

Original Research Article

The influence of immediate dentin sealing techniques on marginal micro leakage of composite resin inlays

ABSTRACT

Aim: To evaluate the influence of two immediate dentin sealing (IDS) techniques on the marginal microleakage of composite inlays.

Methodology: Thirty human third molars received MOD preparations, with the mesial proximal box extending above and the distal box extending below the cement-enamel junction. The teeth were randomly divided into groups (n=10): G1 – control, without IDS; G2 – IDS with Clearfil SE Bond; G3 - IDS with Clearfil SE Bond and Protect Liner F. Impressions of the preparations were made, followed by provisory restorations. Composite inlays were luted with ED Primer A and B and Panavia F to the preparations. The samples were thermocycled 2,500x between 5°C and 55°C, then placed in a 0.5% methylene blue dye solution for 24 hours. The samples were sectioned in the mesio-distal direction and evaluated for microleakage at the mesial and distal margins under a stereomicroscope. The dye penetration was scored as 0 (no penetration), 1 (penetration to half of the gingival wall), 2 (penetration along the gingival wall), 3 (penetration along the gingival and axial wall).

Results: According to Kruskal-Wallis analysis ($\alpha=.05$), there were no significant differences in the dye penetration between the groups in either the enamel ($P=.07$) or the dentin ($P=.31$). According to Wilcoxon, the dye penetration in the dentin was higher than in the enamel ($P=.03$) for group 3.

Conclusion: The IDS with Clearfil SE Bond, either associated or not associated with Protect Liner F, was not capable of producing complete sealing of the enamel and dentin margins.

Keywords: enamel; dentin; immediate dentin sealing; microleakage.

1. INTRODUCTION

Composite resins have been largely used in Dentistry for aesthetic reasons. Polymerization shrinkage is an intrinsic characteristic of these materials [1,2]. Shrinkage can lead to marginal microleakage [3,4] when the forces generated during the polymerization cause the rupture of

30 the adhesive-tooth interface [5]. When an indirect technique is used to restore a tooth with
31 composite resin, the polymerization shrinkage occurs outside the tooth, and the possibility of
32 microleakage is reduced [6,7].

33 The traditional technique used for producing indirect restorations consists of making an
34 impression of the tooth immediately after preparation and luting an acrylic resin restoration with
35 provisional cements, or with provisional resin materials applied directly to the prepared tooth.
36 Once the permanent restoration is ready, the provisional material is removed and an adhesive
37 system is applied to the tooth, followed by a resin cement for the adhesive luting procedure.

38 Studies have shown that adhesive systems bond better to freshly cut dentin compared
39 with dentin contaminated with temporary materials [8,9] and that this contamination may cause
40 microleakage [10]. To avoid this problem, the immediate dentin sealing (IDS) technique was
41 suggested in the early 1990s [11]. This technique consists of the application of an adhesive
42 system immediately after finishing the tooth preparation but before the impression. Another
43 technique consists of the application of an adhesive system and a low-viscosity composite resin
44 to the dentin immediately after finishing the preparation [12,13]. It is believed that a layer of low-
45 viscosity composite resin helps to protect the hybrid layer and, consequently, preserve the
46 dentin seal [14].

47 The clinical advantages of the IDS technique are related to the seal and the protection
48 of the dentin-pulp complex immediately after cavity preparation by the application of a resin
49 agent, preventing and decreasing the sensitivity and bacterial infiltration during the provisional
50 stage [15,16].

51 Studies have shown that there is an increase in the bond strength when the IDS
52 technique is applied [17-23]. However, there is no consensus about the correlation between the
53 bond strength and the level of microleakage [24,25]. Therefore, it is important to evaluate how
54 the IDS techniques relate to the marginal sealing ability.

55 The aim of this study was to evaluate the influence of two IDS techniques on the
56 microleakage of composite resin inlays. This study was conducted under the null hypothesis
57 that IDS techniques do not influence the microleakage of composite resin inlays.

2. MATERIALS AND METHODS

Thirty sound third molars were obtained from the Tooth Bank of the Pontifical Catholic University of Rio Grande do Sul after Ethics Committee approval was obtained. The teeth were cleaned and disinfected in 0.5% chloramine for 24 h, then stored in distilled water at 4°C. The buccal-palatal and mesio-distal dimensions of each tooth were measured with a digital caliper rule (Mitutoyo, Suzano, SP, Brazil). A variation of 0.5 mm was allowed for each measurement to standardize the dimensions of the teeth.

The teeth were randomly divided into three groups ($n=10$): group 1, conventional technique (control); group 2, IDS with the adhesive system; group 3, IDS with the adhesive system and a low-viscosity composite resin. The materials used are listed in Table 1.

Table 1. Materials used in the study.

Materials	Composition	Manufacturer
Clearfil SE Bond	<i>Self-etch primer</i> : 10-MDP, HEMA, hydrophilic dimethacrylate, photo-initiator, water. <i>Adhesive</i> : 10-MDP, bis-GMA, HEMA, hydrophilic dimethacrylate, microfiller	Kuraray Medical Inc., Tokyo, Japan
Protect Liner F	TEG-DMA, Bis-GMA, methacryloyl fluoride-methyl, methacrylate copolymer	Kuraray Medical Inc., Tokyo, Japan
Panavia F	<i>ED primer A</i> : HEMA, 10-MDP, 5-NMSA, water, accelerator <i>ED primer B</i> : accelerator, water, sodium benzene sulfinate <i>A-Paste</i> : Methacrylate, 10-MDP, quartz-glass, microfiller, photoinitiator <i>B-Paste</i> : Methacrylate, barium glass, sodium fluoride, chemical initiator	Kuraray Medical Inc., Tokyo, Japan
Filtek Z250	BisEMA, UDMA, BisGMA, TEGDMA, silane treated ceramic	3M ESPE, St. Paul, MN, EUA

HEMA=hydroxyethylmethacrylate; TEGDMA= triethylene glycol dimethacrylate; UDMA= urethane dimethacrylate; Bis-GMA= bisphenol-glycidyl methacrylate; 10-MDP = 10-methacryloyloxydecyl dihydrogen phosphate; Bis-EMA: ethoxylated bisphenol A dimethacrylate; 5-NMSA: *N*-methacryloyl-5-aminosalicylic acid.

2.1 MOD preparation

Each tooth was mounted vertically in a plastic ring with a self-cured acrylic resin (Jet Classico, São Paulo, SP, Brazil) up to 2 mm below the cement-enamel junction (CEJ). A single operator performed cavity preparations on the mesial, distal and occlusal surfaces with a 4159

77 diamond bur (KG Sorensen, Barueri, SP, Brazil) at a high setting speed under constant water
78 and air cooling. The width between the buccal and lingual cavosurface angle was two-thirds the
79 distance between the buccal and lingual cusp tips, and the occlusal isthmus was 3 mm deep.
80 The widths of the proximal boxes corresponded to one-third the distance between the buccal
81 and lingual surfaces of the teeth at the level of the gingival wall, and 1.5 mm deep. The mesial
82 boxes were located 1 mm coronal to the CEJ, and the distal boxes were located 0.5 mm below
83 the CEJ. The internal line angles were rounded, the cavosurface angles were approximately
84 90°, and the angle of divergence of the walls of the preparations was approximately 6°. The
85 dimensions of the cavity were standardized using a digital paquimeter with a precision of 0.01
86 mm. A diamond bur was used for five preparations.

87 **2.2 IDS techniques**

88 Group 1 (the control) received only the cavity preparation. In group 2, the Clearfil SE
89 Bond adhesive system was applied immediately after the preparation as follows: the self-
90 etching primer was applied to the dentin using a brush tip and was left in place for 30 s. Excess
91 solvent was removed by air drying for 5 s. The bond was applied to the surface cavity with a
92 brush tip, and gentle air drying was applied for 3 s, followed by light-curing for 20 s with a light-
93 curing unit (Optilux Plus, Gnatus, Ribeirão Preto, SP, Brazil). The light intensity was controlled
94 by a radiometer (Model 100 Demetron, Kerr, Danbury, CT) between 450 and 500 mW/cm².
95 Polymerization of the adhesive was followed by the application of an air-blocking barrier
96 (glycerin jelly) and 10 s of additional light curing to polymerize the oxygen inhibition layer
97 (MAGNE et al., 2005). In group 3, Clearfil SE Bond was applied as described in group 2 without
98 the air-blocking barrier. After application of the adhesive, Protect Liner F was placed on the
99 adhesive surface using a brush-on technique and light-cured for 20 s. The surface of the cured
100 flowable composite resin was wiped with a cotton pellet soaked in alcohol for 10 s to remove the
101 unpolymerized layer on the surface (UDO et al., 2007).

102 **2.3 Restorative procedures**

103 Impressions of the preparations were taken with polyvinyl siloxane (3M Espe, Saint
104 Paul, MN, USA) with individual trays made from self-cured acrylic resin using the putty/wash
105 one-step technique. The impression material was allowed to set for 10 min before it was

106 removed from the preparation. Temporary self-cured acrylic resin crowns were then luted onto
107 the preparations with non-eugenol cement (Temp Bond NE, Kerr, West Collins, CA, USA).
108 Tooth specimens were stored in water at 37 °C water for 7 days. The impressions were poured
109 after 1 h using Durone Type IV stone (Dentsply, York, PA, USA).

110 The casts were lined with a die spacer, except in the marginal areas. Four horizontal
111 layers of composite resin (Filtek Z250, 3M Espe, Saint Paul, MN, USA) were inserted into the
112 casts with a Thompson spatula (nos. 2 and 12), producing a 90° inclination between the internal
113 slopes and the cusps. Each resin layer was light-cured for 40 s, followed by finishing with
114 polishing discs and silicone tips (Soft-Lex, 3M Espe, Saint Paul, MN, USA).

115 **2.4 Luting procedures**

116 Following storage, the provisional restorations were removed, and the remaining
117 temporary cement on the inlay preparation was scraped off with a dental instrument.
118 Subsequently, the dentin (group 1) and sealed dentin (groups 2 and 3) were cleaned with a
119 mixture of water and pumice using a rotary brush for 10 seconds. The fitting surface of the
120 restoration was cleaned with alcohol and sandblasted with 50 µm aluminum oxide for 5 s, rinsed
121 and dried. A layer of silane (Ceramic Primer, 3M ESPE, St. Paul, MN, USA) was applied,
122 followed by gentle air-drying for 5 s. The coated surfaces of the preparation (except group 1)
123 were then acid-etched with 37% phosphoric acid for 10 s and rinsed and dried to remove debris.
124 A mixture of ED Primer A and B was applied for 30 s and gently air-dried for 5 s. The base and
125 catalyst of the Panavia F resin cement were mixed according to the manufacturer's instructions.
126 The crowns were seated using a 1 kg standard load for 2 min. Excess cement was removed
127 with a microbrush and each surface (buccal, lingual, mesial, distal and occlusal) was light-cured
128 for 40 s. The margins were finished with polishing discs (Sof-Lex). The specimens were stored
129 in distilled water at 37 °C for 24 h.

130

131 **2.5 Microleakage evaluation**

132 After storage, the specimens were thermocycled in water for 2.500 cycles between 5 and
133 55°C, with a dwell time of 30 s. After the thermocycling, the teeth were covered with nail varnish,
134 except for the restoration and a 0.5 mm margin around it. The crowns were immersed in a 0.5%

135 methylene blue dye solution for 24 hours at 37 °C. After the thermocycling and dye penetration steps,
136 the teeth were rinsed thoroughly with water and sectioned in the mesio-distal direction, in the center
137 of the crowns, with a diamond disc mounted on a low speed laboratory cutting machine (Labcut
138 1010, Extec Corp., London, UK) with cooling. The cut surfaces were polished with 1000-, and
139 1200-grit silicone carbide abrasive papers and wetted with water using manual pressure and
140 rotary movements. The extent of the dye penetration was observed in the mesial gingival wall
141 (enamel) and distal gingival wall (dentin) under a stereomicroscope at 20x (Olympus Corp.,
142 Tokyo, Japan). Dye penetration was scored: 0= no penetration; 1= dye penetration to half of the
143 gingival wall; 2= dye penetration along the gingival wall; 3= dye penetration along the gingival and
144 axial wall.

145 **2.6 Statistical analysis**

146 The data were analyzed using the Kruskal-Wallis non-parametric test to compare the
147 microleakage between the groups. The Wilcoxon non-parametric test was used to compare the
148 microleakage between the enamel and dentin. The significance level was 5%.

149

150 **3. RESULTS**

151 According to Kruskal-Wallis non-parametric test, there were no significant differences in the
152 microleakage scores between the groups, for either the enamel ($P = .07$) or the dentin (P
153 $= .31$). Figure 1 shows the microleakage score distribution in the groups.

154 According to the Wilcoxon non-parametric test, there were significant differences in the
155 microleakage scores between the enamel and dentin for group 3, with the microleakage scores
156 in the dentin statistically higher than those in enamel ($P = .03$). There were no significant
157 differences in the microleakage scores between the enamel and dentin for group 1 ($P = .38$) and
158 group 2 ($P = 1.00$).

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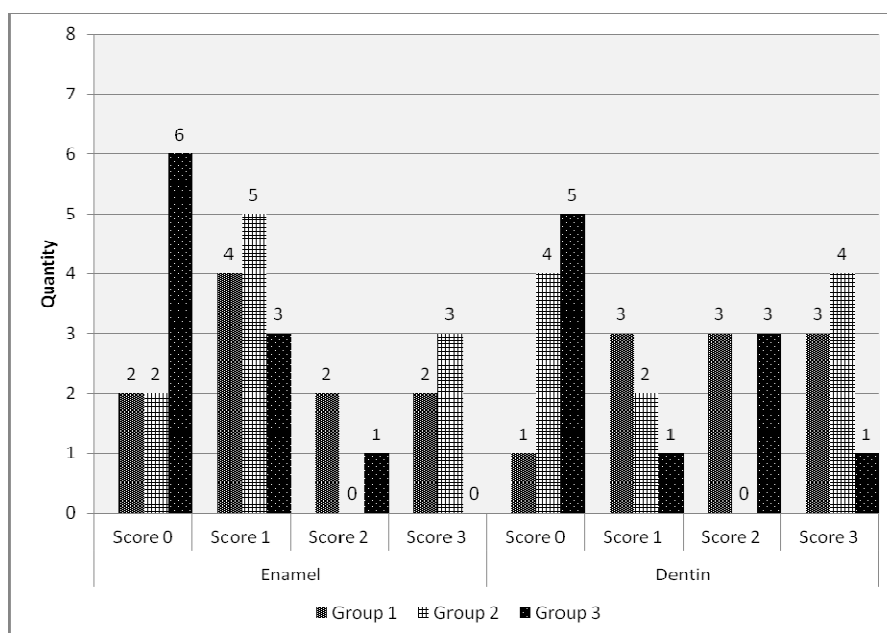


Fig. 1. Microleakage scores distribution in the groups.

4. DISCUSSION

In the literature, microleakage has been defined as the passage of bacteria, fluid, molecules or ions that are not “detectable clinically” between the cavity wall and the restorative material [26]. Microleakage can occur due to the deterioration of the tooth-restoration interface, differences in the thermal expansion coefficients of the tooth and restorative material, or polymerization shrinkage [27]. The clinical consequence is that the higher the marginal microleakage, the greater the chances that fluids, ions and bacteria percolate through the tooth-restoration interface [28]. Microleakage causes dissolution of luting materials [29], which can result in bacterial contamination and post-operative sensitivity, and can even compromise the pulp vitality [28]. Therefore, it is important to use materials and techniques that prevent or reduce microleakage.

According to the results obtained in the present study, the null hypothesis was accepted because there were no significant differences in the microleakage between the three different procedures related to adhesive luting, either in the enamel or dentin. Figure 1 shows that any technique could avoid marginal microleakage, which was consistent with the results of prior studies [23,30].

178 In the present study, the Clearfil SE Bond adhesive system was applied to the dentin
179 immediately after the tooth preparation in groups 2 and 3. In group 1 (conventional technique),
180 ED *Primer* was applied to the dentin after the provisional restoration removal, followed the
181 Panavia F resin cement application.

182 ED Primer is a one-step self-etching primer that has a moderate capacity of dentin
183 demineralization. Due to the presence of the hydrophilic monomer HEMA, ED Primer presents
184 some permeability, allowing changes at the dentin–adhesive interface, and consequently,
185 hydrolytic degradation of this interface [31]. It was suggested that the application of hydrophobic
186 adhesive over the ED Primer layer before luting with Panavia F could decrease the amount of
187 microleakage [31]. Clearfil SE Bond is a two-step adhesive system with a pH close to 2 with that
188 has a self-etching primer and an adhesive, and a moderate capacity to demineralize dentin [32].
189 As the primer in this adhesive system also has the hydrophilic monomer HEMA, it has some
190 permeability. However, the application of the adhesive on the primer, which contains a larger
191 quantity of hydrophobic monomers, tends to reduce the permeability of this adhesive system
192 [33]. Therefore, the expected result of the present study was to obtain less microleakage in
193 group 2 relative to the control group, which was not confirmed. The differing material
194 composition may partially explain this finding.

195 The ED *Primer* and Clearfil SE Bond adhesive system contain the acidic monomer 10-
196 MDP, which chemically bonds to the calcium of the hydroxyapatite that remains partially
197 attached to collagen [34], promoting a hydrolytically more stable dentinal bond. This chemical
198 bond may favor a similar protection effect on the hydrolytic degradation for both adhesive
199 systems, resulting in microleakage scores without significant differences between them.

200 Despite the absence of significant differences between the groups due to the
201 homogeneous microleakage scores distribution, there was a trend towards more scores of 0
202 and fewer scores of 3 when the Clearfil SE Bond adhesive system and the Protect Liner F low-
203 viscosity composite resin were applied to the dentin immediately after cavity preparation (group
204 3). It is possible that the combination of various factors may have contributed to this finding. For
205 example, perhaps additional polymerization on the Clearfil SE Bond at the moment of low-
206 viscosity composite resin photo activation occurred, favoring a higher rate of monomer-to-
207 polymer conversion in the adhesive most closely in contact with the tooth substrate [31]. This

208 higher rate of conversion may reduce the permeability of the adhesive, resulting in a greater
209 durability of the adhesive interface [35]. In addition, the higher rate of conversion may favor a
210 higher adhesive bond strength to the dental substrate, as this an important factor in resisting the
211 polymerization shrinkage that is restricted to the thin layer of Panavia F resin cement [36].
212 Studies show that polymerization shrinkage stress that is generated due to the lack of no-
213 adhered surfaces can break the bond between the resinous material and the cavity walls,
214 resulting in gaps and failures in the interfaces [37,38]. Therefore, the bond of the adhesive to
215 the substrate is important in resisting the shrinkage stresses that are generated during
216 polymerization. In addition to these factors, the adhesive was applied directly over the cut dentin
217 and was not contaminated with the temporary material, thus favoring a better bond [8,9]. All of
218 these factors may have influenced the microleakage values in group 2 because the Clearfil SE
219 Bond also received further polymerization at the time the hydrophilic gel was applied. However,
220 the difference between groups 2 and 3 is the application of low-viscosity composite resin, which
221 may have favored a greater absorption of the stresses generated by the polymerization
222 shrinkage of the resin cement, contributing to an increased relief of polymerization shrinkage in
223 the adhesive interface [37,39].

224 In both IDS techniques, the bond of the cementing agent to the pre-existing resin layer
225 should be promoted by cleaning the surface prior to cementation [40] to remove remnants of the
226 provisional cements that can cause a significant reduction in the bond strength of the luting
227 agent [41,42]. Therefore, after removing the provisional restoration, the preparations in all of the
228 groups received prophylaxis with pumice and water.

229 Although the techniques of IDS have the primary purpose of sealing the dentin, this
230 study also evaluated the microleakage in the enamel. This is because, depending on the clinical
231 case, the preparation margin may be in enamel in the cervical proximal region. Even when the
232 clinician seeks to apply the adhesive system only to the dentin, the enamel is usually also
233 treated.

234 Comparing the microleakage between the enamel and dentin, there was a significant
235 difference between these substrates only in group 3, in which the scores in the dentin were
236 significantly higher than those in the enamel. Despite the lack of significant differences between
237 the enamel and dentin for groups 1 and 2, there was a trend towards scores of 2 and 3 when

238 assessed in dentin. This finding is in agreement with the literature, which shows that
239 microleakage tends to be higher in dentin than in enamel [43,44]. The dentin has a higher
240 biological variability than enamel, making it a more difficult substrate to obtain a stable bond
241 [45]. Possibly, the bond to dentin did not withstand the interfacial stresses generated by the
242 polymerization shrinkage of the resin cement. Another possible explanation for not observing
243 lower microleakage scores in the enamel in relation to the dentin is the fact that ED *Primer* and
244 Clearfil SE Bond have questionable bonding to enamel as they have a moderate level of
245 demineralization capacity. In the case of Clearfil SE Bond, laboratory studies have reported an
246 equal or lesser effectiveness of the bond to enamel compared to adhesive systems that employ
247 phosphoric acid etching [46,47].

248 This study used thermal cycling as the aging feature of the samples to simulate the
249 degradation of bonded interfaces that occurs over time in the oral cavity. The effectiveness of
250 thermal cycling on microleakage as a model to simulate clinical aging has been a subject of
251 controversy among researchers [48,49]. Although there has been discussion on the validity of
252 thermal cycling as a valid model system, it has been frequently utilized in microleakage studies.

253 The transfer of results from laboratory studies to the clinical setting must be performed
254 with caution, as in vitro studies can never fully reproduce the conditions and behavior of the oral
255 cavity behavior. According to the results, it seems that IDS with Clearfil SE Bond, either alone or
256 in combination with Protect Liner F, does not prevent microleakage. However, longitudinal
257 clinical studies are needed to confirm these results in indirect MOD restorations.

258 **5. CONCLUSION**

259 This study showed that an IDS technique with the Clearfil SE Bond adhesive system, either
260 associated or not associated with the Protect Liner F low-viscosity composite resin, does not
261 produce complete sealing of the margins in enamel or dentin.

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