Effect of loading on the microtensile bond strength and microleakage of a self-etching and etch-and-rinse adhesive in direct class II MOD composite restorations in vitro

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The effect of loading on microleakage and microtensile bond strength of MOD composite restorations bonded with either self-etching or etch-and-rinse adhesive was investigated. MOD cavities were prepared in 28 extracted molar teeth. 14 teeth were bonded with a one-step self-etching adhesive (G-Bond) and 14 with an etch-and-rinse adhesive (Optibond Solo Plus) then restored with resin composite. For each adhesive, 7 teeth were loaded and 7 unloaded (controls). Loading was achieved with an axial force of 80 N at 2.5 cycles/s for 250,000 cycles. All the teeth were stored in 0.25% rhodamine solution for 24 h and sectioned in a bucco-lingual direction at the proximal boxes to examine microleakage then further sliced mesiodistally into beams for the μTBS test. Failure modes were determined using confocal and scanning electron microscopy. ANOVA assessed the effect of loading on microleakage and bond strength. After loading, restorations bonded with G-Bond exhibited significantly greater dye penetration compared to Optibond Solo Plus at both the axial walls and cavity floor. On the other hand, loading significantly reduced the μTBS of Optibond Solo Plus, whereas it had no significant effect on the μTBS of G-Bond.

Keywords: Loading, Bond strength, Microleakage, Composite restorations

INTRODUCTION

As clinicians move away from amalgam to “metal-free” restorations in response to patient demands, direct resin composite is being used in increasingly larger sized cavities such as those involving the mesial, occlusal and distal surfaces (MOD cavities) of posterior teeth. When restoring a posterior tooth with direct resin composite, the clinician must first decide which type of adhesive to use. The subsequent bond formed between the adhesive and the cavity margins and floor must be able to resist temperature changes, chewing loads and chemical assaults within the oral cavity(1). It has been reported that deterioration of the marginal seal of composite restorations is a major influence on the longevity of a composite restoration and the subsequent development of marginal leakage remains the most frequent reason for replacing such a restoration(1).

Developments in material technology can be quite rapid and therefore there is still a need for in-vitro testing of adhesives and composite restorations(2). However, it is important that in-vitro testing of adhesive restorations tries to simulate the oral environment and tests that involve accelerated aging through water storage, thermocycling and cyclic loading have been developed(3,4). Mechanical loading of direct composite restorations bonded with self-etching and etch-and-rinse adhesives has been used due to its potential to simulate but not replicate the chewing cycle(5,6). These studies have been carried out on flat surfaces, cervical cavities or class II MO cavities in bovine or human incisor teeth(3-5). However, there has been to date no published information on how the adhesive-dentine bond beneath large MOD resin composite restorations in posterior teeth respond is affected by loading in vitro.

In vitro mechanical load cycling under water is an important method for evaluating their clinical potential(5,6). Following loading of a restoration, several factors may be assessed such as the strength of the adhesive bond or leakage occurring at the margin of the restoration(7,8). The validity of microleakage evaluation as a predictor of the clinical performance of materials has been called into question. It has been suggested that research should be focussed on laboratory tests, which might be able to predict the clinical performance of restorative materials despite the fact that no laboratory test can simultaneously reproduce all the conditions encountered in the oral environment(9). Since clinical trials often include marginal integrity among the evaluation criteria(7,8), combining microleakage measurements with another in vitro test such as bond strength testing within the same tooth may provide useful information on the durability of the adhesive interface in large intra-coronal direct composite restoration. Moreover, it has been reported that bond strength testing can reveal valuable clinical information and if an aging factor is introduced, then the durability of adhesion can be assessed(7,8).

Therefore, the aim of this experiment was to investigate the effect of mechanical loading on the microleakage and microtensile bond strength of MOD direct composite restorations bonded with either a one-step self-etching or a two-step etch-and-rinse adhesive. The null hypothesis was that loading would have no effect on the bond strength and microleakage of the
tested adhesives.

METHODS AND MATERIALS

Specimen preparation
Twenty-eight non-caries human lower third molars, extracted in accordance with the local ethical committee rules (King’s College London Dental Institute research ethics committee approval 04/Q0704/57) and after obtaining informed consent of the patients, were used in the study. Only caries-free upper third molars with no visible cracks and of similar dimensions were used. The teeth were stored in tap water at 4°C, undisinfected, in order to avoid chemical media-induced artefacts and were used within 1 month of extraction.

The root of each specimen was dipped in a two-part silicone model duplicating material (SP8016 Hard, Bracon Dental Laboratory Products, Sussex, UK) up to 2 mm below the cement-enamel junction and left to dry, so that each root was surrounded by a simulated periodontal membrane. Each specimen fitted the centre of the test chamber of the mechanical loading device, so that each tooth would be loaded in the centre of each restoration, through the vertical axis of the tooth. In order to ensure this position was maintained, an aligning method was developed and a dental milling machine was used (Galloni, Milano). On a co-polyester translucent disc (Erkodent, Pfalzgrafenweiler, Germany), a circular outline was scribed with the same diameter as the test chamber of the loading device. This disc was used to determine the centre of the polysiloxane matrix. Then a stainless steel rosehead bur was attached to the drill chuck of the milling machine and set in the centre of the polysiloxane matrix. Each tooth was fixed with red beading wax (Remdent, Dental Products Ltd, Wiltshire, UK) to the rosehead bur. Autopolymerising acrylic resin (Mr Dental, cold cure modelling acrylic, Surrey, UK) was poured in the polysiloxane matrix from a height of 10 cm, to ensure uniform filling of the matrix. Then the root base of each tooth was embedded in the acrylic resin to complete stabilization of the tooth.

Preparation design
Figure 1 illustrates how the specimens were prepared. A cavity was cut into the dentin of each tooth with a diamond bur (FG 845C; Sybron Kerr, CA, USA) mounted on a high-speed hand piece under water coolant to receive an MOD restoration. The dimensions of the preparations were 4 mm buccolingually, 3 mm deep at the isthmus and 4 mm deep at the mesial and distal boxes and the boxes were also 1.5 mm at the base towards the pulp. The cavities were prepared 1–1.5 mm above the CEJ and the boxes were prepared with butt margins gingivally. All the internal line angles were smoothed to reduce the possibility of stress concentrations. The burs were replaced after every four preparations in order to ensure high cutting efficiency. All cavity dimensions were strictly standardized during preparation by securing the specimens to a microscope stage converted into a specimen holder/cutting guide. Each cavity was cut by keeping the position of the handpiece constant and slowly moving the stage in the x, y and z directions.

Fig. 1 Schematic showing method of loading and specimen preparation for microleakage evaluation and microtensile bond strength testing.
All the prepared cavities were randomly divided into two groups of 14 teeth each, which were assigned to be bonded with either a one step self-etching adhesive (G-Bond, GC Corporation, Tokyo, Japan) or a two-step etch-and-rinse adhesive (OptiBond Solo, Kerr, Orange, USA), which were applied according to manufacturers’ instructions (Table 1).

A clear matrix band was then adapted to the prepared tooth before incremental insertion of the restorative material (Gradia Direct Posterior, GC Corporation, Tokyo, Japan). 1.5 mm thickness increments were placed, in an oblique layering technique. Each increment was polymerised using a halogen curing light according to the manufacturer’s instructions for 40 s. The restorations were finally finished with diamond burs and silicone polishing points (CompoMaster, Shofu, Tonbridge, UK).

**Mechanical loading**

Seven specimens in each group were fatigued in a water bath maintained at 37°C (JB1, Grant Instruments Ltd, Sherpreth, UK). The fatiguing regime consisted of 250,000 cycles of 80 Newton loads, at a rate of 2.5 loads per second. Static loading was applied vertically via a 2 mm-wide, round ended, stainless steel shaft attached to a LAL90 linear actuator (SMAC Europe Ltd, Horsham UK), which generates force and motion using speaker coil technology to the midpoint of the composite restoration both mesio-distally and bucco-lingually. The actuator was operated via computer coding stored in a LAC1 controller (SMAC Europe Ltd) linked to a computer by a RS232 interface, so allowing the input of loading parameters using the HyperTerminal programme (Hilgraeve Inc, Monroe, Michigan, USA). The remaining 7 teeth in each group were stored in water at 37°C for an equivalent time span. Figure 1 shows how the specimens were loaded and prepared for microleakage and microtensile bond strength evaluation.

**Microleakage evaluation**

The teeth were sealed with two layers of nail varnish up to 1.0 mm from the restoration margins after the root apices were sealed with wax. They were then immersed in a 0.25% solution of Rhodamine B in distilled water, for 24 h. After storage, the teeth were thoroughly cleaned in an ultrasonic water bath (Biosonic, Coltène/Whaledent Inc, Cuyahoga Falls, Ohio, USA). Each tooth was sectioned twice bucco-lingually with a diamond wafering blade (Benetec Limited, London, UK), yielding two end tooth sections close to axial wall-gingival floor line angles of the proximal boxes. In preparation for examination using confocal microscopy, the sections were manually wet-polished using 1000 grit carborundum paper (Struers, Solihull, UK) for 20 s each. After polishing, the sections were ultrasonicated (Biosonic, Coltène/Whaledent Inc, Cuyahoga Falls, Ohio, USA) in distilled water for 3 min each. Microscopy followed, in which each specimen was examined with a tandem scanning confocal microscope (TSM, Noran Instruments, Middleton, WI, USA) using a ×20/0.80 NA oil immersion objective lens. Following calibration of an acetate measuring sheet to the objective lens’ output to an iXon 885 EM-CCD camera (Andor Technology, Northern Ireland, UK), each margin could be scored for leakage. Detection of Rhodamine infiltration was possible via suitable emission and excitation filters: 546 nm (green) and 600 nm (red) respectively. Using the CCD in fixed-gain mode in order to isolate the fluorescent signal, images were relayed to an LCD monitor via iQ capture software IQ (Andor Technology, Northern Ireland, UK). Leakage was measured using the calibrated scale on the acetate sheet. Leakage was measured in microns on each wall separately and expressed as a percentage. Leakage into enamel and dentin was not considered independently. For the axial walls, each tooth had four walls measured; buccal and lingual walls of both the mesial and distal specimens. For the cavity floor, measurements for the mesial and distal slices were considered together.

**Microtensile bond test**

The remaining slabs were further sectioned vertically in a mesio-distal direction then a bucco-lingual direction to obtain beams with an approximate surface area of 1 mm². The dimensions of each beam were checked using a digital caliper before the microtensile bond test was performed. Then, each specimen was attached to a customized microtensile jig with cyanoacrylate adhesive (Zapit; Dental Ventures of America, Corona, CA, USA) and mounted on a linear actuator (LAL 300; SMAC Europe) for microtensile bond strength testing at a crosshead speed of 1 mm/min.

**Statistical analysis**

Data were analysed using Stata 8 (Stata Corp, TX, USA) software. The microtensile bond strength data were

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**Table 1 Adhesives used, their composition and application method**

<table>
<thead>
<tr>
<th>Adhesive</th>
<th>Composition</th>
<th>Application Procedure</th>
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</thead>
<tbody>
<tr>
<td>G-Bond GC, Tokyo, Japan</td>
<td>Acetone, 4-MET, pHA-m, UDMA, filler, photoinitiator, water</td>
<td>After shaking bottle, apply to cavity surface for 10 s. Air-dry for 5 s then light-cured for 10 s</td>
</tr>
<tr>
<td>OptiBond Solo Plus, Kerr, Orange, CA, USA</td>
<td>Ethanol, HEMA, Bis-GMA, GPDM, silica, barium glass, sodium hexafluorosilicate</td>
<td>Etch with H₃PO₄ for 15 s, apply to enamel and dentin for 15 s using a light brushing action. Light-cure for 10 s</td>
</tr>
</tbody>
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**Note:**

- **G-Bond:** A two-step etch-and-rinse adhesive.
- **OptiBond Solo Plus:** A one-step self-etching adhesive.
- Note the differences in the application procedures and the materials used in each step.
analysed initially using two-way analysis of variance (ANOVA) with ‘beam’ nested within ‘teeth’ within ‘adhesives’. Subsequent post-ANOVA contrasts were performed using a Bonferroni adjustment for multiple comparisons. Percentage micro-leakage showed non-Normal distribution of the data. The data was described using medians and inter-quartile ranges and differences between treatments and between loaded and unloaded conditions were analysed using Mann-Whitney-U tests. For this, proportional values for four axial walls and two cavity floors in each tooth were measured and then a mean value for “axial walls” and “cavity floor” obtained. Differences in the mode of fracture were analysed using chi-square tests. A $p$ value less than 0.05 was regarded as indicating statistical significance.

RESULTS

The results of the microtensile bond strength test and microleakage evaluation are presented in Tables 2, 3 and 4 respectively. For each group, the maximum number of beams that could be harvested for testing was 42. The pre-test failures are included in the mean bond strength values and occurred as follows: G-Bond Unloaded, 12; G-Bond Loaded, 21, Optibond Solo Unloaded, 11; and Optibond Solo Loaded, 12.

When the teeth were unloaded, there was no significant difference in bond strength between the two adhesives. After loading, there was a significant reduction in the bond strength of Optibond Solo ($p<0.001$). However, there was no significant reduction in the bond strength of G-Bond.

<table>
<thead>
<tr>
<th>Table 2</th>
<th>Microtensile bond strengths (Mean (SD), (MPa)) before and after loading</th>
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</thead>
<tbody>
<tr>
<td>Adhesive</td>
<td>Unloaded</td>
</tr>
<tr>
<td>G-Bond</td>
<td>23.75 (6.36)$^{ab}$</td>
</tr>
<tr>
<td>Optibond Solo Plus</td>
<td>37.50 (17.78)$^a$</td>
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</table>

*Groups with the same superscript letter are not significantly different ($p>0.05$)

<table>
<thead>
<tr>
<th>Table 3</th>
<th>Median (inter-quartile range) of percentage micro-leakage at axial walls by treatment and loading</th>
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</thead>
<tbody>
<tr>
<td>Adhesive</td>
<td>Unloaded</td>
</tr>
<tr>
<td>G-Bond</td>
<td>48.6 (36.8–55.8)$^a$</td>
</tr>
<tr>
<td>Optibond Solo Plus</td>
<td>36.9 (22.7–63.0)$^{ab}$</td>
</tr>
</tbody>
</table>

*Groups with the same superscript letter are not significantly different ($p>0.05$)

<table>
<thead>
<tr>
<th>Table 4</th>
<th>Median (inter-quartile range) of percentage micro-leakage at cavity floor by treatment and loading</th>
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<tbody>
<tr>
<td>Adhesive</td>
<td>Unloaded</td>
</tr>
<tr>
<td>G-Bond</td>
<td>69.1 (47.7–97.5)$^a$</td>
</tr>
<tr>
<td>Optibond Solo Plus</td>
<td>94.7 (58.3–100)$^{ab}$</td>
</tr>
</tbody>
</table>

*Groups with the same superscript letter are not significantly different ($p>0.05$)

<table>
<thead>
<tr>
<th>Table 5</th>
<th>Failure mode by treatment and loading</th>
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<tr>
<td>Adhesive</td>
<td>Loading</td>
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<tr>
<td></td>
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<tr>
<td>G-Bond</td>
<td>unloaded</td>
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<tr>
<td></td>
<td>loaded</td>
</tr>
<tr>
<td>Optibond Solo Plus</td>
<td>unloaded</td>
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<tr>
<td></td>
<td>loaded</td>
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</table>

* (Significantly different, $p=0.009$)
Fig. 2  Sequence of confocal scanning micrographs showing fluorescence evaluation of a loaded G-Bond specimen at the gingival floor. C=composite, D=dentin
a) reflection image
b) fluorescence image using 546/600 nm excitation/emission of an unloaded G-Bond specimen. TSM X20/0.80 oil immersion objective
c) reflection image
d) fluorescence image using 546/600 nm excitation/emission of a loaded G-Bond specimen. TSM X20/0.80 oil immersion objective

Fig. 3  Sequence of confocal scanning micrographs showing fluorescence evaluation of a loaded Optibond Solo Plus specimen at the gingival floor. C=composite, D=dentin
a) reflection image
b) fluorescence image using 546/600 nm excitation/emission of an unloaded Optibond Solo Plus specimen. TSM X20/0.80 oil immersion objective
c) reflection image
d) fluorescence image using 546/600 nm excitation/emission of a loaded Optibond Solo Plus specimen. TSM X20/0.80 oil immersion objective
Fig. 4  Scanning electron micrographs of the opposing ends of a fractured beam from the G-Bond group after loading. A mixed failure mode is evident.
   a) Dentin end of the beam; the rectangle indicates the area viewed at higher magnification in b, 70×.
   b) High magnification showing cohesive failure in resin and at top of hybrid layer, 1,000×.
   c) Resin end of the beam, the rectangle indicates the area viewed at higher magnification in d, 70×.
   d) High magnification showing cohesive failure in resin and at top of hybrid layer, 1,000×.

Fig. 5  Scanning electron micrographs of the opposing ends of a fractured beam from the Optibond Solo Plus group after loading. A mixed failure mode is evident.
   a) Resin end of the beam; the rectangle indicates the area viewed at higher magnification in b, 70×.
   b) High magnification showing fractured resin tags, 900×.
   c) Dentine end of the beam, the rectangle indicates the area viewed at higher magnification in d, 70×.
   d) High magnification showing fractured resin tags and open dentinal tubules, 1,000×.
With regards to the microleakage at the axial walls, there was no significant difference between the two adhesives in the unloaded state, however G-Bond exhibited a significant increase in microleakage after loading (p<0.05). There was no significant difference in the percentage microleakage values of the two materials after loading.

With regards to the microleakage at the gingival floor, there was no significant difference between the two adhesives in the unloaded state, however G-Bond exhibited a significant increase in microleakage after loading (p<0.05). There was no significant difference in the percentage microleakage values of the two materials after loading at the gingival floor (Figs. 2 and 3).

The modes of failure of the beams are shown in Table 5. The majority of the beams failed through a mixture of adhesive and cohesive failure at the bonded interface when unloaded and after loading. Representative SEM images the fractured beams of loaded G-Bond and Optibond Solo Plus specimens are shown in Figs. 4 and 5 respectively.

DISCUSSION

The present study evaluated a one step self-etching adhesive and a two-step etch-and-rinse adhesive in MOD cavities restored with direct resin composite recommended for use in posterior teeth, which were subjected to 250,000 cycles of 80 Newton loads. A force of 80 N was chosen as an average of the masticatory forces observed by Anderson and the loading condition of 250,000 cycles has been verified as one year of clinical wear. The load was applied to the mid-point of the occlusal portion of the restoration. It has been reported that the application of a compressive load in the middle of the restoration would create tensile stresses along the bonded interface at the mesial and distal aspects of the restoration. It was suggested that this would mimic the situation when occluso-proximal restorations are loaded directly by the opposing teeth during mastication.

The present study revealed that prior to loading the bond strengths of the cavities restored with the etch-and-rinse adhesive were higher than those restored with the self-etching adhesive although not significantly. However, after loading, the bond strength of the etch-and-rinse adhesive significantly reduced such that it was equivalent to the bond strength of the self-etch adhesive. On the other hand the bond strength of the self-etch adhesive did not significantly reduce after loading. Although, these findings might indicate that the resindentine bonds formed with the total-etch adhesive may not have been as resistant to mechanical stress as those formed with the self-etch adhesive, the result that bond strengths of both adhesives were similar after loading implies that both may offer equivalent performance under large composite restorations in posterior teeth.

The etch-and-rinse adhesive used in the present study has been evaluated previously. It was reported that when Optibond Solo Plus was subjected to mechanical loading in class II cavities, a significant reduction in bond strength was not found after loading for 100,000 and 200,000 cycles but only after loading for 500,000 cycles and thermal cycling. It was considered that this result may be because a low-elastic modulus hybrid layer was formed due to the glass filler particles, which allowed for a more homogeneous distribution of the mechanical stresses. In the present study, Optibond Solo Plus exhibited a significant reduction in bond strength after 250,000 cycles with no thermal cycling, which disagrees with previous research. This disagreement may be because it is not possible to exactly replicate the experimental conditions used in different academic institutions due to variations in specimen preparation and loading conditions and also the fact that human teeth as opposed to extracted teeth were used in the present study. Therefore further *in-vitro* studies are indicated.

With regards to the bond strength testing of the sectioned beams, it has been reported that the loading stress is concentrated mainly at the interface between the adhesive and the top of the hybrid layer but that bonded specimens generally failed beneath the hybrid layer. This was considered to occur because beneath the hybrid layer there may be exposed collagen networks in the demineralised intertubular dentine, which the resin had failed to surround or fill. SEM examination of the loaded Optibond Solo Plus specimens after bond strength testing revealed that all the specimens exhibited a mixed mode of failure at the adhesive interface and some specimens did show areas of failure at the base of the hybrid layer. In the case of the tested one-step self-etching adhesive, G-Bond, some cohesive failures in resin were observed together with mixed failures at the adhesive interface. Self-etching adhesives do not have a prior phosphoric acid etching step and are therefore etch intertubular dentine less aggressively. There is less likely to be a zone of demineralised dentine because the resin infiltrates to the depth of demineralisation, however, the resin-dentine interface is more hydrophilic due to the presence of residual solvent, which may account for the cohesive failures in resin.

In order to ensure that the cavities were prepared into dentine of a standardised depth and therefore similar morphology, only caries-free upper third molars that were of a similar size were selected for the study. For the microleakage evaluation, the axial walls of the proximal boxes of the cavities were considered separately from the gingival floor. Previous research has only investigated the influence of occlusal load cycling on self-etching and etch-and-rinse adhesives in class II cavities in bovine teeth. It was reported that 50,000 cycles did not effect the microleakage of the tested adhesives, which included Optibond Solo Plus. The results of the present experiment for Optibond Solo Plus agree with this finding. However, at the gingival floor, dye penetration was over 90% before loading and 100% after loading (Fig. 3). For the self-etching adhesive, G-Bond, there was a significant increase in dye penetration after loading at both the axial walls and cavity floor. Previous research has studied the
microporomeability of self-etching and etch-and-rinse adhesives\(^2\). Microporomeability was observed around the resin tags and along the entire resin-bonded interface between the hybrid layer and the adhesive resin using both fluorescence and reflection confocal microscopy of the G-Bond specimens\(^2\). Moreover, it was observed that silver nitrate accumulated around water droplets in the adhesive interface of G-Bond\(^2\). These findings may explain the high dye penetration percentages observed in the present study using fluorescence and reflection confocal microscopy.

Previous research has investigated the bonding durability of G-Bond to flat dentin surfaces surrounded by bonded enamel using the microtensile bond strength test in combination with the hour-glass trimming technique\(^3\). Although the bond strength of G-Bond gradually reduced over a period of 12 months water storage, there was no significant difference between the bond strengths at the 24 h, 3 month, 6 month and 12 month storage periods. The authors concluded that bonded enamel margins could not maintain the integrity of the resin-dentin interface\(^3\). The present study found that there was no significant reduction in the bond strength of G-Bond after mechanical loading and despite including enamel at the cavity margins, dye penetration was observed at the resin-dentin interface. These findings therefore agree with previous research and indicate that the resin-dentin bond strengths of G-Bond do not significantly reduce when stressed under different experimental conditions and bonded enamel margins cannot maintain the integrity of the resin-dentin interface.

Previous research has investigated the effect of the cavity configuration factor (c-factor) on the adhesion of three types of adhesives by looking at resin-dentin bond strengths on flattened dentin surfaces and in 4-mm deep occlusal cavities\(^4\). Resin-dentin bond strengths were lower in the occlusal cavities and a higher number of pre-test failures were observed along with larger standard deviations even when composite was placed in increments compared to those of flat cavities\(^4\). In the present study, pre-test failures and large standard deviations were observed in both the unloaded and loaded groups. Although, the statistical analysis revealed that Optibond Solo exhibited a significant reduction in bond strength after loading, when unloaded this group exhibited the largest standard deviation. It is therefore speculated that although an incremental technique was used to restore the MOD cavities, their c-factor may have influenced resin-dentin bond strengths prior to loading as well as after loading.

Moreover, previous research has investigated the effect of cavity size and filling using either an incremental or bulk filling technique with or without the placement of a flowable composite for large cavities cut in bovine teeth\(^5\). Although, the incremental filling technique resulted in the highest bond strengths in large (5-mm diameter cavities), the authors recommended the use of an incremental technique in combination with placement of a flowable liner on the cavity floor because of the unpredictable influence of a high C-facto\(^5\). Further research is therefore indicated on the performance of different types of adhesive-composite systems with or without a flowable liner in complex situations such as large posterior cavities under conditions resembling those experienced in the oral cavity.

The dye penetration results of the present study if considered in isolation without the bond strength data indicate that the self-etching adhesive might perform less well than the etch-and-rinse adhesive in large posterior restorations. However, the bond strength results contradict this assumption and therefore it must be considered that there was no relation between microleakage and bond strength. This finding agrees with the research by Frankenberger who looked at published literature on the marginal integrity in vitro and in vivo between 1990 and 2005 and found that after two years of clinical service, restorations bonded with self-etching adhesives did not clinically fail but exhibited significantly more marginal gaps\(^7\). They concluded that clinical outcome is not predictable from marginal integrity alone. Furthermore, previous research has found that restorations bonded with self-etching adhesives may exhibit greater microleakage at the enamel margins but do not clinically fail\(^9\).

In conclusion, after MOD direct resin composite restorations were loaded, the tested one step self-etching and two step etch-and-rinse adhesives exhibited equivalent bond strengths to dentine. However, the microleakage data for the two adhesives before and after loading contrasted the bond strength results and therefore within the limitations of the present study, it was also concluded that microleakage and bond strength are not related.

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