Adaptation and radiographic evaluation of four adhesive systems

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ABSTRACT

Objectives: The purpose of this study was to compare microleakage, gap formation, thickness of the adhesive layer and its radiographic appearance associated with four adhesive restorative procedures for class I cavities.

Methods: Adhesive systems with easy handling characteristics were selected for the restoration of class I cavities in extracted third molars. Bite wing radiographs were taken of each tooth and four observers were asked to assess the presence of the adhesive layer. Microleakage, gap width and the thickness of the adhesive layer of each restoration were measured upon sectioning of the teeth.

Results: Microleakage in the experimental restorations was minimal. The thickness of the adhesive layers and gap formation varied among different adhesive systems. The adhesive system with self-etching primer produced the highest percentage gap-free restorations. Thick adhesive layers could be detected on the radiograph. ROC analysis of the results validates the diagnosis from the radiograph.

Conclusions: The four restorative systems performed well in the prevention of microleakage. The use of a resin modified glass-ionomer cement base did not prevent gap formation compared with the all-etch bonding systems used in this study. The presence of an adhesive layer contributed to the prevention of gap formation, independently of the bonding system used. Thick adhesive layers could be detected on the radiograph. © 1997 Elsevier Science Ltd.

KEY WORDS: Marginal adaptation, Bonding, Composites, Radiography

INTRODUCTION

In search for alternatives for dental amalgam the interest for adhesive techniques is growing. Adhesive techniques have the advantage that a maximum of sound tooth structure can be saved. For the treatment of primary occlusal caries a conservative cavity design combined with the use of adhesive restorative techniques can be an alternative to the more traditional class I cavity preparation and restoration techniques. The adhesive cavity design is characterized by a small opening in the enamel which provides access to a more extensive lesion in dentine. Upon excavation, a cavity with undermined enamel margins will generally remain.

To bond the restorative material to the tooth, the acid-etch technique in combination with a glass-ionomer cement or a dentine adhesive can be selected. Both the resin modified radiopaque glass-ionomer cements and the ‘all-etch’ adhesive systems possess easy-to-use handling characteristics compared with the third generation adhesives. The adhesive properties of the fourth generation of all-etch adhesive systems are based on the formation of a resin impregnated (hybrid) layer in the dentine. They provide a stronger bond to the dentine than earlier generations and are easy to handle under clinical conditions as both the dentine and the enamel receive the same treatment. The latest development is an all-etch system using a self-etching primer, replacing a conditioner and primer, for both the enamel and dentinal surfaces.

Dental adhesives may differ in composition, viscosity and mode of application. They can be applied with a
Table I. Restorative groups

<table>
<thead>
<tr>
<th>Group</th>
<th>Base</th>
<th>Etching</th>
<th>Primer</th>
<th>Adhesive</th>
<th>Composite resin</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>—</td>
<td>PA*</td>
<td>—</td>
<td>PB†</td>
<td>CRP‡</td>
</tr>
<tr>
<td>2</td>
<td>—</td>
<td>MA§</td>
<td>SMP‖</td>
<td>SMP‖</td>
<td>CRPt</td>
</tr>
<tr>
<td>3</td>
<td>—</td>
<td>—</td>
<td>LB**</td>
<td>—</td>
<td>CRP†</td>
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<td>4</td>
<td>VM††</td>
<td>PA*</td>
<td>—</td>
<td>PB†</td>
<td>CRP†</td>
</tr>
<tr>
<td>5</td>
<td>—</td>
<td>—</td>
<td>—</td>
<td>—</td>
<td>CRPt</td>
</tr>
</tbody>
</table>

*Superlux Thixo-etch (DMG, Hamburg, Germany, 37% phosphoric acid).
†Clearfil Photo Bond (Kuraray corporation, Osaka, Japan, No. 41154).
§Clearfil Ray Posterior (Kuraray corporation, Osaka, Japan, No. 0028).
‖10% Maleic acid (3M, St. Paul, MN, USA, No. 19930729).
‡Scotchbond Multi Purpose (3M, St. Paul, MN, USA No. 19930729).
**Liner Bond 2 (Kuraray corporation, Osaka, Japan).
††Vitremer (3M, St. Paul, MN, USA, No. 19930512).

brush or a sponge and sometimes need to be air-thinned. As such, differences in material properties and application procedures will affect the thickness of the adhesive layer. A thick adhesive layer may act as an elastic buffer between tooth and composite resin, thereby reducing stress introduced by polymerization shrinkage. However, a thick layer of radiolucent adhesive may show on a radiograph as a radiolucent zone between the cavity wall and a radiopaque composite. This can mistakenly be interpreted as a marginal gap or secondary caries.

In view of the large number of variables involved in adhesive cavity preparation and restoration it was preferred to investigate characteristics of adhesive systems rather than specific materials and procedures. The aim of this study was to evaluate microleakage, gap formation, the thickness of the adhesive layer and the radiographic appearance associated with four adhesive systems in class I composite restorations.

MATERIALS AND METHODS

Sixty extracted sound upper third molars were selected. The teeth had been stored in a 1% chloramine solution immediately after extraction. Standardized cavities with a depth of 3 mm were prepared in the central fissure of the teeth, using a Horico 001025 diamond stone. Dentine was removed using a Horico 001016 diamond stone to simulate the excavation of carious tissue.

The teeth were randomly divided into five groups of 12 teeth, each group receiving a different restorative treatment. Four different adhesive systems with varying composition and handling characteristics were applied. All adhesive systems were combined with the same posterior composite resin (Clearfil Ray Posterior, Kuraray, Osaka, Japan). Curing of materials was performed by a visible light-curing unit (Translux, Kulzer, Wehrheim, Germany, intensity 600 mW/cm²). The five groups were restored according to the following protocols (Table 1).

Group 1

Etching with 37% phosphoric acid was followed by the application of a phosphonated bonding agent (Clearfil Photo Bond, Kuraray).

The entire cavity was etched for 15 s and thoroughly rinsed and dried. Photo Bond was mixed according to the manufacturers instructions and applied into the cavity with a brush. After gentle drying with air, the adhesive was light-cured for 20 s. The composite was subsequently injected into the cavity in two increments using a Hawe Neos Centrix tip. The first layer was applied against the cervical and buccal cavity wall and cured from the buccal and occlusal side for 30 s. With the second layer the restoration was completed. This layer was cured for 30 s from the lingual and occlusal side (Fig. 1a).

Group 2

Etching with 10% maleic acid was followed by the application of a primer and adhesive (Scotchbond Multi Purpose, 3M) which was spread by a gentle airstream.

Since the adhesion to enamel is crucial in this study it was decided to increase the likelihood of good bond strength to enamel by prolonging the application time of maleic acid from the recommended 15 to 60 s. This extension does not jeopardize the bonding to dentine. After thoroughly rinsing and drying, Scotchbond MP primer was applied with a brush and dried until the required shiny surface was established. Scotchbond MP adhesive was applied, spread by gentle
airblowing and cured for 20 s. The cavity was restored as described for group 1.

**Group 3**

An adhesive system, combining a self-etching primer and a light-cured adhesive was used (Clearfil Liner Bond 2, Kuraray). The two-component primer of Liner Bond 2 was mixed and applied into the cavity with a brush. After 30 s, the primer was gently air-dried. The adhesive was applied, spread with a brush and cured for 20 seconds. The cavity was restored as described for group 1.

**Group 4**

The specimens in this group were restored using a radiopaque resin modified glass–ionomer cement (Vitremer, 3M). For this purpose, the traditional 'sandwich technique' was modified as the application of a glass–ionomer lining cement through the narrow opening of the cavity would likely contaminate the enamel walls and prohibit proper etching.

The primer was applied, left undisturbed for 30 s, gently dried with air and cured for 20 s from the occlusal surface. The Vitremer was mixed according to the manufacturer's instructions and syringed into the cavity using a Hawe Neos Centrix tip as if it were a restorative material. The material was cured for 30 s from the buccal and 30 s from the lingual side of the tooth. After setting of the cement, excess material was removed from the entrance of the cavity to a depth of 1.5 mm using a diamond stone (836012 Meisinger, Düsseldorf, Germany) under water coolant. After drying, the cavity was etched for 15 s with 37% phosphoric acid, rinsed and air-dried. Photo Bond (Kuraray) was mixed according to the manufacturer's instructions and applied into the cavity with a brush. After air-drying, the adhesive was cured for 20 s. The remaining cavity was filled in bulk with composite, which was injected into the cavity and cured for 30 s from the occlusal side (Fig. 1b).

**Group 5**

In this positive control group, etching and bonding was omitted to induce gap formation and marginal leakage. The composite was injected into the cavity which was filled in bulk and was subsequently cured for 60 s from the occlusal surface.

All restorations were finished with a fine grit diamond stone (830 C 016, Meisinger, Düsseldorf, Germany) and stored in water for 24 h before thermocycling (500 cycles at 5–55°C). The teeth were mounted in artificial jaws to simulate the *in vivo* situation, and bitewing radiographs were exposed (D-speed film, 15 mA, 70 kV, 0.3 s).

The teeth were immersed in a dye solution (basic fuchsin) for 24 h and embedded in acrylic resin (Fastacryl). Each tooth was sectioned twice with a diamond saw (Bronwill, Rochester, NY, USA) along the fissure system yielding two tooth parts and a 1 mm thick section. Hence, four section planes per restoration were available for validation. Dye penetration was scored on a five-point ordinal rating scale: 0, no dye penetration; 1, dye penetration restricted to the enamel; 2, dye penetration into the dentine not reaching the cavity floor; 3, dye penetration reaching the cavity floor; 4, leakage beyond the cavity floor into the pulp. Microleakage was assessed jointly by two observers until both observers arrived at the same decision. The most unfavourable dye penetration score from the four section planes was selected to represent the dye penetration score for the entire restoration.

The 1 mm thick section was viewed under a transmitting light microscope (Zeiss, W, Germany) at ×160 magnification to measure the gap width (GW) and the adhesive layer thickness (ALT). GW was measured in all groups, whereas ALT was measured in groups 1–3 only, at five predefined sites along the tooth restoration interface (Fig. 2).

Differences in microleakage and the mean width of gaps and thickness of adhesive layers between groups were tested for statistical significance using a Mann–Whitney *U*-test at *p*<0.05 and with correction for multiple group testing. To investigate the relation between thickness of the adhesive layer and gap width, all ratings for gap width and layer thickness were dichotomized. Gaps were considered 'absent' when no gap was measured and 'present' when any gap was measured. Likewise, an adhesive layer was considered 'absent' when no layer was measured and 'present' when any layer was measured. The dichotomized ratings for gap and layer presence were cross-tabulated in a four-fold contingency table (Fig. 3). The risk of a gap being absent given a presence of the adhesive layer is expressed as $a/n_1$. Likewise, the risk of a gap being present given the presence of an adhesive layer is
The null-value of RR is 1.00, indicating that no consistent relation exists between adhesive layer and gap presence. The 95% confidence interval for RR is defined:\(^{14}\)

\[ RR = \frac{a n_0}{b n_1} \]

The bitewing radiographs were examined independently by three dentists, all experienced in the field of operative dentistry. The observers were asked to assess the presence of a radiolucent layer in relation to each restoration using a four-point ordinal likelihood rating scale: 0, translucent layer very likely present; 1, translucent layer likely present; 2, translucent layer likely absent; 3, translucent layer very likely absent. The inter-examiner reliability was expressed in kappa values. The mean of the ratings by three observers was determined for all restorations with an actually present adhesive layer as observed from the sections. Subsequently, the mean of the ratings by three observers was considered 'small' when less than the median, and 'large' when equal to or larger than the median. ROC analysis plots sensitivity as a function of (1-specificity), whereas the area under the ROC curve indicates the accuracy of the observations independent of the cut-off at the radiographic rating scale\(^{16}\).

**RESULTS**

The results for dye penetration, gap width and thickness of the adhesive layer are shown in Table II. Leakage into the dentine was only found in the positive control group. Specimens from the control group 5 had the largest mean gap width (15.46 µm) and no gap-free restorations. Mean gap width of this group was statistically significantly different from those in all other groups.

Specimens from group 3, restored with the adhesive system with the self-etching primer had the smallest mean gap width (0.29 µm). Eighty-three percent of the restorations in this group were gap free. Mean gap width of group 3 was statistically significantly different from that in group 1, yet not statistically significantly different from those in groups 2 and 4.

Group 1 specimens had the thinnest adhesive layer (mean thickness 23 µm), which was statistically significantly thinner than the layers in groups 2 (mean thickness 93 µm) and 3 (mean thickness 140 µm).

Table III contains the RRs for the presence of gaps and adhesive layers in groups 1-3, the crude RR and their 95% confidence intervals. Because the RRs for the methods were smaller (group 1) as well as larger (groups 2 and 3) than that of the crude RR, possible effects of the restorative procedures on the presence of gaps were not confounded with those with the presence of the adhesive layer\(^{14}\). RRs for groups 1 and 2 and the crude RR were statistically significantly different from 1. A crude RR of 11, indicating that the risk of gaps being present given the presence of an adhesive layer is 11 times smaller than the risk of gaps being present given an absent adhesive layer, demonstrates that the presence of a visible adhesive layer considerably reduces the risk of gap formation.

The results from the radiographic observations are contained in Table II. Kappa values for inter-examiner agreement were 0.31, 0.41 and 0.47. From the restorations with an actually present adhesive layer as observed from the sections, 20 restorations received a mean rating equal to or lower than 2, whereas 16 restorations had mean ratings higher than 2. The medians of the distribution of ALT+GW for these restorations were 124 and 42 µm, respectively. Thus, 42 µm was selected as the threshold beyond which a translucent zone between restoration and cavity wall was considered detectable from the radiographs. Table IV contains the results from the ROC analysis and Fig. 4 depicts the ROC curves reflecting the observers performance to detect a translucent zone. The areas under ROC for all observers as well as a mean area under ROC of 0.76 indicate substantial accuracy of the observations.
Table II. Dye penetration, mean gap width, % gap-free restorations, adhesive layer thickness and radiographic score in five groups of 12 teeth, restored with five different adhesive systems

<table>
<thead>
<tr>
<th>Group</th>
<th>Dye penetration</th>
<th>Mann-Whitney U-test*</th>
<th>Mean gap width (S.D.) (µm)</th>
<th>Mann-Whitney U-test*</th>
<th>% Gap free restorations</th>
<th>Adhesive layer thickness (S.D.) (µm)</th>
<th>Mann-Whitney U-test*</th>
<th>Radiographic score</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0 1 2 3 4</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>8 4</td>
<td>ab</td>
<td>4.17 (4.25)</td>
<td>b</td>
<td>25</td>
<td>22.97 (21.56)</td>
<td>a</td>
<td>7 23 9 9</td>
</tr>
<tr>
<td>2</td>
<td>8 4</td>
<td>ab</td>
<td>1.26 (2.67)</td>
<td>ab</td>
<td>75</td>
<td>92.84 (79.39)</td>
<td>b</td>
<td>23 12 5 8</td>
</tr>
<tr>
<td>3</td>
<td>1 11</td>
<td>b</td>
<td>0.29 (0.72)</td>
<td>a</td>
<td>83</td>
<td>140.07 (53.11)</td>
<td>b</td>
<td>33 9 4 2</td>
</tr>
<tr>
<td>4</td>
<td>10 2</td>
<td>a</td>
<td>1.50 (1.85)</td>
<td>ab</td>
<td>33</td>
<td>79.64 (79.39)</td>
<td>b</td>
<td>6 12 15 15</td>
</tr>
<tr>
<td>5</td>
<td>1 11</td>
<td>c</td>
<td>15.46 (6.22)</td>
<td>c</td>
<td>0</td>
<td>40.07 (53.11)</td>
<td>b</td>
<td>7 14 18 9</td>
</tr>
</tbody>
</table>

*Results within the same level of significance are marked identically (Mann-Whitney U-test; P<0.05).

Table III. Risk ratios (RR) and 95% confidence intervals for groups 1–3

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
<th>Crude</th>
</tr>
</thead>
<tbody>
<tr>
<td>a</td>
<td>21</td>
<td>3</td>
<td>0*</td>
<td>24</td>
</tr>
<tr>
<td>b</td>
<td>6</td>
<td>2</td>
<td>2</td>
<td>10</td>
</tr>
<tr>
<td>n&lt;sub&gt;1&lt;/sub&gt;</td>
<td>26</td>
<td>6</td>
<td>0*</td>
<td>32</td>
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<tr>
<td>n&lt;sub&gt;0&lt;/sub&gt;</td>
<td>34</td>
<td>54</td>
<td>60</td>
<td>148</td>
</tr>
<tr>
<td>RR</td>
<td>4.58</td>
<td>13.5</td>
<td>14.75</td>
<td>11</td>
</tr>
<tr>
<td>(\chi^2)</td>
<td>23.32</td>
<td>14.9</td>
<td>1.24</td>
<td>79</td>
</tr>
<tr>
<td>95% Confidence interval</td>
<td>2.47–8.49</td>
<td>3.60–50.62</td>
<td>0.13–1669.15</td>
<td>6.49–18.72</td>
</tr>
</tbody>
</table>

*In calculating RR's, 0 was substituted by 0.1.

Table IV. Results from ROC analysis

<table>
<thead>
<tr>
<th>Observer</th>
<th>Area under ROC (S.D.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.81 (0.06)</td>
</tr>
<tr>
<td>2</td>
<td>0.70 (0.08)</td>
</tr>
<tr>
<td>3</td>
<td>0.83 (0.06)</td>
</tr>
<tr>
<td>4</td>
<td>0.72 (0.08)</td>
</tr>
<tr>
<td>Mean</td>
<td>0.76 (0.04)</td>
</tr>
</tbody>
</table>

DISCUSSION

The intention of this study was to evaluate combinations of adhesive materials and application techniques (adhesive systems) with respect to marginal leakage, gap formation and radiographic appearance. The systems were designed such that they would be comparatively easy to handle in clinical practice. It should therefore be stressed that differences found in this study between adhesive systems cannot be attributed to single variables constituting a system such as the mode of application or the materials used.

In this study materials were selected on their handling characteristics. Three all-etch systems with different application techniques and a glass–ionomer composite ('sandwich') restoration were evaluated on the aspects of gap formation in relation to the presence and thickness of the adhesive layer. To study gap formation and marginal leakage four adhesive systems were applied. Marginal integrity is considered important in preventing secondary caries and pulpal disease. All restorations in groups 1–4 showed no marginal leakage or only minimal leakage restricted to the enamel. All restorations in the positive control group showed severe dye penetration, indicating that the method to validate microleakage used in this study is acceptable. It can therefore be concluded that all applied restorative techniques prevent marginal leakage into the dentine under in-vitro conditions. This finding is consistent with that of another study which also found only minimal marginal leakage in similar occlusal composite restorations.

Although restricted to the enamel, almost all restorations in group 3 using the self-etching primer, showed
dye penetration, whereas the acid etched composite resin restorations of group 1, 2 and 4 showed virtually no dye penetration. It is hence questionable if a self-etching primer is as effective as phosphoric acid or maleic acid etching in achieving a tight bond to enamel. This finding seems to be conflicting with the high bond strength values for Liner Bond 2 of 28.2 MPa to enamel and 19.4 MPa to dentine. However, the results of bond strength tests have shown to correlate poorly with the behaviour of adhesive systems in restorations.

The applied method for detecting a translucent zone using a transmitting light microscope has the restriction that the observations of layers and gaps thinner than about 1 μm are not accurate. Therefore, when no adhesive layer was observed, the presence of a very thin layer is still possible. The applied method for detecting a translucent zone under composite restorations is not used until now. Kappa values for inter-examiner agreement indicate fair to moderate agreement between observers.

Especially in class 1 cavities with their unfavourable cavity design, polymerization stress may lead to gap formation at the resin–dentine interface, because the bond strength to dentine is lower than the cohesive strength of the restorative material and its bond to enamel. Therefore, although marginal leakage reaching the dentine is not observed with any of the adhesive systems, the long-term success of adhesive restorations is also determined by the presence or absence of gaps at the tooth–restoration interface. When the restoration is loaded by an opposing tooth or during mastication of solid food, deformation of the restoration may occur and, as a result, the dimensional change of the gap may cause pain, due to percolation of fluids in the dentinal tubules. From that point of view it is considered important to minimize gap formation during polymerization. In this study, the specimens in group 3 combined 83% of gap-free restorations with the highest mean adhesive layer thickness. Moreover, specimens in group 3 showed significantly less gap formation than those in group 1, which had a comparatively thin adhesive layer.

In a recent study it was assumed that the adhesive bonding resin, which is characterized by a low modulus of elasticity, compensates for polymerization shrinkage, thus preventing microleakages. Accordingly, the RRs obtained from this study indicate that the presence of an adhesive layer significantly contributes to the prevention of gaps in groups 1 and 2. The results of this study show that thick adhesive layers are more capable of preventing gap formation than thin layers, indicating that differences in the occurrence and size of gaps between groups 1, 2 and 3 can at least in part be attributed to differences in adhesive layer thickness. However, differences between the adhesive systems cannot only be attributed to the thickness of the adhesive layer but will also be influenced by the quality of the bond of the applied system. In group 1 the acid-etch technique was combined with the use of an adhesive without application of a dentine primer. The additional use of a dentine primer in combination with Photo Bond would probably have improved the results as is shown by another study.

The teeth in group 4 were restored using a resin modified glass–ionomer cement combined with a superficial layer of composite resin. From the samples of these restorations many tiny porosities were observed throughout the glass–ionomer material. These were probably introduced by the mixing procedure. Both the porosities and the relatively low modulus of elasticity of the glass–ionomer cement could have contributed to a relief of shrinkage stress. However, only 33% of the restorations in group 4 were gap free. Apparently, the bond strength of this glass–ionomer material to dentine did not withstand the polymerization stresses of the material itself. The presence of gaps in this group is not likely to be caused by the polymerization shrinkage of the composite, as its volume was relatively small.

Radiographic assessment of the restoration interface revealed that translucent zones larger than 40 μm were detectable from radiographs. ROC analysis of these assessments showed only small differences in accuracy between the four observers. A mean area under ROC of 0.76 indicates that the presence of a fairly thin adhesive layer can be accurately detected from radiographs. Because the observation of a translucent zone on a radiograph can be associated with either the presence of a thick adhesive layer, secondary caries or a gap, it remains uncertain if such a restoration requires replacement. Therefore, in aiding restorative treatment decision making, the adhesive layer should either be limited in thickness or the adhesive should be made radiopaque. Meanwhile, dentists should remain cautious about replacing restorations only because a radiolucent zone is seen on the radiograph.

Contrary to the specimens in group 3 which had measurable adhesive layers at all sites, specimens in groups 1 and 2 showed many sites where the adhesive layers were too thin to be measured. This may be due to a difference in application technique, since the adhesives in groups 1 and 2 were spread with air, whereas the adhesive in group 3 was spread with a brush. Differences in the viscosity of the adhesive resins may also have influenced the thickness of the adhesive layers, as high viscous resins, applied into a cavity by a brush, may stick to the brush in larger volumes. Furthermore, the design of the cavity will hinder the removal of an excess of resin. For this reason it is likely that high viscous resins produce thicker adhesive layers. Further investigations should highlight the influence of the viscosity and the application method of an adhesive on adhesive layer thickness and its ability to provide a durable bond.

Although the use of Photo Bond and Clearfil Ray Posterior resulted in gaps under restorations in several specimens, the clinical significance of this finding is uncertain as these materials performed very well in a
three year clinical study\textsuperscript{24}. \textit{Also in an in vivo study the combination of phosphoric acid etching, Photo Bond and Clearfil Ray Posterior showed very good results regarding the prevention of microleakage\textsuperscript{25}. However, the clinical relevance of microleakage found in \textit{in vitro} studies can be disputed as is stated by Wilson, who concluded that in a five year clinical study on the longevity of posterior composite restorations no negative effects due to the presence of microleakage were found\textsuperscript{26}. Thus, dentists should focus on aspects other than marginal leakage alone to select an adhesive system. The aspects focused upon in this study, i.e. handling characteristics and radiographic assessment of the margins, will probably be more appealing to dentists, but have no clinical history yet.

It can be concluded that all four restorative systems evaluated performed well in preventing microleakage. The presence of an adhesive layer contributed to the prevention of gap formation. The thickness of the adhesive layers varied among the adhesive systems and layer thicknesses larger than 40 \textmu m could be accurately detected from radiographs.

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\section*{References}


