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Microtensile bond strength to ground enamel by glass-ionomers, resin-modified glass-ionomers, and resin composites used as pit and fissure sealants

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Summary *Objectives*. To measure the microtensile bond strength to ground enamel of different types of materials used as pit and fissure sealants in combination with different substrate conditioners.

Methods. From 40 sound extracted molars, eight groups of five teeth were randomly formed. The experimental groups were: (1) (C) 37% phosphoric acid/ClinPro Sealant (3M ESPE); (2) (G) 37% phosphoric acid/Guardian Seal (Kerr); (3) (E/TF) 37% phosphoric acid/Excite/Tetric Flow (Ivoclar-Vivadent) (4) (OS/UF) 37% phosphoric acid/One Step (Bisco)/UniFil Flow (GC); (5) (OS/Æ) 37% phosphoric acid/One Step/Æliteflo (Bisco); (6) (UB/UF) UniFil Bond/UniFil Flow (GC); (7) (CC/FVII) GC Cavity Conditioner/Fuji VII (GC); (8) (CC/FII) GC Cavity Conditioner/Fuji II LC Improved (GC).

On the buccal of each tooth, a 5 mm high build-up was created by incrementally adding layers of the sealing material on the conditioned enamel. By serially cutting the built-up tooth, multiple beam-shaped specimens about 1 mm$\times$1 mm in cross section were obtained, and loaded in tensile (0.5 mm/min) until failure occurred.

Results. The bond strengths measured in MPa were: (C) 20.41$\pm$11.79; (G) 16.02$\pm$7.99; (E/TF) 24.06$\pm$9.67; OS/UF 15.63$\pm$9.00; (OS/AE) 9.31$\pm$6.05; (UB/UF) 4.96$\pm$3.46; (CC/FVII) 1.70$\pm$2.19; (CC/FII) 2.19$\pm$1.44.

Conclusions. The conventional and the resin-modified glass ionomers measured bond strengths significantly lower than those of any resin-based materials. Failure frequently occurred cohesively within the cement. Flowable composites in combination with phosphoric acid and a total-etch adhesive performed similarly to resin-based materials specifically conceived for sealings, such as ClinPro Sealant

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and Guardian Seal. The bond achieved by resin composite when treating enamel with the self-etching primer used in this study (UniFil Bond) was significantly lower than that developed when the substrate was etched with 37% phosphoric acid.

Introduction

The procedure of pit and fissure sealing, introduced more than thirty years ago, has proved to be effective for caries prevention in several studies,[1] and has a primary role in minimal intervention dentistry.[2]

Resin composites with Bis-GMA monomers are traditionally used for sealing, in combination with 37% phosphoric acid for etching. The composites are lightly filled in order to keep the viscosity low, thus allowing for a deep penetration of the material into pits and fissures,[3] where a resin impregnated layer of enamel is formed, producing the sealing effect.[4]

Low viscosity and high wettability are peculiar properties of a recently introduced class of resin-based materials, the flowable composites.[5] Flowables have been tested as pit and fissure sealants both in vitro[6,7] and in vivo.[8,9]

The potentials of self-etch adhesives in pit and fissure sealings have also been investigated.[7,10-15] Self-etching primers simplify the bonding procedure by combining etching and priming actions into one step and eliminating the need for rinsing.[10,16]

Also glass-ionomer cements have long been used for sealings.[17-21] Glass-ionomer cements, although providing the benefit of fluoride release, however, offer mechanical properties and a wear resistance inferior to composite resins.[21-23] These characteristics have been improved in resin-modified glass-ionomer cements,[21,23-25] which differ from their precursors also for the photopolymerization ability.[26]

The variety of materials available to clinicians for pit and fissure sealing is then evidently wide. A large amount of research data has been collected for sealants. This includes findings from microleakage tests,[7,11,14,29] observations of retention and caries protection,[8,10,19,20,21,23,27,28] conventional and environmental scanning electron microscopy,[15] transmission electron microscopy[15] and bond strength measurements.[12,13,24,25,30] In particular, the strength of the adhesion to enamel of sealing materials has so far been assessed only with shear[13,24,25,30] and traditional tensile tests.[12] However, in recent years, an increasing number of researchers has turned to the microtensile technique as a more accurate method for the assessment of interfacial bond strength.[31]

The purpose of this study was to use the microtensile non-trimming technique to measure the bond strength to ground enamel of different types of materials, which were used as sealants in combination with different substrate conditioners. The tested null hypothesis was that there were no statistically significant differences in the strength of the adhesion achieved on enamel by the different materials tested.

Materials and methods

A sample of 40 extracted human third molars was collected. The teeth were intact and free from caries, and had been extracted for periodontal or orthodontic reasons.

The sample was randomly divided into eight groups of five teeth each. The experimental groups differed for the material used as a sealant and/or for the procedure followed in enamel conditioning. The eight groups were thus defined:

Group 1: 37% phosphoric acid (Scotchbond Etchant 3M ESPE, St Paul, MN, USA) and ClinPro Sealant (3M ESPE);

Group 2: 37% phosphoric acid (Gel Etchant, Kerr, Orange, CA, USA) and Guardian Seal (Kerr);

Group 3: 37% phosphoric acid (Total Etch, Ivoclar-Vivadent, Schaan, Liechtenstein), Excite and Tetric Flow (Ivoclar-Vivadent);

Group 4: 37% phosphoric acid (All Etch, Bisco, Schaumburg, IL, USA), One Step (Bisco) and UniFil Flow (GC, Tokyo, Japan);

Group 5: 37% phosphoric acid (All Etch, Bisco), One Step and Æliteflo (Bisco);

Group 6: UniFil Bond and UniFil Flow (GC);

Group 7: GC Cavity Conditioner and Fuji VII (GC; in US Fuji VII is commercialized as Fuji TRIAGE);

Group 8: GC Cavity Conditioner and Fuji II LC Improved (GC).

After a storage period never longer than 1 month in a 37° saline solution, on the buccal aspect of each tooth (Fig. 1a) a flat surface was created in enamel by means of a 180-grit wet sand paper (Buehler, Lake Bluff, IL, USA) (Fig. 1b), taking care not to expose the underlying dentin. The surface thus

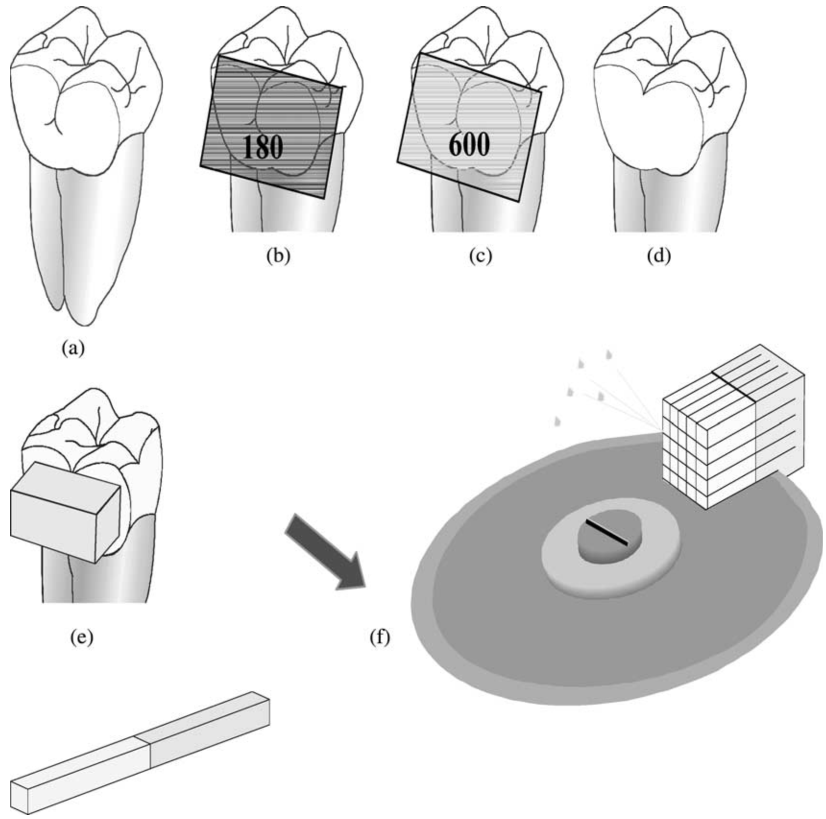

Figure 1 Procedure for the preparation of microtensile specimens. On the buccal aspect of a sound extracted molar (a) a flat enamel surface was created by grinding with a 180-grit (b) and a 600-grit (c) wet sand paper. On the bonding surface thus obtained (d), a block of sealant material was progressively built up (e), after having treated the enamel according to the instructions of the sealant's manufacturer. By means of a water cooled diamond saw, the built-up tooth was serially sectioned into multiple 1 mm × 1 mm sticks (f). (From Ferrari M et al.,[42] modified).

created was then polished with a 600-grit polishing paper (Buehler) (Fig. 1c), rinsed with a water spray, and lightly dried (Fig. 1d).

Then, the enamel surface was prepared for adhesion following different procedures depending on the group. The chemical composition and handling steps of the tested materials are reported in Table 1.

After having prepared the enamel for bonding, the material meant for pit and fissure sealing, be it a resin-based material (Guardian Seal, ClinPro Sealant, UniFil Flow, Tetric Flow, and Æliteflo), or a glass-ionomer (Fuji VII), or a hybrid ionomer (Fuji II LC Improved), was used to build up on the bonding surface a block of about 5 mm in height (Fig. 1d). The build-up was created by progressively adding 2 mm-thick increments of material, which were singularly cured for 20 s with the already mentioned curing device.

After a 24 h-storage in a 37° saline solution, each tooth was longitudinally cut into a series of 1 mm-thick slabs by means of a water-cooled diamond blade (Isomet, Buehler). By rotating the specimen 90° and sectioning it again lengthwise, 15–20 sticks about 1 mm × 1 mm in cross-section were obtained per tooth (1e). Each stick was made up by enamel for about a half of its length and by the sealing material for the remaining portion (1e).

Each microtensile specimen was then glued with cyanoacrylate (Zapit, Dental Ventures of America, CA, USA) to the two free-sliding parts as previously described.[32] This jig is designed to transmit the specimen purely tensile forces, without any torquing component, when mounted on a universal loading machine (Controls, Milano, Italy). The tensile load was applied at a cross-head speed of 0.5 mm/min, until specimen failure occurred. At this point the load at failure in Newtons was recorded, and the specimen's fragments cautiously removed from the grips with a scalpel. The cross-sectional area at the site of fracture was measured to the nearest 0.01 mm with a digital caliper, in order to calculate the bond strength at failure in MegaPascals. For each specimen the mode of failure was also defined as either adhesive, cohesive in enamel, or cohesive in sealing material by examining the fractured stick with an optical microscope (Bausch&Lomb, Rochester, USA) at 200 magnifications.

Table 1 Chemical composition and handling of the tested materials.

Group	Material	Manufacturer	Batch#	Chemical composition	Handling
I	Scotchbond Etchant	3M ESPE	3BN	37% H_3PO_4	A, D
	ClinPro Sealant		1 AR	Bis-GMA, TEGDMA, EDMAB, CQ, BHT, silica, TBATFB, TiO_2, rose Bengal sodium (C.I. 45440)	
II	Gel Etchant	Kerr	31297	37% H_3PO_4	A, D
	Guardian Seal		104818	Fluoride releasing monomer (agent BF_3), Bis-GMA, CQ, silica	
III	Total Etch	Ivoclar-Vivadent	E25773	37% H_3PO_4	A, B, D
	Excite		E37320	HEMA, TEGDMA, phosphonic acid acrylate, silicon dioxide, catalysts, stabilizers, ethanol	
	Tetric Flow		E46593	Bis-GMA, UDMA, TEGDMA, barium glass, ytterbium trifluoride, Ba-Al fluorosilicate glass, silicon dioxide, catalysts, stabilizers, pigments	
IV	All Etch	Bisco		37% H_3PO_4	A, B, D
	One Step		0200002921	BPDMA, HEMA, acetone, glass filler	
	UniFil Flow	GC	0108061	Aluminofluorosilicate glass, UDMA, dimethacrylate resin, silica fine powder, photoinitiator, pigment	
V	All Etch	Bisco		37% H_3PO_4	A, B, D
	One Step		0200002921	BPDMA, HEMA, acetone, glass filler	
	Æliteflo		0100013461	PTMDA, barium glass, catalysts, stabilizers, pigments	
VI	UniFil Bond	GC	0111061	Primer: HEMA, 4-META, CQ, ethanol, distilled water. Bonding: UDMA, HEMA, silica filler, CQ	C, D
	UniFil Flow		0108061	Aluminofluorosilicate glass, UDMA, dimethacrylate resin, silica fine powder, photoinitiator, pigment	
VII	GC Cavity Conditioner	GC	0206131	20% PAA	E, F
	Fuji VII		0206335	Powder: Aluminofluorosilicate glass, pigment. Liquid: PAA, distilled water, polybase carboxylic acid	
VIII	GC Cavity Conditioner	GC	0206131	20% PAA	E, F
	Fuji II LC Improved		0206204	Powder: Aluminofluorosilicate glass, pigment. Liquid: PAA, distilled water, HEMA, CQ	

BHT, butylated hydoxytoluene; Bis-GMA, bisphenol A diglycidyl methacrylate; BPDMA, byphenyl dimethacrylate; CQ, camphorquinone; EDMAB, ethyl 4-(dimethylamino)benzoate; HEMA, hydroxyl ethyl methacrylate; 4-META, 4-methacryloyloxyethoxycarbonylphthalic anhydride; PAA, polyacrylic acid; TBATFB, tetrabutylammoniumtetrafluoroborate; TiO_2, titanium dioxide; TEGDMA, triethylene glycol dimethacrylate; UDMA, urethane dimethacrylate. A, apply etching for 30 s, rinse, and dry until the enamel surface appears chalky; B, apply adhesive and light cure for 20 s; C, apply the primer solution and leave it undisturbed for 20 s, gently dry for 5 s until the enamel surface appears glossy; D, apply the resin composite and light-cure for 20 s; E, condition with 20% polyacrylic acid for 10 s, wash and gently dry; F, apply the GIC and light-cure for 20 s.

Statistical analysis

All the sticks that failed prematurely while being cut or glued were included in the statistical calculations as 'zero bond' values.

As a preliminary linear regression analysis showed that the tooth of origin did not have a significant influence on the measured bond strength, within each group the data from all the teeth were pooled together, and the statistics were calculated per stick.

Square root transformation of bond strength data was performed in order to ensure for normality of data distribution and allow for the application of

One-Way Analysis of Variance to test for significance of the differences among the eight experimental groups. The Newman–Keuls test was used for multiple comparisons.

For all the analyses the level of significance was set at the $P=0.05$.

Results

Table 2 reports mean and standard deviation values of the microtensile bond strength together with the percentage of premature failures for all the experimental groups. In the table the statistically significant differences are highlighted.

The highest bond strength values to enamel were achieved by the flowable composite Tetric Flow used in combination with the adhesive Excite (24.06±9.67 MPa) and by ClinPro Sealant (20.41±11.79 MPa). Lower bond strengths, although statistically similar to that of ClinPro Sealant, were established by Guardian Seal (16.02±7.99 MPa) and UniFil Flow, used in combination with One Step (15.63±9.00 MPa). The same adhesive measured a lower bond strength when utilized with the proprietary flowable composite as Æliteflo (9.31±6.05 MPa). This difference was, however, statistically insignificant. The weakest bonds were attained by UniFil Flow in combination with the self-etch adhesive UniFil Bond (4.96±3.46 MPa), as well as by the traditional (1.70±2.19 MPa), and the resin-modified glass-ionomer (2.19±1.44 MPa).

Regarding the type of failure, all of the Fuji II specimens that failed prematurely; in fact, they exhibited a cohesive fracture within the hybrid ionomer. Also in half of the prematurely failed Fuji VII sticks the fracture was cohesive within the cement. All of the other tested specimens failed adhesively at the bonded interface.

Discussion

The clinical success of sealings depends on the ability of the material to solidly adhere to the enamel surface and securely isolate pits and fissures from the oral environment. As an indicator of a sealant retention ability, the strength of its bond to enamel can be measured in vitro with different techniques.

Although providing several advantages over shear and conventional bond strength tests,[31,33] never before had the microtensile technique been applied to quantify the adhesive potentials of sealing materials. According to researchers routinely resorting to microtensile for adhesion testing, the greatest value of this technique lies in the ability to more accurately assess the interfacial bond strength between material and dental substrate. This is the result of the uniform stress distribution expectedly occurring over small-sized specimens.[31,33]

In the current study, as a matter of fact, the microtensile test led to the assessment of interfacial bond strength on the great majority of the prepared specimens. Only for those Fuji II and Fuji VII specimens that failed cohesively within the cement was the measurement of the interfacial bond strength precluded. The frequent occurrence of cement fractures in the Fuji II and Fuji VII groups can reasonably be related to the relatively low cohesive strength of hybrid and conventional glass-ionomers, as compared with resin-based materials. The clinical implications of this finding will be further discussed.

Among the different variants of the microtensile technique, the non-trimming method producing 1 mm×1 mm-thick beams was chosen, as a previous investigation had shown that such shape and size were the most appropriate for microtensile bond strength testing on specimens from enamel.[34]

Table 2 Mean and standard deviation values of the microtensile bond strengths measured in each experimental group.

Group	Mean (MPa)	S.D.	Premature failures (%)
H_3PO_4, ClinPro Sealant[a,b]	20.41	11.79	4.16
H_3PO_4, Guardian Seal[b]	16.02	7.99	8.00
H_3PO_4, Excite, Tetric Flow[a]	24.06	9.67	0.00
H_3PO_4, One Step, UniFil Flow[b]	15.63	9.00	0.00
UniFil Bond, UniFil Flow[c,d]	4.96	3.46	13.33
H_3PO_4, One Step, Æliteflo[c]	9.31	6.05	18.75
Polyacrylic acid, Fuji VII[e]	1.70	2.19	54.54
Polyacrylic acid, Fuji II[d,e]	2.19	1.44	16.00

Also the percentage of specimens that failed before prematurely during cutting or gluing is reported. Statistically similar groups are labeled with the same superscript letter ($P<0.05$).

Regarding the procedure of substrate preparation, we are aware that the substrate obtained by grinding the buccal tooth surface with 180- and 600-grit sand paper may to some extent differ from the unground enamel of natural pits and fissures; however, it was necessary to create a flat bonding surface in enamel for the purpose of proper microtensile specimens cutting. The same protocol for substrate preparation had been previously followed in a laboratory research, where the shear bond strength of sealants to bovine enamel had been measured.[24]

As for the materials tested in this investigation, they were all of recent introduction on the market, and have properties that make them suitable for pit and fissure sealing.

ClinPro Sealant is a light-cure, low viscosity, fluoride releasing resin composite. It is provided with a light-activated color-change feature from pink to opaque, which helps the clinician thoroughly place the correct amount of material. The manufacturer recommends using ClinPro Sealant in combination with 37% phosphoric acid, in order to create microporoties by selectively dissolving the enamel inorganic component, as well as to change the enamel surface from a low energy, weakly reactive, hydrophobic state to a high-energy, strongly reactive hydrophilic substrate, into which the sealant is attracted to flow.

Less strong but still dependable was the adhesion developed by the other material specially marketed for sealings, Guardian Seal. Guardian Seal is a light-cure, 30% filled resin, with fluoride release ability from both the monomer and the filler (manufacturer's information). It requires 37% phosphoric acid for enamel etching.

Although not specific, flowable composites are, however, suitable for pit and fissure sealings thanks to their viscous properties.[5] The decision to evaluate the bonding ability of flowables in comparison with materials traditionally used as sealants was dictated by the consideration that these versatile materials are very likely to be present in the dispensary of the general practitioner, as routinely used for Class III and V restorations, for small occlusal restorations, and by some clinicians for the gingival wall of direct Class II restorations.[35] A flowable composite would also be the material of choice when a fissurotomy is indicated.[36,37]

Concerning the use of these materials for preventively sealing pits and fissures, Autio-Gold[8] reported that a flowable composite tested clinically as a sealant did not perform any better than a conventional unfilled resin in terms of retention and caries protection. However, it should be pointed out that no bonding agent was used in this trial prior to the placement of the flowable. In our investigation, on the other hand, a one-bottle adhesive was applied after phosphoric acid etching in Groups 3, 5, and 6. There is indeed indication in the literature that applying an intermediary layer of adhesive solution onto the etched enamel prior to sealant placement significantly improves marginal adaptation[8] and sealant retention.[38]

In the present investigation flowable composites provided a satisfactory performance in terms of bond strength when they were used in combination with phosphoric acid and an adhesive agent. As a matter of fact, it was a flowable material, Tetric Flow, with the proprietary total-etch adhesive Excite, that recorded the highest bond strength of all (Table 1). Tetric Flow had previously been tested as a sealant in combination with a one-bottle bonding, and proved even more effective than Helioseal F in obturating deep fissures.[7]

Significantly less strong was the adhesion to enamel of the flowable composites bonded with One Step, UniFil Flow and Æliteflo. However, this finding can plausibly be related to the greater bonding ability of Excite as compared with One Step, rather than to properties of the flowables themselves.

Far less dependable the bond to enamel appeared when the flowable UniFil Flow was used in combination with the proprietary self-etch adhesive UniFil Bond (Table 1). Although a simplification of the bonding procedure is understably appealing to the clinician, in particular when working on uncooperative patients, there is abundant evidence that on uncut enamel etching with 37% phosphoric acid creates more favourable conditions of adhesion than the acidic resin monomers of even the most aggressive self-etching primers.[12,13,16,39] As regards in particular UniFil Bond, Torii et al.,[12] who recently compared in vitro this self-etching primer with a traditional and a total-etch system, report that UniFil Bond measured the lowest tensile bond strength to enamel. Microscopically, the self-etching primer appeared able to create at the interface with enamel only shallow penetrations, which evidently differed from the thick tag-like extensions penetrating into the substrate etched with phosphoric acid.[12,40]

The results of our microtensile test, in agreement with previous studies[11,12,16] confirm an impaired bonding ability of the self-etching primer as compared with 37% phosphoric acid. It is obviously understood that for a thorough evaluation of the potentials of self-etch adhesives in sealings, also microleakage tests should be carried out, and a clinical trial should eventually complete the investigation. It is, however, true that, based on the findings of previous microleakage tests,[11,14] the use of self-etching primers for bonding to pits and fissures does not appear advisable.

Contrary to the expectations, a significantly higher bond strength was reported when the adhesive One Step was used in combination with UniFil Flow than when it was employed with the proprietary flowable composite Æliteflo. The different filler content of the two composite resins may provide an explanation for this finding. UniFil Flow has a higher filler content than Æliteflo (67 versus 60%, respectively, according to the manufacturers). It is then plausible that UniFil Flow, undergoing a lower polymerization shrinkage, stressed the newly established adhesive interface less than Æliteflo on curing.

Some considerations have also to be made regarding the performance of the ionomer-based materials in our trial. Fuji VII is a glass-ionomer cement specifically targeted for early protection in erupting first and second molars. According to the manufacturer, in such a working field, where saliva contamination is very likely to occur, the bonding mechanism of resin-based materials would be seriously compromised, whereas the chemical adhesion of glass-ionomers can effectively take place also on a moist substrate. Another peculiarity of Fuji VII entails its polymerization. Although of a chemical nature, the setting reaction of Fuji VII can be accelerated by irradiation with an halogen or a plasma light, whose energy is captured by the material thanks to the pink chroma.

Properties such as fluoride release and the ability to chemically adhere to enamel even in the presence of humidity and without the need to preliminarily etch the substrate would make glass-ionomers preferable to resin-based materials for pit and fissure sealing. However, it should be pointed out that conditioning the dental substrate with an acid, though considered unnecessary with glass-ionomers, in fact significantly increases the shear bond strength to enamel.[24] In particular, a 20% polyacrylic acid solution has been reported to produce a favorable combination of good shear bond strength and relatively limited enamel erosion.[24] This same solution (GC Cavity Conditioner) was used in our study to treat the enamel surface prior to the placement of Fuji VII and Fuji II.

In addition, several clinical studies have documented a lower retention rate of glass-ionomer sealant as compared with resin-based materials.[18,20,26] This may be related to the relatively poorer mechanical properties and wear resistance of glass-ionomer cements.[18,23] These characteristics have been improved in resin-modified ionomers by the introduction of HEMA monomers.[21,23–28,41]

In our microtensile study the conventional glass-ionomer Fuji VII and the resin-modified ionomer Fuji II LC Improved revealed the lowest bonding potential to enamel of all the tested materials. The fragility of the specimens was such that 16% of the Fuji II LC sticks and more than half of the Fuji VII sticks fractured before being tested. As already mentioned, many of these premature fractures in fact occurred cohesively within the cement portion. This type of failure of the material may not mean a complete failure of the sealing intervention from a clinical standpoint, for, as long as the cement adheres to enamel, the protective effect is not lost. Together with fluoride release, this is the explanation some authors provide for the finding that with glass-ionomer sealants, despite what may macroscopically appear as a conspicuous loss of material, the caries-preventive effect is somewhat maintained.[17,21,24,27,28] However, it should also be considered that thin layers of glass-ionomer material, though still adhering to the substrate, are anyway at risk of rapidly wearing off.[23]

Finally, the glass-ionomer and the resin-modified ionomer might have performed better if a varnish had been applied to initially protect the material from water contamination and prevent its hygroscopic expansion, as suggested by the manufacturer. However, the opinion of the authors is that the protective coating should not significantly affect the adhesive potential of the material, but rather have an effect on microleakage prevention.

In conclusion, the findings of our microtensile study can be thus summarized:

- the tested null hypothesis that there were no statistically significant differences in the strength of the adhesion achieved on enamel by the different materials on trial had to be rejected;
- resin-based materials developed on enamel significantly stronger bonds than a glass-ionomer and a hybrid ionomer;
- if microtensile bond strength can be taken as an indicator of the material's retention ability, flowable composites, used as pit and fissure sealants in combination with phosphoric acid and a total-etch adhesive, should perform similarly to resin-based materials specifically conceived for this purpose, such as ClinPro Sealant and Guardian Seal;
- the bond achieved by treating enamel with the self-etching primer used in this study (UniFil Bond) was significantly weaker than that

developed when the substrate was etched with 37% phosphoric acid;
- the retention ability expressed by a conventional glass-ionomer and a resin-modified ionomer was inferior to that of any resin-based material.

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