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# Push-out bond strength and SEM evaluation of a new bonding approach into the root canal

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## ABSTRACT

**O**bjective: This study evaluated the performance of different adhesive systems in fiber post placement aiming to clarify the influence of different hydrophobic experimental blend adhesives, and of one commercially available adhesive on the frictional retention during a luting procedure. Material and Methods: One luting agent (70 Wt% BisGMA, 28.5% TEGDMA; 1.5% p-tolyldiethanolamine) to cement fiber posts into root canals was applied with 4 different adhesive combinations: Group 1: The etched roots were rinsed with water for 30 s to remove the phosphoric acid, then rinsed with 99.6% ethanol for 30 s, and blot-dried. A trial adhesive (base to catalyst on a 1:1 ratio) was used with an experimental luting agent (35% Bis-GMA, 14.37% TEGDMA, 0.5% EDMAB, 0.13% CQ); Group 2: A trial adhesive (base to catalyst on a 1:2 ratio) was luted as in Group 1; Group 3: One-Step Plus (OSP, Bisco Inc.) following the ethanol bonding technique in combination with the luting agent as in Group 1; Group 4: OSP strictly following the manufacturer's instructions using the luting agent as in Group 1. The groups were challenged with push-out tests. Posted root slices were loaded until post segment extrusion in the apical-coronal direction. Failure modes were analyzed under scanning electron microscopy. Results: Push-out strength was not significantly influenced by the luting agent ( $p > 0.05$ ). No statistically significant differences among the tested groups were found as Group 1 (Exp 1 – ethanol-wet bonding technique) = Group 2 (Exp 2 – ethanol-wet bonding technique) = Group 3 (OSP – ethanol-wet bonding technique) = Group 4 (control, OSP – water-wet bonding technique) ( $p > 0.05$ ). The dominating failure modes in all the groups were cohesive/adhesive failures, which were predominantly observed on the post/luting agent interface. Conclusions: The results of this study support the hypothesis that the proposal to replace water with ethanol to bond fiber posts to the root canal using highly hydrophobic resin is plausible, but this seems to be more the proof of a concept than a clinically applicable procedure.

**Key words:** Root canal. Luting cement. Hydrophobic adhesives. Dentinal bonding.

## INTRODUCTION

The process of hybrid layer formation in etch-and-rinse dentin bonding systems (DBS) involves the penetration of resin monomers into a delicate layer of unsupported collagen fibrils exposed by the etching agent (usually 35-37% phosphoric acid). The etching agent was inactivated, and removed by copious air/water spray. This is

because etch-and-rinse DBS impregnate the substrate in accordance with the "water-wet" bonding technique, i.e. collagen fibrils should remain wet to avoid excessive shrinkage due to desiccation that can impair resin impregnation. The residual water within the fibrillar network was then displaced by hydrophilic monomers to allow the gradual penetration of hydrophobic monomers into the demineralized dentin layer. The process of

hydrophilic resin impregnation into water-wet dentin collagen matrices consists in a passive diffusion mechanism<sup>20-23</sup>. Solvents also have an important role in monomer impregnation as they can reduce resin viscosity, and increase the water substitution rate, thus facilitating water displacement within the demineralized collagen fibrils. The more common solvents used for DBS are ethanol, acetone, and water<sup>14</sup>.

As hydrophobic resin blends showed higher stiffness, improved stability over time, and reduced water uptake<sup>11,15,16</sup> when compared to more hydrophilic ones<sup>10</sup>, hydrophobic monomers should be preferred to produce a stable bond over time. However, if the adhesive blend is too hydrophobic, suboptimal impregnation occurs since the solvents cannot replace all the residual water within the demineralized dentin collagen fibrils. This insufficient resin penetration leads to the formation of a hybridoid layer characterized by voids and porosities with reduced sealing ability<sup>3,4,8,20</sup>. Areas of incomplete resin impregnation can result in nanoleakage, and can be identified using a tracer (silver nitrate) under an scanning electron microscopy (SEM)<sup>19</sup>.

In order to coax hydrophobic monomers into demineralized dentin collagen matrices, the "ethanol-wet bonding technique" has recently been proposed<sup>16,22</sup>. This technique is characterized by sequential rinses with ethanol at ascending concentrations to replace interfibrillar water<sup>16,18,22</sup>. Since the ethanol-saturated dentin is more compatible with hydrophobic resin monomers, collagen shrinkage is prevented, and impregnation is facilitated<sup>6</sup>. This technique has been shown to produce adhesive interfaces with higher bond strength, reduced interfacial nanoleakage expression, and increased stability over time when compared to the "water-wet bonding technique"<sup>9</sup>. The use of the ethanol-wet bonding technique has also been shown to produce encouraging results when used to lute posts to intra-radicular dentin<sup>5</sup>. In addition, the possibility of using high hydrophobic resin possibly minimizes endogenous collagenolytic activities<sup>9</sup>.

Despite promising *in vitro* results, the ethanol-wet bonding technique is time consuming, as several ethanol rinses should be performed to completely replace the residual water within the dentin collagen, and to allow hydrophobic monomers to infiltrate into a fully ethanol saturated dentin. Recently, a simplified ethanol wet-bonding procedure with the reduction of time of application has been proposed to bond to coronal dentin<sup>17</sup>. However, no studies have clarified if the proposed "simplified ethanol-wet bonding technique" could be beneficial when luting fiber posts to intra-radicular dentin.

The aim of the present study was to compare the bond strength and interfacial morphology created by an experimental or a commercially-available DBS in association with resin-based cements used to lute fiber posts within the endodontic space, by using the simplified ethanol-wet bonding technique. The null hypotheses tested were that: (1) no differences exist between the push-out bond strengths of hydrophobic experimental resin blends and a commercially available two-step etch-and-rinse adhesive; (2) no differences exist between the push-out bond strengths of a simplified ethanol-wet bonding technique and those of a commercially available two-step etch-and-rinse adhesive.

## MATERIAL AND METHODS

### Specimen preparation

Twenty single-rooted premolars showing a single-canal, and extracted for orthodontic reasons were selected for the study after informed consent was obtained under a protocol approved by the University of Siena (Siena, Italy). Exclusion criteria were teeth shorter than 20 mm, apex larger than a size 25 K-file before instrumentation, presence of caries, root fissures, or fractures.

The teeth were hand-scaled, and stored in 1% chloramine, T at 4°C, and used within 1 month after extraction. Crowns were removed cutting the teeth 2-mm over the cementum-enamel junction, using a slow-speed diamond saw (Micromet, Remet, Casalecchio di Reno, BO, Italy). Canals were shaped using nickel-titanium rotary instruments (size S1, S2 and F3; Protaper, Dentsply-Maillefer, Ballaigues, Switzerland). Root canals were prepared using the Protaper Universal System according to the sequence: S1 and SX for the 2/3 coronal third, and then, instruments S1, S2, F1, F2, and F3 at the work length for the final preparation in accordance with the crown-down fashion to an ISO size 30/0.07 taper. Irrigation with 5% NaOCl was performed (Nicolor 5; Dentale-Ogna, Milan, Italy) during instrumentation using a syringe with a 30G endodontic needle (Perio/Endo Irrigation Needle Biaggio Switzerland). Removal of the smear layer was obtained after irrigation with 3 mL of 17% EDTA for 2 min, followed by 3 mL of saline. After the final rinse, root canals were completely dried with air stream and absorbent paper points (Dentsply-DeTrey, Konstanz, Germany) and filled by lateral condensation of gutta-percha cones and a resin-based sealer (AH-26, Dentsply-DeTrey). The filled roots were coronally sealed with glass ionomer cement (Fuji VII, GC Corporation, Tokyo, Japan) as the coronal temporary restorative material, and stored in 100% humidity in labeled white film containers for 24 h at 37°C.

After removing the temporary coronal seal, the

gutta-percha was removed in all groups using a low-speed universal drill provided by the manufacturer, and keeping at least 4 mm of apical seal. A standardized 7-mm post space was drilled in each root with the #2 drill that corresponded to RelyX Fiber Post size #2 (3M ESPE, St. Paul, MN, USA).

The n value was obtained after a power analysis of 80%, in order to calculate the minimum effective size that is likely to be detected in a study using a given sample showing that the sample size was adequate; teeth were equally (n=5) and randomly divided into 4 groups according to the adhesive procedure (Table 1). All monomers were purchased by Sigma-Aldrich, St. Louis, MO, USA, Group 1: experimental adhesive (35% Bis-GMA, 14.37% TEGDMA, 0.5% EDMAB, 0.13% camphoroquinone (CQ); co-monomers to ethanol on a 1:1 ratio) applied in accordance with the simplified ethanol-wet bonding technique; Group 2: experimental adhesive (composition similar to Group 1 with co-monomers to ethanol on a 1:2 ratio) applied in accordance with the simplified ethanol-wet bonding technique; Group 3: One-Step Plus (OSP) applied in accordance with the simplified ethanol-wet bonding technique; Group 4: OSP applied strictly following the manufacturer's instructions (i.e. water-wet bonding technique; control group). Chemical compositions of all materials and systematic clinical procedures are described in Table 1. In brief, root canal walls were etched with 32% H<sub>3</sub>PO<sub>4</sub> gel (Bisco Inc., Schaumburg, IL, USA) for 15 s, using an intracanal tip, then specimens of groups 1, 2 and 3 were rinsed with water for 15 s using an endodontic needle, and root canals were filled with 99.6% ethanol (Sigma-Aldrich) for 30 s. Canals were then gently dried with paper points leaving an evident visual aspect of ethanol-wet saturated surfaces. Adhesive blends of groups 1-3 were then immediately applied on ethanol-wet dentin, and light-cured using a conventional quartz-tungsten-halogen light (600-mW/cm<sup>2</sup> output; VIP; Bisco Inc.). The final ethanol rinse was avoided in specimens of group 4 that were prepared applying OSP on water-wet dentin.

The conical epoxy resin posts size #2 (RelyX-

Posts; 3M ESPE) were cleaned in ethanol, and surface-treated with a silane solution (Porcelain Primer; Bisco) using a disposable brush, and gently air-dried for 5 s. An unfilled resin (Scotchbond MP/3M ESPE) was used as a luting agent (2-hydroxyethylmethacrylate, bis-phenol, A diglycidylmethacrylate, photoinitiator; Table 1), and placed into the canal using a disposable syringe. All fiber posts were then seated under finger pressure, and the excess of luting material was removed while maintaining a seal of the exposed dentin along the coronal part of the root. Light curing was performed using a conventional quartz-tungsten-halogen light (600-mW/cm<sup>2</sup>; VIP; Bisco Inc.) by placing the light tip perpendicularly on the post for 40 s. All bonded specimens were then placed in individually labeled containers in 100% humidity for 24 h at 37°C.

### Preparation of specimens for the push-out strength test

After 24 h, the portions of the roots corresponding to the bonded fiber post were transversely sectioned into 1-mm-thick serial slices using a slow speed diamond saw under water irrigation (Micromet M, Remet; Casalecchio di Reno, Bologna, Italy). The apical surfaces of the slices were marked with a permanent black-ink dot. The push-out load was applied using a universal testing machine (Controls S.P.A., Milano, Italy) at a crosshead speed of 0.5 mm/min. The apical surface displaying the ink dot was placed facing the punch tip ensuring that loading forces were introduced in an apical to coronal direction. With regard to the tapered design of the post, three different sizes of punches were used for the push-out testing. The diameter of the punch pin was 1.2 mm for the coronal slices, 1.0 mm for the middle slices, and 0.8 mm for the apical slices. This guaranteed that the strength was applied as more adequately as possible to the bonded area during the loading process.

Bond failure was manifested by the complete dislodgment of the fiber-post from the root section. Push-out strength data were converted to MPa by dividing the load in Newton by the bonded surface area ( $S_L$ ) in mm<sup>2</sup>. Stereomicroscopic images<sup>12</sup>

**Table 1-** Median push-out bond strength values\* (SD) expressed in MPa, number of specimens (N), and percentage of failure mode distribution recorded in the experimental groups. OSP= One-Step Plus

Groups	Bond Strength (SD)	Number of Slices	Failure mode (%) A/M/PA/C
1. Experimental adhesive 1 + simplified ethanol-wet bonding technique	6.9 (5.9) <sup>a</sup>	33	0/38.89/11.11/50
2. Experimental adhesive 2+ simplified ethanol-wet bonding technique	6.7 (5.4) <sup>a</sup>	32	0/0/40/60
3. OSP + simplified ethanol-wet bonding technique	6.8 (4.3) <sup>a</sup>	32	0/32/4/64
4. OSP + water-wet bonding technique (Control)	6.9 (5.1) <sup>a</sup>	31	7.14/25/0/67.86

of the upper and lower sides of each specimen were obtained, and the failure limits were traced with a closed line using image analysis software (Image Pro Plus 5.0; Media Cybernetics, Bethesda, MD, USA). Limits were then measured (after software calibration) in accordance with Ferrari, et al.<sup>7</sup> (2009), and the thickness of the slice was individually measured using a digital caliper with 0.01-mm accuracy. SL was calculated as the lateral surface area of a truncated cone using the formula:

$$S_L = \pi(R+r)[(h^2+R-r)^2]^{0.5}$$

where R is the coronal post radius, r the apical post radius, and h the thickness of the slice.

Modes of failure were classified as (A) adhesive

between dentin and cementing agent, (M) mixed, (PA) adhesive between post and cementing agent, or (C) cohesive if cementing agent failures were assessed with a stereomicroscope (Nikon SMZ645, Nikon, Tokyo, Japan) at 30x magnification.

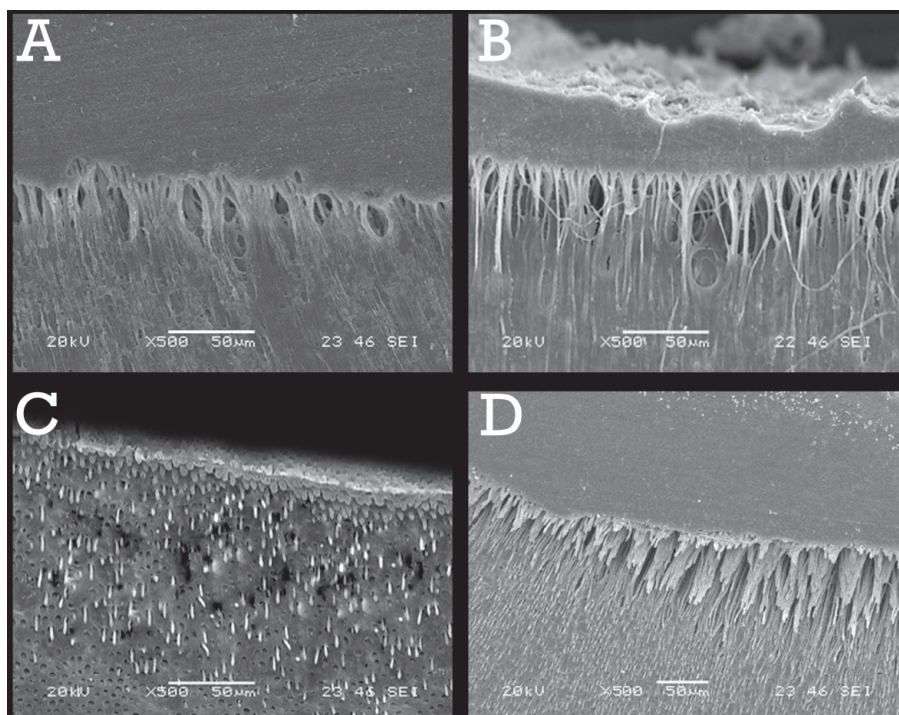
### Statistical analysis

The normally distributed data (Kolmogorov-Smirnov test) with no homogeneous group variances (Levene's test) were analyzed by Kruskal-Wallis's one-way analysis with push-out strength in MPa as a dependant variable. To improve the accuracy of the *post-hoc* statistical testing, the data were analyzed by Dunn's test for multiple comparisons, not including an adjustment for ties. The level of

Dentin Treatment	Groups	Bonding system	Luting agent	Application procedure
32% phosphoric acid etching; rinse after 15 s; air-dry and paper points	1	Adhesive 35% Bis-GMA, 14.37% TEGDMA, 0.5% EDMAB, 0.13% CQ light-cure	Unfilled Resin (batch #4NR, 3M ESPE, St Paul, MN, USA) 2-hydroxyethylmethacrylate (HEMA); Bis-phenol A diglycidylmethacrylate; photoinitiator Light-cure	Procedure: rinsed with water for 30 s to remove the phosphoric acid, then rinsed with 99,6% ethanol for 30 s and blot-dried lightly to obtain a visibly moist. Mix Experimental Adhesive with Ethanol (1:1). Light cured for 20 s. Luting: Apply unfilled resin using a disposable syringe, then light cured for 40 s.
	2	Adhesive as in Group 1	Unfilled resin as in group 1	Rinse with water/ethanol as in group 1. Mix Experimental Adhesive with Ethanol (1:2). Light-cured for 20 s Luting: as in group 1
	3	One-Step Plus (batch # 0500002430, Bisco Inc., Schaumburg, IL, USA) Biphenyl dimethacrylate; 2-hydroxyethyl methacrylate; Acetone; amine; photoinitiator; dental glass	Unfilled resin as in group 1	Rinse with water/ethanol as in group 1 Apply One Step Plus adhesive in 2 coats with agitating movements for 10 s. Gently air dried for 10 s. Light-cure for 10 s. Luting: as in group 1
	4	One-Step Plus	Unfilled resin as in group 1	One-Step Plus procedure: Rinsed with water/air spray for 20 s. Apply adhesive in 2 coats with agitating movements for 10 s. Air dry after 10 s. Light-cure for 10 s. Luting: as in group 1

Figure 1- Chemical composition and application mode of the materials used in the study





**Figure 2-** Scanning electron microscopy images (SEM) (magnification 500x, bar 50 µm) from a representative push-out tested on dentin slice specimens: (A) Experimental Group 1; and (B) Experimental Group 2 showed an evident hybrid layer with long resin tags; (C) One Step Plus/Ethanol/Experimental Luting Agent resulted in the formation of short and discrete resin tags; (D) One Step Plus as the control group using the regular wet bonding technique demonstrated the formation of long, deep, and compact resin tags into the dentin

significance was set at  $p < 0.05$ . The analyses were performed using SigmaStat 3.5 (Windows Version; SPSS, Chicago, IL, USA).

### SEM sample preparation

Representative fractured slices from each group were randomly assigned to scanning electron microscopy (SEM) analysis<sup>22</sup>. Each slice was smoothed with wet silicon carbide paper of decreasing abrasiveness (up to 1200 grit). To analyze hybrid layer morphology and resin tag formation, the specimens were etched with silica free 32%  $H_3PO_4$  (Etching gel; Bisco) for 20 s, and, subsequently, immersed for 2 min in 2.5% NaOCl to remove the organic and mineral components of the dentin, rinsed with water, and dehydrated with 99.8% ethanol to analyze hybrid layer morphology, and resin tag formation. Specimens were then mounted on aluminum stubs, sputter coated with gold (Polaron Range SC7620, Quorum Technologies, Newhaven, England), and observed under a scanning electron microscope SEM (JSM 6060 LV, JEOL, Tokyo, Japan). Micrographs were taken at different magnifications in order to provide an overview of each area, and to evaluate the type of micro morphologic pattern of the representative specimens.

## RESULTS

Mean values and SDs expressed in MPa of push-out bond strength, and numbers of slices/group and failure modes (%) are summarized in Table 1. No premature failures were found during the cutting procedure or during the testing procedure.

No statistically significant differences were found among the tested groups ( $p > 0.05$ ).

### SEM evaluation

The majority of the specimens showed cohesive/adhesive failures on the post/luting agent interface. By the images, Groups 1, 2, and 3 demonstrated a visually deeper penetration of the bonding material different from Group 4 (Figure 1). All groups formed a distinct hybrid layer.

## DISCUSSION

The results of this study showed that experimental hydrophobic resin blends (Groups 1, 2) have similar bond strength values to OSP (Group 3) if applied in accordance with the simplified ethanol-wet bonding technique, and, thus, the first null hypothesis was accepted. In addition, similar bond strength was obtained when OSP was applied with the simplified ethanol-wet bonding technique (Group 3) if compared to control application (Group 4);

thus, the second null hypothesis was also accepted.

The rationale on the use of the ethanol-wet bonding technique is that hydrophobic monomers can better infiltrate the ethanol-saturated demineralized dentin due to reduced polarity of the collagen network to match the low polarity of high hydrophobic resins. Since hydrophobic resin blends have higher stiffness and stability than hydrophilic ones, bonding to dentin with hydrophobic resin blends in association with the ethanol-wet bonding technique showed excellent results both on immediate dentin bonding and on the longevity of the bond created on coronal dentin substrates<sup>2,9,13,18</sup>.

An essential pre-requisite for the achievement of the complete impregnation of the collagen fibrils exposed by acid etching with hydrophobic monomers is that dentin interfibrillar residual water is fully replaced by ethanol, and, since ethanol is a water chasing solvent, it removes water from the tissues. If water remains within the collagen network, hydrophobic monomers cannot fully embed the demineralized dentin substrate, and water-rich domains remain within the hybrid layer, constituting areas of early degrading phenomena over time<sup>3</sup>.

The ethanol-wet bonding technique was initially proposed by five sequential rinses at ascending ethanol concentrations (for 30 s) followed by absolute ethanol (re-applied three times) before the application of an hydrophobic DBS<sup>16</sup>. Because this procedure is time-consuming due to its extended clinical application time, the support of a simplified dehydration protocol could make the ethanol-wet bonding technique more attractive. In a recent study, it was shown that the use of a simplified ethanol wet-bonding technique applied on coronal dentin (a single 30 s application of absolute ethanol) results in a 50% bond strength reduction compared to the "standard" multi-step ethanol-wet bonding technique after 6 months of aging in artificial saliva, and high interfacial nanoleakage expression<sup>17</sup>. It was speculated that the short application time of absolute ethanol is probably ineffective to completely replace water from the etched coronal dentin<sup>19</sup> in which the simulated pulp pressure was applied. Ethanol could rapidly evaporate or be replaced by water permeating from open and funnelled dentin tubules after smear-layer removal.

It can be speculated that the short dehydration protocol proposed by the simplified ethanol-wet bonding technique could be beneficial in luting fiber posts in endodontically treated teeth due to the absence of pulp pressure. Despite the absence of water permeating through the tubules, the results of the present study support the findings of Sadek, et al.<sup>17</sup> (2010) showing that a single final rinse of ethanol for only 30 s before the application of an

experimental hydrophobic adhesive blend does not improve push-out bond strength if compared to the bond produced by a commercially available DBS (OSP) if applied with the "standard" water-wet bonding technique. These data support the hypothesis that optimal impregnation of etched dentin cannot be achieved with the simplified technique probably due to the presence of inter fibrillar water that needs multiple ascending ethanol concentration rinses, and appropriate contact time to allow complete interfibrillar water replacement by ethanol.

In this study, the analysis of the failure modes demonstrated that most failures occurred at the post/luting material interface, and this is in accordance with the results of a recently published investigation<sup>6</sup>. This type of fracture could be due to the lack of chemical union between the cured epoxy resin matrix fiber-post and the unfilled resin (HEMA, Bis-GMA; Figure 2).

Further studies are advisable to confirm the supported hypothesis, and to evaluate the effect of concentration and application time of ethanol rinses to improve the bond to intraradicular dentin.

## CONCLUSION

Within the limitations of this study, it can be concluded that the simplified ethanol-wet bonding technique is not sufficient to enhance the push-out strength in the root canal using the tested materials. The present study supports the hypothesis that the purpose of replacing water with ethanol to bond fiber post to the root canal using highly hydrophobic resin is plausible, but this seems to be more the proof of a concept than a clinical applicable procedure.

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