Long-term Evaluation of the Sealing Ability of Two Root Canal Sealers in Combination with Self-etching Bonding Agents

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Purpose: To compare the short- and long-term sealing ability of root canal fillings consisting of AH-26 and laterally compacted gutta-percha in combination with a self-etching dentin bonding system and the Epiphany-Resilon system.

Materials and Methods: Ninety-eight single-rooted human teeth were used in this study. The root canals were prepared using the step-back technique. Sodium hypochlorite 5% was used as the irrigant solution. The teeth were divided into 4 groups. In groups 1, 3, and 4, irrigation with ethylene diamine tetracetic acid (EDTA) 15% was used to remove the smear layer. In the specimens of all groups, a final irrigation with 5 ml distilled water was performed. In groups 1 and 2, the Nanobond bonding system was used. The specimens in groups 1 to 3 were obturated with AH-26 and laterally compacted gutta-percha. In the last group (4), the Epiphany-Resilon system was used. Microleakage was measured at 7 days, 1 month, and 1 year using a fluid transport model. Two specimens from each group and 4 where only Nanobond had been applied were prepared and examined in a scanning electron microscope.

Results: The Epiphany-Resilon system and the group obturated with AH-26 sealer and gutta-percha, in combination with the self-etching bonding system after removal of the smear layer with EDTA, demonstrated similar sealing ability. Kruskal-Wallis and Mann-Whitney tests showed that he results for these two groups were statistically significanty better than the other groups ($p < 0.05$). All the groups examined showed increased apical leakage over time, but the increase was statistically significant after one year only in the case of the Epiphany-Resilon system.

Conclusion: The use of a self-etching bonding system improved the sealing ability of AH-26 sealer only after removal of the smear layer. Significantly higher microleakage over time was only observed with the Epiphany-Resilon system.

Keywords: self-etching bonding agents, resin-based sealers.

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Successful root canal therapy requires chemomechanical preparation, and then a complete obturation of the root canal system with nonirritating biomaterials. One of the major causes of root canal treatment failures is the incomplete sealing of the root canal[23] and shortcomings in coronal restoration,[22] confirming the necessity of using materials capable of forming a bacteria-proof seal between the root canal system and both the periradicular tissues and the oral cavity.

An important factor in bonding between filler and root canal wall is the smear layer formed on the root canal walls as a result of root canal instrumentation; it acts as a barrier to the adaptation and penetration of root canal sealers into dentinal tubules. The removal of smear layer improves the bonding interface between the sealer and root canal dentin and reduces microleakage.[5,25]

A great variety of root canal filling materials are currently in use, but particularly the use of dentin bonding agents in combination with gutta-percha cones and a resin-based sealer has been the focus of important research.[7,10] The adhesion of composite resins to dentin is known, from experience in restorative dentistry, to be im-

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proved by dentin bonding agents. The bonding agents create a micromechanical interlock between the dentin collagen and resin by forming a hybrid layer.[13]

Self-etching dentin bonding agents were introduced on the dental materials market in response to dentists' needs for materials that were easier to manipulate and less technique sensitive. Since the self-etching materials have higher pH values than the acids used with etch-and-rinse adhesive systems, and the self-etching materials are not rinsed away but remain in situ, the smear layer is incorporated in the bonding layers.[15,35]

In 2004, a new product was proposed based on this strategy: the Epiphany-Resilon system (Epiphany, Pentron Clinical Technologies; Wallingford, CT, USA). This contains Resilon as a core filling material (a thermoplastic synthetic polymer-based material), a resin-based sealer (Epiphany), and a self-etching dentin bonding agent.

Many studies have evaluated the sealing ability of this new obturating system in comparison with epoxy resin sealers and gutta-percha.[2,3,27,34] In these studies, epoxy resin-based sealers were used without the application of a dentin bonding system. The use of dentin bonding agents seems to improve the sealing ability of AH-26 sealer,[10] and the adhesion of this sealer to dentin.[7]

Long-term stability of the tooth-biomaterial bond is very important for maintaining an effective seal. It is known from coronal application that biomaterial/tooth interfaces degrade over time via exposure to water and/or human bacterial enzymes present in saliva. In the case of root canal dentin, the hybrid layer may be degraded by enzymes released by bacteria[24] and by dentin itself.[20] Subsequently, the resultant breakdown products and residual monomers can leach out, compromising the interfacial bond. As far as can be ascertained from the literature, the results of most studies evaluating apical leakage of these products were obtained immediately after or within three months of root filling.[2,36] Only one study[19] compared the sealing ability of gutta-percha/AH Plus and Resilon Epiphany over a longer period (16 months).

The aim of the current study was to compare the sealing ability of an epoxy resin-based root canal sealer (with and without a self-etching dentin bonding system) with the Epiphany-Resilon system at time intervals up to 12 months.

The null hypotheses were that 1. the use of a self-etching bonding agent in combination with the epoxy resin-based sealer would improve the short- and long-term sealing ability of the system as compared to the Epiphany-Resilon, and 2. there would be no difference between the long- and short-term sealing ability of the two obturating systems.

MATERIALS AND METHODS

The materials used were AH-26 sealer (Dentsply DeTrey; Konstanz, Germany), Epiphany sealer (Pentron Clinical Technologies), Resilon cones (Pentron Clinical Technologies), Nanobond dentin bonding system (Pentron Clinical Technologies), and gutta-percha cones (Spident, SPI Dental; Incheon, Korea).

Microleakage Evaluation

The 98 single-rooted human teeth used for this study had been extracted for periodontic or prosthetic reasons and subsequently stored in distilled water before use. The crowns were sectioned with a high-speed bur under water spray so that all roots were 14 to 15 mm long. A #20 K-file was used in all teeth, 1 mm longer than root length, to assure apical patency.

The root canals were prepared with the step-back technique (steps of 1.0 mm between different file sizes) using K- and Hedstrom files (0.02 taper) (Dentsply Maillefer; Ballaigues, Switzerland). The working length was the same for all specimens (14 mm), and a #35 master apical file was used. NaOCl 5% was used as an irrigant solution after the use of each instrument. Delivery of irrigant into the canal was performed with the use of a syringe equipped with a 27-gauge needle. All the specimens were prepared to #55. Paper points were used to dry the root canals.

The teeth were randomly divided into 4 groups of 23 teeth each (20 for microleakage evaluation, 3 for SEM evaluation, 1 only with the bonding agent) numbered from 1 to 4. Six specimens were used as controls. In three of these, the external surface, including the apex, was covered with two layers of nail varnish (negative control). The other three specimens were filled by lateral compaction of gutta-percha, without sealer (positive control).

In groups 1, 3, and 4, ethylene diamine tetracetic acid (EDTA) 15% solution (3 ml/3 min) was used for irrigation to remove the smear layer. All groups (1 to 4) were given a final irrigation with 5 ml distilled water. A #20 K-file was used to confirm the apical patency, and the root canals of all specimens were dried with paper points.

The tested dentin bonding system was used according to the manufacturer's instructions. In the specimens of groups 1 and 2, Nanobond was placed as follows: Using a cylindrical microbrush (Tepe; Malmö, Sweden), self-etching primer was applied into root canals. Any excess was removed with paper points. After 20 s, two drops of Nanobond adhesive resin were mixed with one drop of Dual Cure Activator and applied to the root canal with a microbrush. Any excess was removed with paper points. Finally, AH-26 sealer was placed in combination with gutta-percha cones and lateral compaction.

Teeth in group 3 were filled with AH-26 sealer in combination with gutta-percha cones.

In the specimens of group 4, Epiphany self-etching primer was placed in root canals with a microbrush and any excess removed with paper points. The roots were then filled with Epiphany sealer and Resilon points, using lateral compaction. The obturation with sealer (AH-26 or Epiphany) and cones (gutta-percha or Resilon) was performed by the same operator as follows. The master cone, corresponding to the master apical file (#35), was inserted into the root canal (at 14 mm) coated with sealer. Lateral compaction was performed with the successive use of finger spreaders sizes 25 and 20. Standardized cones (#20) were used as accessory points. Root filling was considered complete when it was not possible to place an accessory cone further than 2 mm into the root canal. Excess gutta-percha or Resilon was removed with a heated instrument

Table 1 Overview of the groups

Groups	Chemomechanical preparation	Obturation
1	Step-back technique (MAF #35) with NaOCl 5% as irrigant solution EDTA 15% to remove smear layer Distilled water Drying with paper points	Bonding agent: Nanobond Application of the primer with microbrush Excess removal with paper points Leave for 20 s Excess removal with paper points Mixing 2 drops of adhesive with one drop of activator and application with microbrush Excess removal with paper points Sealer: AH-26 Laterally compacted gutta-percha cones
2	Step-back technique (MAF #35) with NaOCl 5% as irrigant solution Distilled water Drying with paper points	Same as group 1
3	Same as group 1	Sealer: AH-26 Laterally compacted gutta-percha cones
4	Same as group 1	Bonding agent: Epiphany Application of the primer with microbrush Excess removal with paper points Sealer: Epiphany sealer Laterally compacted Resilon cones

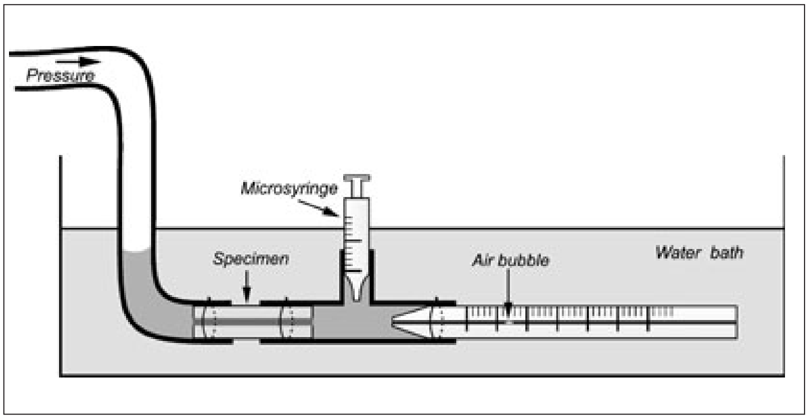

Fig 1 The fluid transport apparatus.

at the canal orifice and final vertical compaction was completed with a finger plugger. Excess sealer was removed with a cotton pellet. The specimens in group 4 were light cured for 40 s to create a coronal seal according to manufacturer's instructions. After obturation, all specimens, except for the 2 apical millimeters and the flat coronal surfaces, were coated with nail varnish. The experimental groups and procedures are summarized in Table 1.

Microleakage was evaluated by a fluid transport model as described by Wu et al[38] (Fig 1).

The root sections were connected to a plastic tube on either side of the specimen and kept in distilled water at 37°C for 7 days. The plastic tubes containing each root section were filled with distilled water. A standard glass capillary tube was connected to the plastic tube on the outlet side of the specimen. Using a syringe, water was retracted approximately 3 mm into the open end of the glass capillary. The whole setup was then placed in a water bath (20°C), and, using a syringe, the air bubble was adjusted to a suitable position within the capillary. A pressure of 0.2

Table 2 Results of measurement after 7 days (L = µl/24 h)

Group	L = 0	0 < L ≤ 10	10 < L ≤ 20	L > 20
1 [a]	12	4	0	4
2 [b]	4	1	2	13
3 [b]	5	2	0	13
4 [a]	14	1	1	4

There are no significant differences in groups with the same superscript letter.

Table 3 Results of measurement after 1 month (L=µl/24 h)

Group	L = 0	0 < L ≤ 10	10 < L ≤ 20	L > 20
1 [a]	12	4	0	4
2 [b]	4	1	2	13
3 [b]	5	2	0	13
4 [a]	14	1	1	4

There are no significant differences in groups with the same superscript letter.

Table 4 Results of measurement after 1 year (L=µl/24 h)

Group	L = 0	0 < L ≤ 10	10 < L ≤ 20	L > 20
1 [a]	6	2	2	10
2 [b]	0	1	2	17
3 [b]	1	0	2	17
4 [a]	6	2	2	10

There are no significant differences in groups with the same superscript letter.

atm was applied from the inlet side to force the water through the voids along the filling, thus displacing the air bubble in the capillary tube. The volume of the fluid transport was measured by visual inspection of the air bubble position by the same observer. The displacement of the air bubble was recorded as the fluid transport results (L), expressed in µl/24 h. Microleakage was measured at 7 days, 1 month, and 1 year. Between measurements, the specimens were kept in distilled water at 37°C.

The results were statistically analyzed by nonparametric Kruskal-Wallis and Mann-Whitney tests ($\alpha = 0.05$).

SEM Evaluation

Two specimens from each group and 4 specimens solely with bonding system (two with smear layer and two without) were prepared for examination by scanning electron microscope. After 7 days of storage in distilled water, cross sections of bonded interface were prepared using a microtome. The sections were polished with 1200-grit SiC papers under copious water irrigation, then exposed to 6 mol/L HCL for 30 s and 1% NaOCl for 10 min.[14] After carbon sputtering, the specimens were examined by SEM (JSM 840 A, JEOL; Tokyo, Japan).

RESULTS

In the case of the positive controls, the air bubbles moved rapidly along the tube as soon as the pressure was applied. No movement of the air bubble in the capillary tubes was observed in the negative controls. The microleakage in root sections was measured, as mentioned before, at 7 days, 1 month, and 1 year, and the results expressed in µl/24 h.

Short-term Results

At 7 days and 1 month (Tables 2 and 3), groups 1 (EDTA, Nanobond, AH-26) and 4 (EDTA, Epiphany, Resilon) leaked significantly less ($p < 0.05$) than groups 2 (Nanobond, AH-26) and 3 (EDTA, AH-26). No other significant differences were found between experimental groups.

Long-term Results

At 1 year (Table 4), groups 1 and 4 leaked significantly less than the other two groups ($p < 0.05$).

Short-term vs Long-term Results

The microleakage increased statistically significantly over time in group 4 ($p < 0.05$). In other groups, the microleakage values increased over time, but without statistically significant differences. Figure 2 summarizes the number of samples showing no leakage over time (qualitative data).

Scanning electron microscopy showed the formation of a hybrid layer with the self-etching adhesive Nanobond, without pretreatment with EDTA 15% (Fig 3A). When the teeth were treated with EDTA, tags of bonding resin were observed in the dentinal tubules (Figs 3B and 3C). In the specimens filled with gutta-percha, AH-26 sealer, and using Nanobond, tags were also observed in the dentinal tubules after smear layer removal by EDTA (Fig 3D).

DISCUSSION

The first null hypothesis of the present study was partially accepted, since the results showed that the sealing ability of AH 26 in combination with the self-etching bonding agent was similar to that of the Epiphany/Resilon only after pretreatment with EDTA. The second null hypothesis was rejected. All the materials tested showed an increase in apical leakage over time, but this increase was only statistically significant for the Epiphany/Resilon system.

The method used for the evaluation of the sealing ability of root canal sealers tested in this study was the fluid

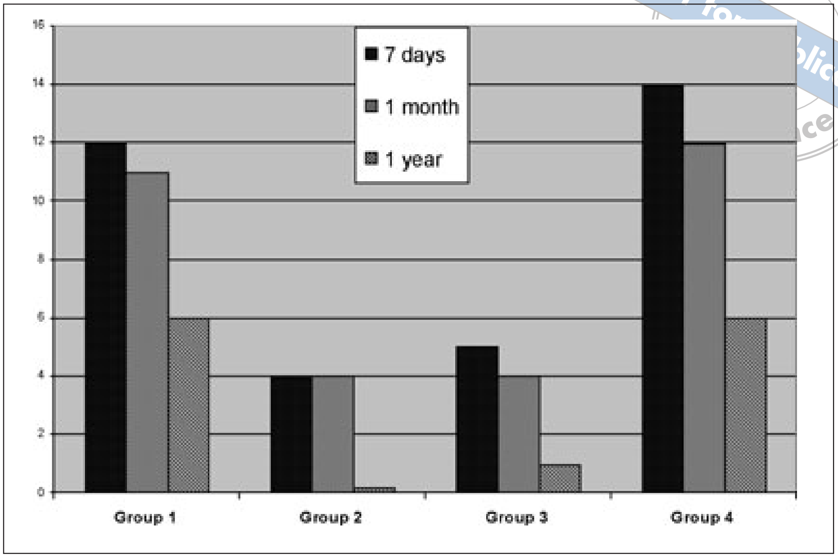

Fig 2 Number of samples with no leakage over time. Decrease of no leakage samples, after one year, was observed mainly in group 4.

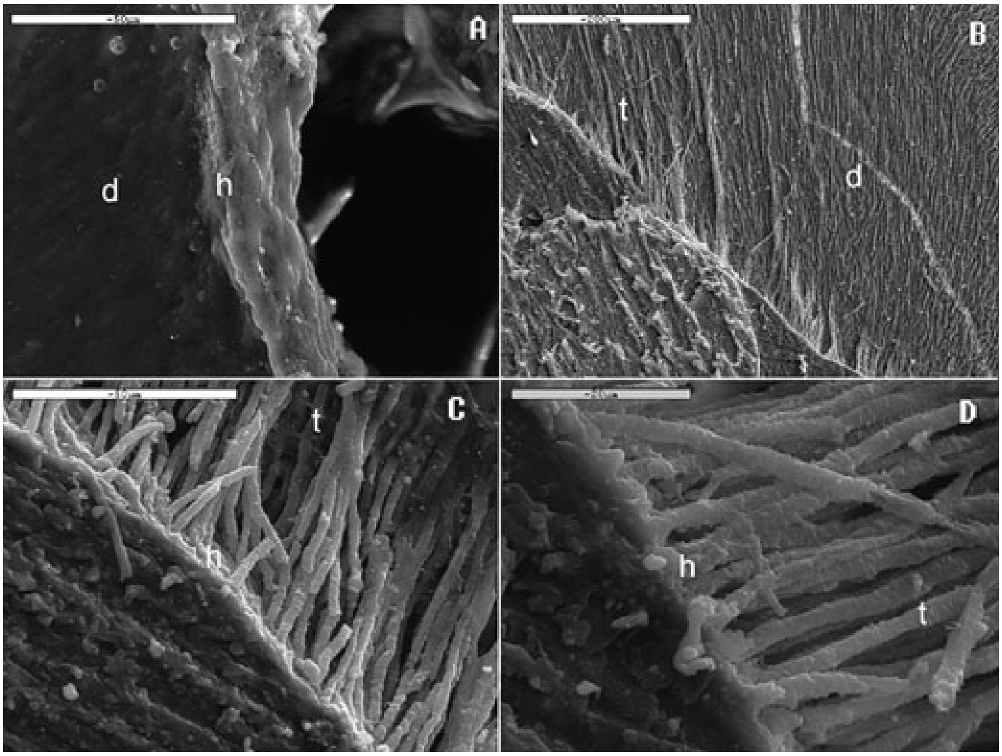

Fig 3A Nanobond without smear layer removal (1400X). d: dentin, h: hybrid layer.
Fig 3B Nanobond, AH-26 sealer and gutta-percha after smear layer removal (800X). d: dentin, t: resin tags.
Fig 3C Nanobond, AH-26 sealer and gutta-percha after smear layer removal (1000X). t: resin tags, h: hybrid layer.
Fig 3D Nanobond, AH-26 sealer and gutta-percha after smear layer removal (2000X). t: resin tags, h: hybrid layer.

transport model. Several methods have been used to evaluate the sealing ability of root canal sealers. The fluid transport model proposed by Wu et al[38] offers several advantages over more commonly used techniques of assessing leakage; it is both more sensitive than dye penetration for the detection of full-length voids along the root canal and highly reproducible. Since this method does not destroy the samples, it is possible to obtain measurements from the same specimens at intervals over extended periods.[37] However, the method requires careful manipulation of the connections between plastic tubes and glass capillaries to avoid erroneous measurements.

Combinations of bonding agents and resin sealers have been proposed for endodontic obturation. Several studies have investigated the effectiveness of these restorative root canal systems, with contradictory results.[1,3,7,10,21,26-29,34]

The reduced apical leakage observed in the present study using AH-26 in combination with the self-etching bonding agent and pretreatment with EDTA is comparable with the results of Mannocci and Ferrari.[10] They evaluated the apical leakage of fillings performed with AH-26, with and without the use of two etch-and-rinse adhesive systems. Their results showed that the use of etch-and-rinse dentin bonding agents significantly reduced apical microleakage. In another study[7] the shear bond strength of AH-26 root canal sealer to human root canal dentin exposed to different intracanal medications was compared, both with and without the use of three different bonding agents (Single Bond and Bond 1 etch-and-rinse, and Clearfill SE Bond self-etching). The results showed that using the bonding agents improved the shear bond strengths of AH-26 sealer to dentin.

On the other hand, Perdigao et al[21] recently examined the interfacial adaptation of an etch-and-rinse and a self-etching adhesive to root canal dentin. The authors reported that although hybrid layers were formed, interfacial gaps were observed with both adhesive systems they tested. They hypothesized that the complexity of the substrate and the increased shrinkage stress, due to the high C factor of the root canal, impedes bonding between adhesives and root canal dentin.

In the present study, the best results on apical leakage were obtained from the Epiphany-Resilon system and AH-26/gutta-percha/self-etching bonding agent plus pretreatment with EDTA 15%. Both systems showed statistically significantly better performance than the other groups examined. The self-etching adhesive system Nanobond was used in combination with AH-26 root canal sealer. Nanobond was selected because it is from the same manufacturer as Epiphany/Resilon and its primer has a similar composition to Epiphany primer (AMPS and hydrophilic monomers solution), apart from the phosphoric acid present in the former and the existence of adhesive resin.

Self-etching adhesive systems include primer containing an acidic monomer, obviating the need for an etch-and-rinse phase, which not only lessens clinical application time, but also significantly reduces technique sensitivity or the risk of errors during application and manipulation.[35] This is particularly convenient when used in root canals. The self-etching adhesive systems dissolve the smear layer and partially demineralize the underlying dentin surface. However, self-etching adhesives with high pH values demineralize dentin only to a depth of 1 μm,[35] and the thickness of the hybrid layer they produce is much smaller than produced by etch-and-rinse adhesives. This has been proved to be of minor importance with regard to actual bonding in the case of coronal dentin.[6,9]

Root dentin is a difficult substrate to bond to. Beside irregular structures such as accessory root canals, resorption, and repaired resorption areas, there are a lower number of dentinal tubules, irregular secondary dentin, and cementum-like tissue in the apical portion of the root canal wall.[12] Physiological phenomena such as tubular sclerosis and dentinal fluid transportation must also be considered in this technique-sensitive treatment approach.[4,18] Furthermore, the thick smear layer generated in root canal instrumentation may be difficult to dissolve and incorporate in the hybrid layer generated by a mildly acidic self-etching bonding agent. This might explain why the use of the EDTA in groups of AH-26/gutta-percha/Nanobond improved the sealing ability of the system.

The Epiphany-Resilon system combines Resilon obturating points, a resin-based sealer, and a self-etching primer to create a solid monoblock. Many studies have evaluated the sealing ability of this system. Bodrumlu and Tunga[3] compared the apical sealing ability of Epiphany-Resilon obturating material with AH-26 or AH plus sealer (in combination with gutta-percha) using the dye penetration method. Their results showed that the Epiphany-Resilon material had the least apical leakage. Similar results were obtained with the fluid transport method.[34] In the present study, the group in which the Epiphany-Resilon system was used also leaked significantly less than the group with AH-26 and gutta-percha, without the use of a dentin bonding system. Tay et al[29] compared the ultrastructural quality of the apical seal achieved with Resilon-Epiphany and gutta-percha/AH Plus using scanning electron microscopy and transmission electron microscopy. They concluded that a complete hermetic seal cannot be achieved with either material. Concerns have been raised regarding the alkaline hydrolysis susceptibility of Resilon points as compared to gutta-percha.[32] Hiraishi et al[8] also reported observing gaps between the points and the sealer in the Resilon group. This physical separation was not expected in view of the philosophy behind the monoblock concept. They also reported that the bond strengths of Resilon to a methacrylate based resin, as found in Resilon sealer, are 4 to 5 times lower than the bond strengths of a composite resin to that sealer. Shemesh et al[26] compared the sealing ability of the Epiphany-Resilon system with AH-26 and gutta-percha, using two different leakage models. With the fluid transport model, no statistically significant differences were found, while with the glucose penetration model, Resilon showed greater leakage throughout the experimental period.

Results of the same groups at three experimental periods in this study showed increasing microleakage values over time, although differences were statistically significant only with the Epiphany-Resilon system ($p < 0.05$). The increased microleakage over time observed in the Epiphany-Resilon group is in agreement with the findings of Paque and Sirtes,[19] who reported a dramatic increase in fluid movement with specimens filled with Epiphany/Resilon after 16 months. Little is known about the long-term effects of incorporating dissolved hydroxyapatite crystals and residual smear layer remnants within the bond in self-etching adhesives.[35] Moreover, the surplus of primer/adhesive solvent retained within the interface structure could directly weaken the bond integrity and provide channels for nanoleakage, or may affect polymerization of the infiltrated monomers. The interfacial structure also becomes more hydrophilic and thus more prone to hydrolytic degradation.[31,33] Papa et al[17] reported that teeth with vital pulps and root-filled counterparts do not differ significantly in their moisture content. In addition, after rinsing the root canal, fluid droplets are retained in the dentinal tubules and may not be removed through the use of paper points. As previously mentioned, there is the additional problem that chemical degradation of the hybrid layer in root canal dentin might occur by enzymes released by bacteria and by dentin itself. Tay et al[30] reported that self-etching adhesives with high pH values (called "mild") activate latent matrix metalloproteinases without denaturing these enzymes, and may thus adversely affect the longevity of bonded root canal fillings. The fact that the Epiphany/Resilon group showed a statistically significant increase in apical leakage over time could be attributed to the degradation of the bond in combination with the hydrolysis of Resilon, while the expansion of gutta-percha over time in the other groups may compensate for the leakage occurring through sealer dissolution or bond degradation.[39]

The results of the present study suggest that the permeation of smear layer by bonding agents does not lead to a seal as satisfactory as that obtained by the removal of the smear layer with EDTA. The elimination of the smear layer facilitates better infiltration of resin and sealer into the dentinal tubules, which may be responsible for the better seal obtained. In fact, scanning electron microscopy evaluation showed that when the smear layer has been removed by EDTA, tags of the bonding resin were observed in the dentinal tubules.

The use of EDTA solution for removing the smear layer is accepted practice for root canal treatment. The etch-and-rinse approach to adhesion, making use of a strong acid (usually phosphoric acid 32%) to dissolve the mineral phase of dentin, also results in a recession of the collagen matrix, raising the possibility of interference with the formation of a hybrid layer.[11] EDTA solution for endodontic use is less acidic than a phosphoric acid-based etchant, which is potentially more toxic to the periapical tissues. Osorio et al[16] concluded that the collagen network is better preserved after EDTA demineralization than after phosphoric acid etching, leading to the possibility of improving the bond durability of adhesives. The results of the current study suggest that using bonding agents in root canal obturation does not require any variation in the normal protocol for removing the smear layer. Removal of the smear layer allows bonding system and sealer to adhere more effectively to root canal dentin.

Further investigation is needed into the many parameters related to clinical practice, such as interactions with intracanal medicaments, difficulties in placing, biocompatility, etc.

CONCLUSION

The sealing ability of AH-26 in combination with gutta-percha was improved using a self-etching bonding agent and pretreatment with EDTA 15%.

The Epiphany-Resilon obturating system showed a similar sealing ability to AH-26 sealer and gutta-percha with the application of a self-etching adhesive and pretreatment with EDTA 15%.

All the obturating systems examined showed increased microleakage over time. A statistically significant difference was observed only with the Epiphany-Resilon system after 1 year of storage.

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Clinical relevance: The use of a self-etching bonding system can improve the sealing ability of AH-26. The sealing ability of the Resilon-Epiphany system decreased over time.