sample size was calculated to be 15 specimens in each group considering 95% confidence interval (CI), 90% power and the results of previous studies (4, 15-17). Thus a total of 30 specimens were selected for this study and stored in distilled water (5, 17). Prior to the experiment, all teeth were disinfected using 0.2% Thymol solution for 24 hours.After gross debridement of the teeth, the occlusal surfaces were cleaned with rotary bristle brush and pumice paste without fluoride (Sultan, USA) for 10 seconds. The teeth were then rinsed with air-water spray for 30 seconds, and air-dried for 10 seconds (14).The teeth were coded and randomly divided into two groups of case (group B) and control (group A) (18). Thus, 15 teeth were evaluated in each group.

Group A: Occlusal surfaces remained intact.

Group B: The central grooves were mechanically prepared with sweeping motion of hand piece using fissurotomy bur (S.S. White, USA) (18). All fissures were etched with 35%phosphoric acid gel (3M ESPE, USA) for 15 seconds, rinsed with water spray for 15 seconds, and dried with oil-free compressed air for 15 seconds. Next, light-cure resin-based sealant (Clinpro Sealant, 3M ESPE, USA) was applied to the fissures with a microbrush and cured for 40 seconds using dental light curing unit (Carton/ Swiss) (14). To reach uniformity, distance from the light curing tip to the tooth surface was less than1mmin allcases. The specimens were then stored in saline solution at 37°Cuntil thermocycling (12). To simulate thermal changes in oral environment, all specimens were thermocycled (Vafaii Industrial Co., Iran) in distilled water bath (500 cycles, at 5°C-55°C with dwell time of one minute and transfer time of 30 seconds) (5).The apices of the teeth were then sealed with self-polymerizing acrylic resin. The teeth surfaces were coated with two layers of nail varnish except for0.5-1.0mm around the restoration margins. Samples were immersed in 50% silver nitrate for two hours in a dark room and were then removed, washed and immersed in the processing solution under fluorescent light. The specimens were then embedded in clear self-polymerizing acrylic blocks (AcroPars, Iran) and longitudinally sectioned in buccolingual direction into three slices of 1 mm thickness (mesial, middle and distal sections) with a diamond saw (Mecatome T201A).The specimens were evaluated under a stereomicroscope (Olympus, Japan) at 32× magnification by observers blinded to the surface preparation of specimens twice with one week interval (12).The highest amount of dye penetration was recorded as the degree of microleakage for the respective specimen. Dye and sealant penetration depths were measured in microns by a ruler under stereomicroscope. By calculating the ratio of dye penetration to the depth of sealant, degree of microleakage was determined for each specimen as follows:

0: No dye penetration; 1: Penetration by0 to 1/3 of the sealant depth; 2: Penetration by 1/3 to 2/3 of the sealant depth; 3: Penetration by more than 2/3 of the sealant depth (7). Data were analyzed using SPSS 14.0.2 (SPSS, Chicago, IL, USA). The difference in dye penetration was quantitatively compared between the two groups using t-test. The Mann Whitney U test was applied for qualitative assessment of the two groups with 95% CI.

Results:

Statistically significant differences were detected between the two groups in terms of dye penetration. The degree of dye penetration in group B was significantly lower than that in group A (Table 1).

Table 1- Comparison of dye and sealant penetration depths between the two groups

Groups	Dye penetration depth (microns) X (SD)	Sealant penetration depth (microns) X (SD)
Group A: No preparation	13.4 (4.6)	21.10 (3.90)
Group B: Enamel preparation	0.08 (0.4)	33.5 (5.10)
p-value	*p*<0.001	*p*<0.001

The amount of dye penetration was 13.4 (4.6) µin group A (no preparation) and 0.4 (0.08) µin group B (with preparation). The difference between the two groups in this respect was statistically significant (*p*<0.001). Sealant depth was 33.5 (5.1) µin group Band 21.1 (3.9) µin group A. In other words, sealant penetration in group A was 12.4 µor 37% less than that in group B. According to t-test, this difference was statistically significant (*p*<0.001).

Diagram 1 indicates the degree of microleakage.

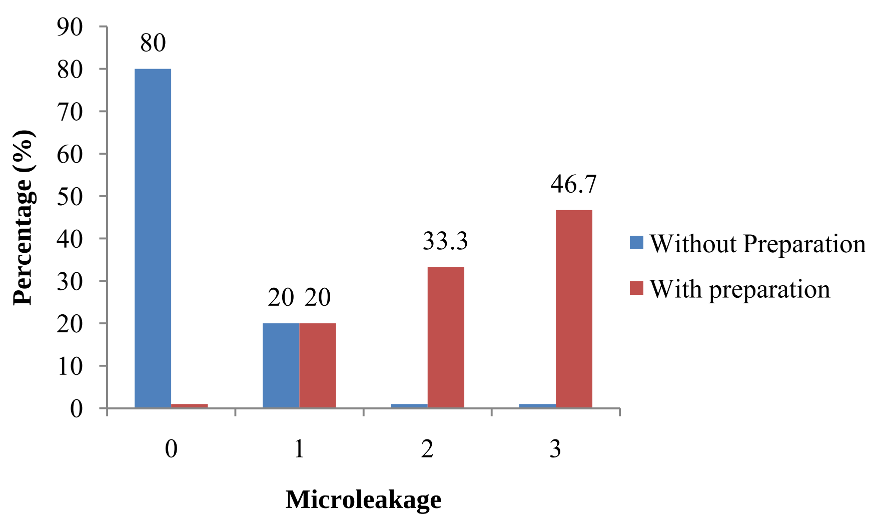

Diagram 1- Comparison of degree of microleakage between the two groups of with and without enamel preparation

As seen in this diagram, degree 1 microleakage was seen in 20%, degree 2 in 33.3% and degree 3 in 46.7% of the specimens in group A; whereas, in group B, 80% of the specimens had no microleakage (degree 0) and 20% had degree 1 microleakage. According to the Mann Whitney U test, the difference in this respect between the two groups was statistically significant (p<0.001).

Discussion:

This study showed that mechanical enamel preparation using fissurotomy bur prior to the application of fissure sealant reduced microleakage at the tooth-sealant interface. However, conflicting results have been reported in this respect as some researchers obtained similar results (7-10, 18, 19), while some others reported otherwise (11-14).

A significant correlation has been reported between the retention of sealant and its cariostatic role (7). When the retention of sealantis optimal, microleakage is reduced and decay is subsequently prevented. Some studies have demonstrated that the retention of sealant decreases to 85% after a year, andto50% after five years (11). Preparation of fissures can increase the retention of sealants. Mechanical preparation widens the fissures and enhances the penetration of sealant into the fissures. As a result, fissure sealant penetrates deeper into the fissures. Mechanical preparation of fissures also facilitates debris removal and increases the surface energy of enamel. All these contribute to minimize microleakage (8). Although various enamel preparation methods are available, no consensus has been reached on the superiority of a specific method. Different burs are used for preparation of fissures like diamond round no. ½ and ¼, diamond Komet bur no.8392, *etc*. (7, 11, 13, 14). We used fissurotomy bur in this study, which is compatible with the shape of fissures. Type of bur used can explain the differences between the results of the current study and those reporting no difference due to enamel preparation. For instance, Youssef and colleagues in 2006 used 1191F diamond bur (12), Lupi-Pegurier *et al.* in 2004 used 8392 diamond bur (13) and Castro *et al.* in 2004 and Srinivasan in 2005 used ¼ round bur (11, 14). Study of microleakage can be carried out using different methods. We used 50% silver nitrate staining solution in our study. Silver nitrate has smaller particles in comparison to other staining solutions (1% methylene blue, 0.2% Rhodamine and 0.5% fuchsin solutions) and consequently has more penetration capacity (12). In fact, silver ions have 0.059 nm penetration capacity resulting in more penetration depth compared to bacteria (0.01-1 µm).The penetration capacity of silver ions is almost similar to that of bacterial products (12). This explains the difference between our results and those of Lupi-Pégurier *et al.* in 2004 and Srinivasan *et al.* in 2005 who used methylene blue for microleakage tracing. Methylene blue has lower penetration capacity; which explains the results obtained by Lupi-Pegurier *et al.* (2004) and Srinivasan, *et al.* (2005) reporting no significant effect due to enamel preparation (13, 14). Youssef *et al.* (2006) used silver nitrate for microleakage tracing but found no significant effect attributed to enamel preparation (12). It should be noted that they compared mechanical preparation of enamel with laser irradiation. In their study etching and preparation were both done with laser; which may be responsible for their obtained results since etching with laser may not be capable of forming efficient enamel porosities. As stated earlier, preparation of fissures may result in elimination of debris and increase the surface energy of enamel; all these factors contribute to reduction of microleakage (8). In a study by Castro *et al.*, the efficacy of laser, bur and air abrasion preparation methods was compared using shear and tensile loads. They suggested the use of bonding agent prior to the application of sealant to enhance shear bond strength. In terms of tensile bond strength, laser in conjunction with bonding agent offered the highest and use of bur in conjunction with bonding agent yielded the lowest tensile bond strength (11). However, an accurate comparison cannot be made between our study and that of Castro *et al*., considering different methodology and application of bonding agent in their study.

In the current study, the specimens were thermocycled to simulate temperature alterations the teeth are subjected to in the oral cavity. Thermocycling can increase microleakage. This issue may also explain the difference between our results and those of Srinivasan *et al*., since they only immersed the specimens in artificial saliva (14).

There are various tools for assessment of the degree of microleakage; among which, we used a stereomicroscope with 32X magnification, which has a high accuracy (8). Widely variable observation methods may be responsible for different results obtained in our study in comparison to those of Srinivasan *et al*. (2005) who used a light microscope with 15X magnification (14), Youssef *et al.* (2006) who used a digital camera with 2X magnification (12) and Lupi-Pegurier *et al.* (2004) who used a digital camera with 26X magnification (13), which might have lower accuracy than the stereomicroscope used in the current study. Another reason for the difference between our results and those of Youssef *et al*. (2006) may be that they used flowable composite along with the bonding agent as fissure sealant material (12); which may explain the lower penetration depth reported in their study.

Conclusion:

It seems that enamel preparation by fissurotomy bur can effectively reduce the microleakage of fissure sealants. Similar *in vitro* studies are required to evaluate and compare the effect of other enamel preparation methods on degree of microleakage.

Conflict of Interest: "None Declared"

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