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In Vitro Evaluation of Giomers Microleakage After Exposure to 33% Hydrogen Peroxide: Self-etch vs Total-etch Adhesives

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Clinical Relevance

Microleakage of a total-etch adhesive system was not influenced by the application of hydrogen peroxide; conversely, enamel dye penetration significantly increased for a self-etching primer.

SUMMARY

This study evaluated the microleakage of a giomer resin bonded with total-etch and self-etch adhesive systems after exposure to hydrogen peroxide. Thirty freshly extracted, caries-free human premolars and molars were used. The teeth were randomly divided into two groups: Group I was exposed to 33% hydrogen peroxide (Niveous-Shofu) for 30 minutes daily for five consecutive days; Group II received no treatment (control). A week later, Class V standardized preparations were performed on the facial and lingual surfaces, with the gingival margin placed 1 mm below the CEJ. Each group was then divided into two subgroups: in Groups IA and IIA, a self-etching adhesive system (FL Bond–Shofu) was applied, and in Groups IB and IIB, a total-etch adhesive system (Prime & Bond NT–Dentsply/Caulk) was applied according to manufacturers' instructions. The teeth were restored using 2-mm increments of Beautifil A2 resin-based giomer material (Shofu). Each layer was cured using a Spectrum 800 curing light (Dentsply/Caulk) for 20 seconds at 600mW/cm². The teeth were thermocycled 500x between 5°C and 55°C with a dwell of 30 seconds; they were then placed in a 0.5% methylene blue dye solution for 24 hours at 37°C. Samples were sectioned longitudinally and evaluated for microleakage at the occlusal and gingival margin under a stereomicroscope at 20x magnification. Dye penetration was scored using the following scoring system 0 = no penetration; 1 = partial dye penetration along the occlusal or gingival wall; 2 = partial dye penetration along the occlusal or gingival wall but did not include the axial wall; 3 = dye penetration to and along the axial wall. A Mann-Whitney test revealed a statistically significant

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difference between subgroups at the occlusal level ($p<0.0001$). Group IA yielded the most microleakage. No statistically significant difference was reported at the gingival level.

Microleakage was affected by hydrogen peroxide exposure only at the enamel cavosurface margin when a self-etching primer adhesive system was used.

INTRODUCTION

Resin composite and adhesive materials have been rigorously investigated and further developed since their introduction, with an attempt to find an adequate alternative to dental amalgam. This research has resulted in the development of resin composites with improved physical, aesthetic and handling properties (Jordan & Suzuki, 1991; Christensen, 1998; Hickel, Manhart & García-Godoy, 2000). Recently, a new fluoride-releasing light-cured restorative material containing pre-reacted glass ionomer fillers has been introduced on the market with promising results (Matis & others, 2004). This new material is called "Giomer" and is based on the incorporation of surface pre-reacted glass-ionomer fillers (S-PRG) into a resin matrix. According to claims made by the manufacturer, the result is a restorative material that combines the advantages of composite and glass ionomer.

It was found that giomers did not show the gel transition phase characteristic of glass-ionomer cements (GICs); however, the glass-ionomer phase is readily available in PRG particles (Tay & others, 2001). The glass-ionomer phase is responsible for the fluoride release mechanism of GICs. Although the long-term fluoride release of giomers is questionable (Yap & others, 2002), a recent research study reported that giomers have demineralization inhibition properties similar to glass-ionomers *in vitro* (Gonzalez, Yap & Hsu, 2004). Like compomers, giomers are light polymerized and require the use of a bonding system for adhesion to tooth structure.

Self-etching primers are gaining popularity and represent an alternative approach to total-etch adhesives. They are based on the use of non-rinse acidic monomers that simultaneously condition and prime dentin and enamel. Elimination of the post-conditioning step and the need to apply primer in a specific condition of wetness render use of these adhesives less technique sensitive (Prati & others, 1998; Hannig, Reinhardt & Bott, 1999, 2001; Tay & Pashley, 2001; Pashley & Tay, 2001). However, the bonding effectiveness of self-etching primers to enamel is still questionable (Hannig & others, 1999; Pashley & Tay, 2001); the use of total-etch adhesive systems for enamel bonding is considered a milestone in adhesive dentistry. Conversely, the bonding effectiveness to dentin of both self-etching primers and total-etch adhesive systems is comparable (Hannig & others, 1999; Tay & Pashley, 2001).

In the last decade, tooth whitening has been one of the fastest growing areas of cosmetic dentistry and is very often followed by placement of esthetic restorations. However, it should be noted that increased microleakage and reduced bond strength has been reported when bonding composite to bleached teeth (Crim, 1992; Stokes & others, 1992; García-Godoy & others, 1993; Barkhordar, Kempler & Plesh, 1997; Swift & Perdigão, 1998). The mechanism of this adverse effect is still unknown; it may be related to inhibition of the adhesive polymerization process due to residual oxygen and peroxides or as a consequence of enamel and dentin ultra-morphology alteration (Titley & others, 1993; Perdigão & others, 1998; Shinohara, Rodrigues & Pimenta, 2001).

Nikaido and others (1999) reported less sensitivity to chemical irrigants for self-etching primers vs total-etch adhesives when bonding to endo-treated teeth. The authors of this study are not aware of other studies that have compared the ability of self-etching primers and total-etch adhesives to providing adequate sealing after bleaching treatments.

The performance of total-etch and self-etch adhesives has been constantly improved in the last decade. However, the influence of chemical agents on the sealing ability of adhesive systems has not yet been fully explored; the enamel and cementum/dentin interface may be affected by the application of hydrogen peroxide.

This study evaluated the ability of total-etch and self-etch adhesives in reducing microleakage in Class V Giomer restorations after exposure to 33% hydrogen peroxide.

METHODS AND MATERIALS

Thirty freshly extracted, caries-free human premolars and molars were kept in distilled water at 4°C for 24 hours. The teeth were randomly divided into two groups. In Group I, the teeth were exposed to 33% hydrogen peroxide (Niveous, Shofu Dental Corporation, San Marcos, CA, USA) for 30 minutes a day for five consecutive days; in Group II, the teeth were not exposed to any bleaching treatment (control). Upon completing the bleaching treatment, all teeth were kept in distilled water for one week before starting the restorative procedures. The preparations were standardized as 4-mm long, 3-mm wide and 1.5-mm in

Table 1: *Distribution of Specimens in the Two Groups; The Giomer Resin Beautifil Was Used in All Groups*

Group I-Bleach (Niveous)	Group II-No Bleach (Control)
A) FL-Bond (self-etch)	**A)** FL-Bond (self-etch)
B) Prime & Bond NT (total-etch)	**B)** Prime & Bond NT (total-etch)

depth. Class V restorations were prepared with the gingival margin 1 mm below the CEJ using a #4 round bur (Shofu Dental Corporation) with a high speed handpiece and copious amounts of water. No bevel was prepared for the coronal enamel margins. Class V preparations were performed in both the facial and lingual surface of each tooth in order to double the number of restorations, keeping the same number of teeth. The facial preparation on each tooth was assigned to the adhesive system FL-Bond subgroups; conversely, the lingual surface was assigned to the adhesive system Prime & Bond NT subgroups. Each group was then divided into two subgroups corresponding to each adhesive system (Table 1).

In Groups IA and IIA, a two-step self-etching fluoride releasing adhesive system (FL Bond, Shofu Dental Corporation) was applied on dried dentin and enamel surfaces according to the manufacturer's instructions.

In Groups IB and IIB, each prepared tooth was etched with 34% H_3PO_4 (Tooth Conditioner Gel, Dentsply/Caulk-Mildford, DE, USA) for 15 seconds, rinsed for 20 seconds, then gently blown to remove excess water, being careful to maintain a moist surface; a total-etch nanofilled acetone-based adhesive system (Prime & Bond NT, Dentsply/Caulk) was applied according to manufacturer's instructions. Both adhesive systems were cured for 20 seconds at 600mW/cm^2 using a Quartz Tungsten Halogen light (Spectrum 800, Dentsply/Caulk).

The restorations were completely filled using wedge-shaped A2 Giomer increments (Beautifil, Shofu Dental Corporation), each layer not being more than 2 mm, and cured for 20 seconds at 600mW/cm^2 using a Quartz Tungsten Halogen light (Spectrum 800, Dentsply/Caulk). The restorations were finished under water-cooling with fine and superfine diamond points (Hybrid points Shofu Dental Corporation) and polished with SuperBuff disks (Shofu Dental Corporation). All teeth were stored in distilled water at 37°C for 24 hours.

The restored teeth were thermocycled 500x in a 5°C-55°C water bath with a dwell time of 30 seconds in each bath. The specimens were then blotted dry with a paper towel and the root sealed with sticky wax. Pink and yellow nail polish were applied, respectively, on the facial and lingual occlusal surfaces and all around the teeth except for 1 mm from the cavosurface margins. The teeth were embedded in acrylic resin blocks (Orthodontic Resin, Dentsply/Caulk). All specimens were then immersed in 0.5 methylene blue dye solution for 24 hours. The dye solution was buffered to pH 7 in order to avoid demineralization of the tooth structure, thus giving false readings. The teeth were rinsed in running water, blotted dry, then sectioned longitudinally from facial to lingual surface with a water-cooled diamond wheel saw (Isomet, Buehler, Lake Bluff, IL, USA). Dye penetration at the occlusal and gingival margin was examined using a stereomicroscope at 20x by two independent evaluators pre-calibrated at 85% reliability. If there were any disagreements in score reported by the two evaluators, the higher score was taken. The specimens were scored according to the following criteria: 0 = no dye penetration; 1 = partial dye penetration (up to 1/3 of the occlusal or gingival wall length) along the occlusal or gingival wall; 2 = dye penetration along the occlusal or gingival wall (superior to 1/3 of the occlusal or gingival wall length), but not including the axial wall; 3 = dye penetration to and along the axial wall (Toledano & others, 1999, 2000). Statistical analysis was performed using Kruskal Wallis one-way at $p<0.05$ and Mann-Whitney U-Test.

Table 2: *Enamel Microleakage Scores for Groups I and II*

Subgroup	Scores				Specimens
	0	1	2	3	
IA (self-etch)	2	11	2	0	15
IB (total-etch)	15	0	0	0	15
IIA (self-etch)	14	1	0	0	15
IIB (total-etch)	15	0	0	0	15
Total	46	12	2	0	60

Table 3: *Cementum Microleakage Scores for Groups I and II*

Subgroup	Scores				Specimens
	0	1	2	3	
IA (self-etch)	13	1	1	0	15
IB (total-etch)	11	0	2	2	15
IIA (self-etch)	15	0	0	0	15
IIB (total-etch)	13	2	0	0	15
Total	52	3	3	2	60

RESULTS

Enamel/Occlusal Scores

The dye penetration scores for Groups I and II for the occlusal wall are presented in Table 2. Kruskal-Wallis test was performed to compare significant differences between Groups I and II. Results displayed a statistically significant difference between Groups I and II ($\chi^2_{df=3} = 44.25$, $p<0.0001$).

Pairwise comparison was performed using the Mann-Whitney U-Test. The results yielded a statistically significant difference between Subgroup IA vs Subgroups IIA, IIB and IB ($p<0.0001$), with Subgroup IA having the higher microleakage score. Conversely, no significant difference was reported between Subgroups IIA, IIB and IB ($p>0.05$).

Cementum/Gingival Scores

Dye penetration scores for the gingival wall for Groups I and II are presented in Table 3. The Kruskal-Wallis test reported no significant difference between Groups I and II ($\chi^2_{df=3} = 5.02$, $p>0.05$). Gingival scores between subgroups were compared through a Mann-Whitney U-Test, and the results yielded no statistically significant difference in microleakage score ($p>0.05$).

DISCUSSION

Tooth whitening may be combined with esthetic restorative treatments. The adverse potential effects of tooth whitening on bonding to both enamel and dentin have been reported, but data are missing on the interaction of total-etch and self-etch adhesive systems with hydrogen-peroxide treated tooth structure.

Depending on their pH, self-etch primers can be subdivided into weak, mild, strong and intermediary strong self-etch systems (Van Meerbeek & others, 2001, 2003). "Strong" and "intermediary strong" self-etch primers were introduced more recently and have ultramorphological characteristics very similar to those produced by total-etch adhesives on both enamel and dentin. The majority of these products belong to the "one-step self-etch" category of adhesives. Research has reported that these products are not yet as reliable as two- and three-step total-etch and self-etch adhesives (De Munck & others, 2003). The FL-Bond adhesive system belongs to the category of "mild" self-etch primers. With an ineffective etching of enamel using "weak" self-etch adhesives, the bonding effectiveness of "mild" self-etch primers to enamel is questionable. "Mild" self-etch primers do not bond well to uncut enamel (Perdigão & Geraldeli, 2003); and it was reported that a self-etch procedure with "mild" self-etch primers may result in equal or reduced bonding effectiveness to enamel when compared to a total-etch approach (Kanemura, Sano & Tagami, 1999; Hannig & others, 2001; Pashley & Tay, 2001).

Bonding to intact enamel is very challenging since it consists of approximately 70% prismless enamel. This phenomena may be the consequence of a reduced micromechanical interlocking due to insufficient treatment with phosphoric acid. However, some "mild" self-etch primers reported bond strength values similar to that of total-etch adhesive systems when applied to instrumented enamel (Van Meerbeek & others, 2001, 2003). Although the etching pattern of "mild" self-etch primers is not as deep as that with phosphoric acid etching, a nano-retentive interlocking is created between ground enamel and resin, which may explain the increased bonding effectiveness. It was suggested that the bonding effectiveness of self-etch primers may result from a combined micromechanical and chemical interaction with tooth substrate; the chemical component may be able to compensate for the reduced bonding effectiveness from decreased micromechanical interlocking (Van Meerbeek & others, 2001, 2003). The FL-Bond system contains 4-AET (4-Acryloxyethyltrimellitic acid), which can interact with the Calcium cations of hydroxyapatite to form 4-AETCa, a relatively insoluble calcium salt that improves durability of the adhesive system (Ikemura & others, 2002).

Some authors recommend enamel etching with phosphoric acid in addition to using a mild self-etch primer (Torii & others, 2002). Conversely, other research has reported that pre-treatment with phosphoric acid had little effect on improving enamel bonding effectiveness with most self-etch primers (Ernst, 2004). Dentin etching with phosphoric acid prior to using mild self-etch primers was reported to negatively affect dentin bonding (Erhardt, Cavalcante & Pimenta, 2004; Ernst, 2004). This approach may be responsible for the formation of an incomplete infiltration of the resin monomer through the over-etched dentinal surfaces.

In this study, pre-treatment with phosphoric acid was eliminated prior to application of a "mild" self-etch primer. FL-Bond performed as well as total-etch Prime & Bond NT on both enamel and dentin when teeth were not exposed to hydrogen peroxide. Conversely, a reduced performance of FL-Bond was recorded at the enamel cavosurface margin after tooth whitening.

It may be hypothesized that, after tooth whitening, the increased microleakage score reported with FL-Bond at the enamel cavosurface margins may be the result of the alteration of microhardness, roughness and the chemical composition of enamel and dentin produced by the bleaching agent, thus interfering with the FL-Bond chemical interaction with enamel. Previous studies support the theory that enamel does not undergo irreversible changes when subjected to an in-office or at-home whitening protocol (Spalding, Taveira & De Assis, 2003; Araujo & others, 2003). Low pH drinks and foods have demonstrated similar effects on enamel *in vitro* (Hughes & others, 2000; Hunter & others, 2000). Tooth structure alteration has been reported to be less significant on enamel than on dentin and cementum (Hunter & others, 2000; Zalkind & others, 1996). The dentin/cementum interface was not affected after exposure to hydrogen peroxide in this study. This may suggest that the capacity of the self-etch primer to adhere to cementum/dentin could be more adequate than adherence to enamel, resulting in a substrate-adhesive interaction less affected by chemical agents.

Teeth in the Niveous group were subjected to a very intense bleaching treatment with 33% hydrogen peroxide for five consecutive days. This is rarely reproducible in clinical conditions. Alternately, a recent clinical study reported the use of either 35% or 38% hydrogen peroxide for three consecutive days in a split mouth design. This treatment was also associated with the use of 10% carbamine peroxide (Deliperi, Bardwell & Papathanasiou, 2004a).

In this study, prolonged bleaching treatment was completed to further elucidate the effect of tooth-whitening agent on the two adhesive systems. For the same reason, Class V Giomer resin-based restorations were performed just a week after completing bleaching treatment. Conversely, waiting at least two to three weeks before restoring teeth that have undergone bleaching treatment is recommended (Goldstein & Garber, 1995).

The total-etch adhesive system Prime & Bond NT was previously tested with a very similar performance *in vitro* (Deliperi & others, 2003, 2004b). In this study, Prime & Bond NT performed very well when used both in the Niveous and control groups. This may suggest that the use of phosphoric acid can help to create a deeper micro-mechanical interlocking; this phenomenon may specifically contribute to improved performance of the total-etch adhesive system on enamel.

No significant difference was recorded between the bonding effectiveness to enamel and dentin of both adhesive systems in the control group. This corroborates the data reported by other authors who were surveyed (Van Meerbeek & others, 2003).

Further research should provide enlightening data on the *in vitro* and *in-vivo* performance of the giomer resin-based material when used in combination with differing total-etch and self-etch adhesive systems.

Giomer resin-based material is alleged to have a similar fluoride release and recharge as GICs but without the physical and optical disadvantages of GICs. Further independent studies are required to validate the behavior of giomers both *in vitro* and *in vivo*; the effect of aging on their caries inhibition properties also warrants investigation.

CONCLUSIONS

A scarce microleakage score was reported using both the manufacturer-recommended self-etch adhesive FL-Bond and the total etch Prime & Bond NT in the control group. However, the interaction of self-etch adhesive systems with hydrogen peroxide exposed enamel deserves further investigation.

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