ABSTRACT

Objective: To assess the effectiveness of adhesive system in reducing microleakage in class II amalgam restorations.

Methods: The teeth were divided randomly into 3 equal groups: Group I was the control experiment (Copal Vanish); Group II: single Bond Universal 3M-ESPE (with and without acid – 37%); and Group III: All Bond Universal – BISCO (with and without acid – 37%), after the amalgam was condensed. The root apices were then sealed with acrylic resin and stored for 24 hours in 100% humidity. After storage, the specimens were subjected to 500 cycles of thermocycling (5°C and 55°C). The teeth were isolated with cosmetic nail polish and were immersed in 0.5 basic fuchsin dye for 24 hours. The teeth then were sectioned with a diamond disc and observed under a stereomicroscope. Microleakage was assessed using a 0-3 scale of dye penetration. The data was analyzed using the Kruskal-Wallis test (p<0.05).

Results: The microleakage scores revealed less leakage in the experimental groups than in the control group. The microleakage for the adhesives without acid conditioning was reduced, although no statistical significance was recorded (p<0.05).

Conclusion: The immediate adhesive material acted effectively as a barrier for microleakage when it was applied without acid.

Key words: Adhesives; Universal adhesive; Amalgams; Microleakage; Class II

Sistema adesivo universal em restaurações de amálgama classe II

RESUMO

Objetivo: Avaliar a eficácia do sistema adesivo na redução da microinfiltração em restaurações de amálgama classe II.

Metodologia: Os dentes foram divididos aleatoriamente em três grupos iguais: Grupo I foi o controle (Copal Vanish); Grupo II: Single Bond Universal 3M ESPE – (com e sem ácido – 37%); e Grupo III: All bond Universal – BISCO (com e sem ácido – 37%) com posterior condensação do amálgama. Os ápices foram então seladas com resina acrílica e armazenado durante 24 horas em 100% de humidade. Após a armazenagem, as amostras foram submetidas a 500 ciclos de termociclagem (5°C e 55°C). Os dentes foram isolados com esmalte para unha e foram imersos em 0,5 de fucsina básica por 24 horas. Em seguida os dentes foram seccionados com disco de diamante e observados sob microscópio estereoscópico. A microinfiltração foi avaliada usando uma escala de penetração do corante de 0-3. Os dados foram analisados utilizando o teste de Kruskal-Wallis (p<0.05).

Resultados: Os escores revelaram menor infiltração nos grupos experimentais do que no grupo de controle. A infiltração para os adesivos sem condicionamento ácido foi reduzida, embora sem significância estatística (p<0.05).

Conclusão: O material adesivo imediato atuou efetivamente como uma barreira para microinfiltração, quando foi aplicada sem condicionamento ácido.

Palavras-chave: Adesivos; Adesivo universal; Amálgama; Micro-infiltração; Classe II
INTRODUCTION

The penetration of fluids and bacteria in the restorative interface remains one of the principal problems of odontology and is directly associated with the longevity of restorations. Marginal micro-infiltration leads to problems such as the early loss of the restoration due to recurring caries, post-operative sensitivity or pulpal damage [1,2,3].

Several approaches have been proposed for the use of amalgam restorations in an attempt to reduce marginal infiltration. There is a greater emphasis on basic materials, modified glass ionomer cements and adhesive resin [4]. Among cavity sealers, adhesive systems have exhibited a better performance than cavity varnish [5].

The use of adhesive systems is supported by the results of in vivo [6-7] and in vitro studies [4-8], which reported significantly greater reductions in micro-infiltration with adhesive systems than with varnish [9-11]. Adhesives form a bond and/or molecular interaction between the amalgam and the dental structure, unlike varnish which only establishes a mechanical connection [12].

Recently, the advent of universal adhesive systems has promised adhesiveness to metallic structures [13,14]. The aim of the present study was to perform an in vitro assessment of the efficiency of two universal adhesive systems in terms of the reduction of marginal micro-infiltration in amalgam class II restorations. Conventional and self-etching adhesive systems were applied.

METHODS

Fifteen non-carious and defect-free human premolars and molars, recently extracted for therapeutic purposes, were selected. Immediately after extraction, the teeth were cleaned of all debris with a curette and an aqueous slurry of pumice, using a soft polishing brush at a low speed. The teeth were stored in 0.1% Thymol solution at room temperature. Subsequently, they were rinsed with water and lightly brushed with pumice slurry at 5 °C and 55 °C using a dwell time of one minute. The teeth of all groups were subjected to 500 cycles of thermocycling (5 °C and 55 °C) at a distance of 1mm. The intensity of the light was monitored periodically with the same light unit, remaining consistently in the range of 400-450 mW/cm².

The amalgam was condensed using small condensers and an air-water spray. The root apices were then sealed with acrylic resin and the teeth were stored for 24 hours in 100 % humidity at 37 °C.

The specimens were stored and polished. Samples from each group were subjected to 500 cycles of thermocycling (5 °C and 55 °C) using a dwell time of one minute. The teeth were isolated with a cosmetic nail polish (1 mm around the restoration) and were then immersed in 0.5 basic fuchsin dye for 24 hours at room temperature. Subsequently, they were rinsed with water and lightly brushed with pumice slurry to remove the superficial dye. Each tooth was sectioned longitudinally in the mesio-distal direction along each restoration using a slow-speed water-cooled diamond saw.

Table 1. Manufacturers, composition, mode of application, and batch numbers of materials used.

<table>
<thead>
<tr>
<th>Material</th>
<th>Manufacturer</th>
<th>Composition</th>
<th>Mode of application</th>
<th>Bath no.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Varnal</td>
<td>Biodinâmica, Europa S.L.</td>
<td>Resin staybielite (Ester 10) and Dimethyltectone</td>
<td>Apply first layer and dry lightly (30s)</td>
<td>673-13</td>
</tr>
</tbody>
</table>
2. Dry lightly (30s)  
3. Apply second layer, brush for 20s  
4. Dry lightly (30s)  
5. Light curing (10s) | 41282      |
| All Bond Universal        | BISCO USA                | Bis-GMA, 10-MDP HEMA, ethanol, initiators, water   | Apply first layer, brush for 20s.  
Dry lightly (30s)  
Apply second layer, brush for 20s.  
Dry lightly (30s)  
Light curing (10s) | B-72020K   |
| Velvalloy spherical       | S.S. White Brazil        | Mercury: alloy 1:0.74, 57.5/42.5%, Single composition spherical alloy: 12wt% Cu, 29wt% Sn, and 60wt% Ag | Activate capsule  
Place capsule in amalgamator  
Tinturate for 12 s  
Condense amalgam | 00R        |
Examinations of the specimens were carried out in random order. All investigators were unaware of the exact nature of the restorative treatment being assessed. If conflicts in scores occurred, a consensus was obtained between the observers.

The staining along the tooth restoration interface was recorded according to the following criteria: 0, no dye penetration; 1, dye penetration along the enamel; 2, dye penetration along the dentine–enamel junction (DEJ), but not including the axial wall; 3, dye penetration to and along the axial wall.

Data were analyzed using absolute frequencies and percentages. To verify the hypothesis of significant differences, the Mann-Whitney test for the comparison of two categories or two groups, as well as the Kruskal-Wallis test, were used to compare microleakage in three different groups filled by three different materials ($p<0.05$).

The margin of error used in the statistical tests was 5%. The statistical software used for data entry and the retrieval of statistical calculations was the Statistical Package for Social Sciences (SPSS), version 21.

RESULTS

Table 2 displays the mean values of dye penetration for the cervical margin location in the different groups. Cohen’s Kappa test confirmed an excellent agreement between the examiners ($k=0.91$). Figure 1 displays the statistical analysis of the different combinations from the experimental design. In general, the interaction between the main factors was not significant ($p<0.05$) for the cervical margins. The microleakage adhesive without acid conditioning provided a reduction, although not a statistically significant one ($p<0.05$).

<table>
<thead>
<tr>
<th>Bonding agent</th>
<th>Mean of post</th>
<th>Etching</th>
<th>$p$ value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>With</td>
<td>Without</td>
<td>With</td>
</tr>
<tr>
<td>Varnish – Control</td>
<td>17.2</td>
<td>3.00 *</td>
<td>3.00</td>
</tr>
<tr>
<td>Single Bond Universal</td>
<td>15.40</td>
<td>10.0</td>
<td>6.50</td>
</tr>
<tr>
<td>All Bond 2 Universal</td>
<td>12.40</td>
<td>10.0</td>
<td>6.00</td>
</tr>
</tbody>
</table>

* Not determined due to the presence of only one category.

(1) Using the Mann-Whitney test to compare the presence or absence of acid conditioning.

Figure 1. (a) Amalgam/single bond universal interfaces with acid/dentin; (a') Amalgam/single bond universal interfaces without acid/dentin; (b) Amalgam/all bond universal interfaces with acid/dentin. (b') Amalgam/all bond universal interfaces without acid/dentin. (c) Amalgam/varnish/dentin interfaces (SEM at 100X and 1000X). There is gap formation along the interface.
DISCUSSION

Marginal infiltration in the bond interface remains one of the principal challenges in odontology. Therefore, studies involving this methodology have become necessary, particularly in in vitro experiments. McCurdy Jr. et al. [15] reported that micro-infiltration is greater in in vitro studies than in in vivo studies, when using pulped teeth. This is probably due to the fluid movement of dentinal tubules, which occurs in the opposite direction to dye penetration. It could also be caused by the oxidation and corrosion suffered by the amalgam, sealing the margins of the restoration more quickly upon contact with oral fluids [16]. Another factor that can hinder, or prevent, dye penetration is the presence of sclerotic dentin [17].

In most in vitro tests, there are variations in the period of storage, which can be 24 hours, a week, two weeks, 3, 6 or 12 months [18,11,19]. The storage time in the present study was 24 hours. Thus, the short-term assessment probably affected the results.

The scientific literature contains studies reporting a reduction in infiltration based on the use of adhesive systems [11-20] over a short-term assessment period. Thus, short-term results are material-dependent, given that long-term assessments have shown that the adhesive is not the dominant factor in terms of reducing infiltration around amalgam restorations [11]. Since adhesive systems are susceptible to hydrolytic degradation, this could explain the reduced bond between dentin and restorative material, thereby favoring the penetration of fluids and bacteria [21]. Moraes et al. [22] assessed the influence of storage time by analyzing marginal infiltration related to the sealing of amalgam restorations. The authors reported that restorations sealed with the Single Bond adhesive exhibited significantly lower amounts of infiltration than the other groups (p<0.001). After 15 months of storage, none of the groups tested exhibited an improved sealing capacity, whereas the group sealed with the Single Bond adhesive exhibited significantly higher scores for dye penetration (p=0.001). Moore et al. [23] showed that the positive influence of adhesives, in terms of the prevention of infiltration, only lasted six months and became insignificant after one year of storage.

The aim of the present study was to use marginal micro-infiltration tests to assess the sealing capacity of two universal/multi-mode adhesive systems (Single Bond – 3M/ESPE; All Bond-BISCO), using conventional and self-etching techniques. The results were compared with sealing conducted using cavity varnish. The use of universal adhesive systems in restorations has been justified based on their affinity with metallic components [13,14]. The results of the present study suggest that the presence of an adhesive agent significantly reduced marginal micro-infiltration, when compared with the control group, in amalgam restorations. All-Bond Universal incorporates 10-methacryloyloxydecyl dihydrogen phosphate (MDP) which, unlike other monomers, bonds chemically with Ca^{2+} ions and forms stable MDP-Ca salts, according to the concept of "adhesion-decalcification". This phenomenon makes the interface more resistant to biodegradation [13]. With regards to the Single Bond Universal adhesive system, studies have shown its capacity for chemical adhesion to teeth, although the results are slightly worse than those found for Clearfil SE Bond. This may be due to interaction between the different chemical components present in the material [24,25].

Analysis of the infiltration of the dye confirmed that none of the three systems applied were capable of eliminating marginal micro-infiltration, although it was lower for restorations associated with dentin adhesives than for those associated with cavity varnish. Oliveira et al. [26] concluded that none of these three restorative systems were capable of eliminating marginal infiltration, similar to the results of the present study. However, marginal infiltration it was lower and statistically significant for restorations associated with dentin adhesives, when compared with cavity varnish. Junior et al. [27] also obtained less marginal infiltration when using the adhesive system All Bond 2/Resinomer than when using a glass ionomer cement (Vitreobond), the latter of which was better than the control group (Copaline Varnish). On the other hand, Guiraldo et al. [4] assessed the marginal adaptation of cavities restored with silver amalgam and adhesive systems and concluded that the worst marginal adaptation was found in the control group (restoration without sealers), with a statistically significant difference when compared with the other groups (G I: adhesive Prime & Bond 2.1 – Dentsply; G II: acid conditioning, application of the adhesive Single Bond and the resin cement Rely X – 3M-ESPE; G III: glass ionomer cement Rely X Luting – 3M-ESPE). The experimental groups did not differ statistically between each other.

When the universal adhesive system was applied in its self-etching mode, the dye penetration scores were reduced by 100% in relation to the conventional mode, for both of the adhesives studied (24 hour storage time). Universal adhesive systems exhibit similar components to single-step self-etching adhesives, in that they possess the functional monomer MDP, thereby enabling the acidic function and posterior chemical adhesion.

Self-etching adhesive systems with a pH of 2.7 partially demineralize dentin, leaving a considerable quantity of hydroxyapatite crystals around and inside collagen fibers. This residual hydroxyapatite is fundamental to the chemical interaction of functional monomers and effective chemical and micromechanical adhesion [28]. The use of self-etching adhesives favors the demineralization and infiltration of the dentin surface at the same depth, theoretically impeding adhesive failure. However, Carvalho et al. [29] reported cases of nano-infiltration along the interface, suggesting that acidic monomers were gradually buffered by the mineral content of the substrate and partially conditioned the dentin. Consequently, areas or partially demineralized, although not infiltrated, dentin was produced below the hybrid layer. This result is contrary to the concept that there is no discrepancy between the depth of demineralization and the depth of adhesive infiltration.

Other factors such as the method of application, the type of solvent and the pH could help to explain the results found herein, particularly in relation to vigorous application. It has
previously demonstrated that the vigorous application of self-etching adhesives improves the immediate bond strength and decreases bond degradation over time [30,31]. The adequate control of dentin moisture is fundamental for the posterior diffusion of resinous monomers, particularly the vapor pressure of the solvent, which is directly associated with adhesive quality [31].

The results obtained in the photomicrography (SEN) suggested that less gaps were formed when adhesive systems were used on the restoration. Iregui [32] used photomicrography to demonstrate the occurrence of micro-mechanical retention between the adhesive and the amalgam, although they also showed that most of the adhesion came from the adhesive/dentin interface. Cavity varnish did not prevent marginal infiltration and provided the worst results overall. This could have been caused by flaws in the layers applied, such as porosities or cracks, or even the dissolution of the material. This is contrary to the findings of Barbosa et al. [33] who found that both conventional and fluoride varnish exhibited low levels of marginal infiltration when used to pre-treat amalgam restorations. Results similar to those of the present study have been previously reported by Pucci et al. [34], Pinto et al. [5], Sanvieiro et al. [35].

Within the limitations of the present study, it can be concluded that the immediate adhesive material acted effectively as a barrier for microleakage when it was applied without acid. This adhesive should be recommended for amalgam restorations.

REFERENCES