Sealer Penetration and Adaptation in the Dentinal Tubules: A Scanning Electron Microscopic Study

Eric Balguerie, DDS, Lucas van der Sluis, DDS, PhD, Karen Vallaeys, DDS, MSc, Marie Gurgel-Georgelin, DDS, MSC, and Franck Diemer, DDS, MSc, PhD

Abstract

Introduction: Tubular penetration and adaptation of the sealer can determine the sealability of the root filling. The aim of this study was to assess, *in vitro*, the tubular adaptation and penetration depth and the adaptation to the root canal walls in the apical, middle, and coronal third of the root canal of 5 different sealers used in combination with softened gutta-percha cones. **Methods:** Fifty-two single-rooted teeth were prepared and filled with 5 different sealers and softened gutta-percha cones. Thereafter, the roots were cross-sectioned and prepared for scanning electron microscopic evaluation. Adaptation of the sealer to the root canal and tubular walls and tubular penetration were assessed. **Results:** AH Plus (Dentsply De Trey, Konstanz, Germany), an epoxy resin sealer, showed the best tubular adaptation and penetration. **Conclusions:** The tubular penetration and adaptation varies with the different physical and chemical properties of the sealers used. AH Plus showed the most optimal tubular penetration and adaptation to the root canal wall of the sealers tested. *(J Endod 2011;37:1576–1579)*

Key Words

Adaptation, Herofill, root canal wall, sealers, tubular penetration

From the Department of Conservative Dentistry and Endodontics, Paul Sabatier University, Toulouse, France.

Address request for reprints to Dr Eric Balguerie, Université Paul Sabatier, Département d'Odontologie Conservatrice Endodontie, Faculté de Chirurgie Dentaire, 3 chemin des Maraîchers, 31062 Toulouse, Cedex 09 France. E-mail address: ericbalguerie@hotmail.fr

0099-2399/$ - see front matter

doi:10.1016/j.joen.2011.07.005

The main objective of a root canal filling is to seal the root canal system to prevent reinfection (1). Normally, a root canal filling is associated with a hard core, like gutta-percha, and a sealer to better adapt the root canal filling material and complete the seal of the root canal filling in the most effectual manner (2). Therefore, the sealer-root canal wall interface is crucial for the sealing of the root canal system (3–5). The sealer can fill the irregularities of the root canal wall and the dentinal tubules, which cannot be filled by gutta-percha.

Sealer penetration into the tubules could affect the seal of the root filling because an increase of the contact surface between filling material and dentin is related to an improvement of the sealability (6). Also, sealer penetration can promote an antimicrobial effect in the tubules, which increases when in closer contact with the microbes (1, 7). Furthermore, sealer penetration in *in vitro* models is comparable to *in vivo* (8, 9). Therefore, the aim of this study was to assess *in vitro* the tubular penetration depth in the apical, middle, and coronal third of 5 different sealers and their adaptation to the walls of the root canal and dentinal tubules in combination with a softened gutta-percha cone.

Materials and Methods

Fifty-two freshly extracted single-rooted permanent human teeth of similar size and root shape were selected and stored in a 3% formaldehyde solution. Therefore, this study was conducted in accordance with the ethical guidelines for medical research at the University of Toulouse and the Declaration of Helsinki. All the teeth had intact roots. Teeth with radicular resorption, immature apex, fracture, or an endodontic filling were rejected. The root surface was cleaned by an ultrasonic device (P-Max; Acteon, Merignac, France). The teeth were fixed on a Synea TA97 turbine (W&H, Eckbolsheim, France), and the crowns were sectioned at the cementoenamel junction with a parallel chamfer diamond bur 014 (Stoner, Brent, Switzerland) mounted on a turbine under continuous water spray.

The working length (WL) is determined to be short 0.7 mm of the point where the K-file size 10 was first visible with a binocular at $\times 16$ magnification. The root canals were prepared with nickel-titanium rotary files (HeroShaper; Micro-Mega, Besançon, France) until file size 30 taper 0.04 at the WL. Between each instrument, the canal was irrigated with 2 mL 3% sodium-hypochlorite (NaOCl) solution with a syringe and a 27-G side-vented needle (Endoneedle G27; Elsodent, Cergy-Pontoise, France). At the end of the preparation, the canal was irrigated with 3 mL 15% EDTA (Largal Ultra; Septodont, Saint Maur, France) for 3 minutes (10–12). The final rinse was performed with 5 mL 3% NaOCl (10–12). Then, the canal was dried with a sterile paper point size 30, taper 0.02 (Pierre Rolland, Merignac, France).

The specimens were randomly divided into 5 groups (n = 10) and filled with 5 different sealers (Table 1). The sealers are mixed according to the manufacturers' recommendations. The sealers were placed in the root canals with a K-file size 15 (Micro-Mega) placed at the WL in a counter-clockwise motion. Thereafter, a hot gutta-percha carrier HeroFill n°30 (Micro-Mega) was placed in the root canal following the recommendations of the manufacturer. Two teeth were not filled and served as the control group to verify the absence of smear layer on the dentinal walls. The coronal part of the root canal was filled with Cavit (3M ESPE, Seefeld, Germany). The samples were then stored at 37°C and 100% humidity for 2 weeks, allowing complete setting of the sealers (13).

The roots were embedded in resin (Synolite 0328-A1; Gaches Chimie, Toulouse, France). Thereafter, the samples were transversely sectioned at 2, 5, and 8 mm from the WL with a diamond wire saw (Well 3241; Well Dental Laboratory Company, Hong Kong, Hong Kong). To remove the (in)organic debris, the specimens were cleaned in a bath with 15% EDTA for 2 minutes and then 3% NaOCl for 2 minutes. The samples were dehydrated in an evaporator (Boc Edwards, West Sussex, UK) for 4 hours.

The specimens were mounted on a tub and cold sputtered (Sputter Coater S-150-B, Boc Edwards). The specimens were observed in a scanning electron microscope (JEOL JSM–840A; JEOL, Tokyo, Japan), and of each specimen one photomicrograph most representative of the section was taken from root-sealer interface at a magnification between 500× and 1,500×. On each of these photomicrographs, the minimum and maximum depth of sealer penetration in the tubules was measured.

Mean scores per tooth and tooth level were used for further calculations and comparisons using analysis of variance and Fisher's protected least significant difference (PLSD) (Statview 5.0.1; SAS Institute Inc, Cary, NC). The Fisher's PLSD test was used to compare the penetration depth between the different sealers and the 3 levels. The P value was set at .05.

The sealer adaptation to the root canal wall or the tubules was described as follows:

1. *Good*: The majority of the sections showed no gaps between the sealer and dentin.
2. *Reasonable*: The majority of the sections showed some small gaps (<1 μm) between the sealer and dentin.
3. *Poor*: The majority of the sections showed many gaps (between 1 and 10 μm) between the sealer and dentin.
4. *No adaptation*: The majority of the sections showed no adaptation between the sealer and dentin (gaps >10 μm).

All the images were analyzed by 2 calibrated independent observers who were both blinded. The percentage of interagreement should be more than 95%; if this percentage was lower than 95%, a consensus should be reached.

Results

The results are shown in Tables 1 and 2 and Figure 1. For the image analysis of the sealer adaptation, the interobserver agreement was high (kappa coefficient = 0.91). In case of a disagreement, an agreement was reached after discussion.

Discussion

AH Plus scores the best for adaptation to the root canal wall, tubular penetration, and adaptation to the peritubular dentine directly followed by Acroseal (Septodont, Saint-Maur, France). Both are epoxy resin sealers. The findings for AH Plus are supported by other studies (14, 15). Epoxy resins sealers like AH Plus are also correlated with a higher adhesion to dentin and gutta-percha (16). The good penetration, adaptation, and adhesion properties will have 2 positive effects, in the first place on sealing because of the increased surface contact between sealer and dentin (6) and second on the antimicrobial effect by locking the residual microorganism in the dentinal tubules (17, 18).

The sealer penetration depth in the dentinal tubules depends on many factors like smear layer removal (19), dentinal permeability (the number and the diameter of tubules), root canal dimension, and the physical and chemical properties of the sealer (20–22). The flow is one of the main chemical/physical factors to influence the

TABLE 1. Adaptation to the Root Canal Wall and Tubules, Structure, and Mean Tubular Penetration of the 5 Sealers

Groups	Sealer type	Sealer name	Adaptation root canal	Adaptation tubules	Sealer structure	Tubular penetration: minimal/mean/maximal (μm)		
						WL-8 mm	WL-5mm	WL-2mm
1	Calcium hydroxide epoxy resin	Acroseal	Good	Good	Homogeneous	23/90.3/157	16/84.3/240	0/0/0
2	Zinc oxide eugenol	Endobtur	Reasonable	Reasonable	Homogeneous	12/40.2/153	8/37.6/115	0/0/0
3	Glass ionomer	Ketac-Endo	Poor	No adaptation	Not homogeneous	0/0/0	0/0/0	0/0/0
4	Epoxy resin	AH Plus	Good	Good	Homogeneous	14/62.5/122	16/53.3/132	0/22.3/36
5	Silicon	RSA	Reasonable	Poor	Granular (wall tubules)	13/22.7/28	12/23.4/40	0/0/0

TABLE 2. The Fisher's PLSD Test Shows the Differences in Tubular Penetration in the Different Sections

	Apical	Middle	Coronal
Endobtur vs AH Plus	S	NS	S
Endobtur vs RSA	/	NS	NS
Endobtur vs Acroseal	/	NS	S
Endobtur vs Kétac-Endo	/	NS	NS
AH Plus vs RSA	S	NS	S
AH Plus vs Acroseal	S	NS	NS
AH Plus vs Kétac-Endo	S	S	S
RSA vs Acroseal	/	S	S
RSA vs Kétac-Endo	/	NS	NS
Acroseal vs Kétac-Endo	/	S	S

/, no tubular penetration; NS, not significant; S, significant

tubular penetration and is defined as the ability of a sealer to penetrate in irregularities, lateral canals, or dentinal tubules of the root canal system (23). The flow is determined by the consistency, particle size, shear rate, temperature, time, internal diameter of the root canal, and the rate of insertion (22). Of these factors, the particle size and consistency of the sealers were typical for the sealers used and therefore not standardized during the experiments. Other studies have shown that the flow of AH 26 (Dentsply De Trey) and AH Plus is significantly higher than other sealers tested (17, 23).

The observation of the interface sealer/dentin by scanning electron microscopy can be done with longitudinal or cross-sectional sections. The direction of the tubules is mostly perpendicular to the root canal wall. The chance to obtain a section longitudinal of the tubule is even for both cutting directions (24). In most studies, longitudinal sections are made, especially when the coronal or middle part of the root canal is evaluated or when the location of the evaluation is not mentioned (8, 19, 25, 26). However, for thin or curved roots, this could create problems in the apical root canal; therefore, we used cross-sectional sections.

After the cutting of the roots, the specimens were cleaned in a bath filled with EDTA 15% and NaOCl 3% to remove the smear layer produced by the sectioning, notwithstanding a continuous flow of water on the diamond wire saw. This procedure could influence the sealer adaptation in the tubules facing the cutting surface and the dimension of the tubule opening. However, this should be the same for all the groups.

The penetration in the dentinal tubules was significantly greater in the coronal and middle of the root canal than the apical part of the root canal ($P < 0.0416$) except for Endobtur (Septodont, $P > 0.0554$). This is supported by other studies (21, 27). This could be because of the fact that the apical root canal contains less tubules, and when present, the diameter is smaller or they are more often closed (8, 28, 29). Furthermore, the apical portion of roots shows a pronounced variation in structure (29). For example, primary dentinal tubules

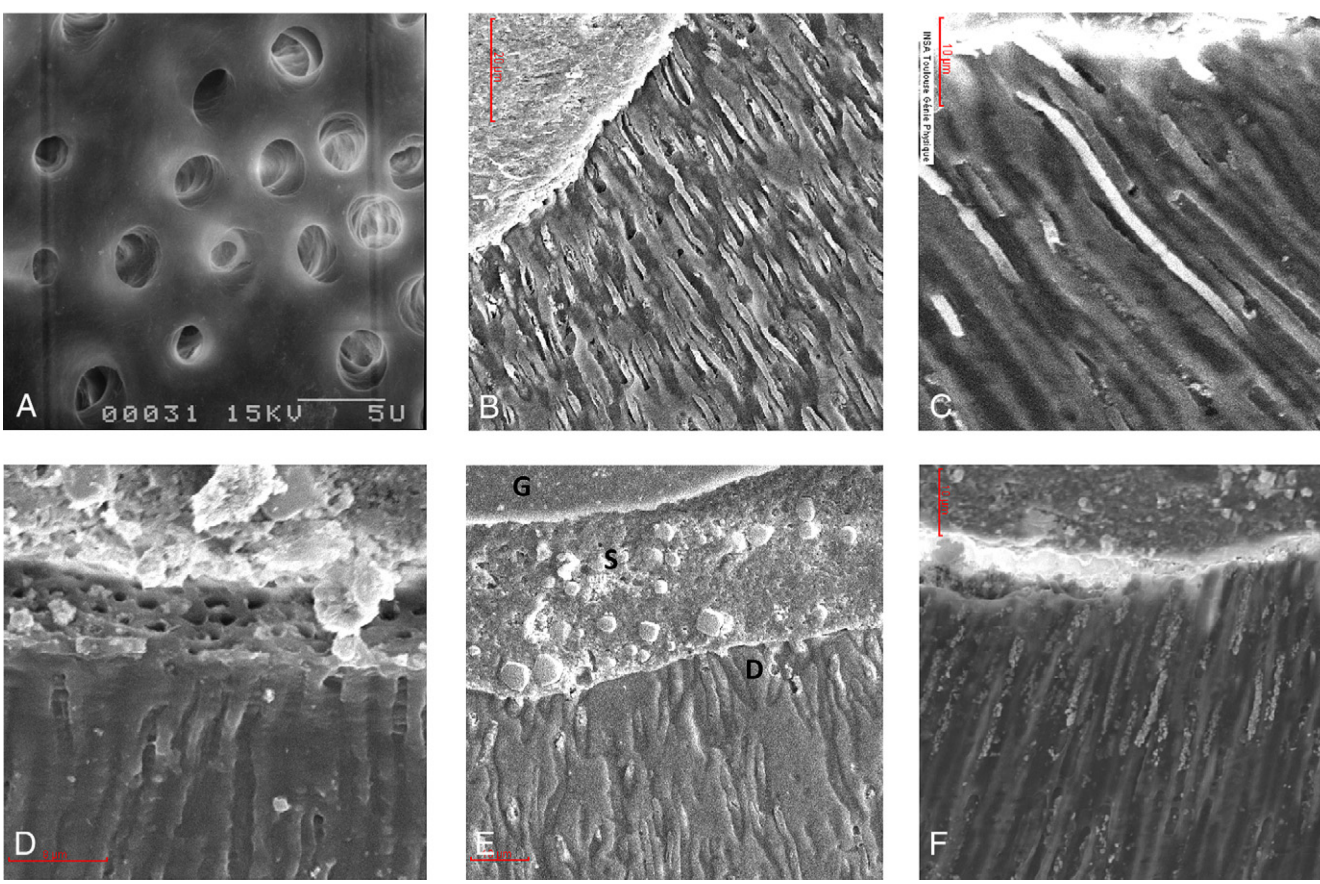

Figure 1. The scanning electron microscopic images show the interface of the dentin with different sealers. (*A*) The control group in the coronal part; the tubules are open, and their mean diameter is 3 μm (3,800×). (*B*) The interface of Acroseal in the middle part of the root canal wall. The tubules are filled with sealer (750×). (*C*) The interface of Endobtur and the root canal wall in the coronal third (1,300×). (*D*) The structure of Ketac-Endo (3M ESPE, Seefeld, Germany) is heterogenic and does not penetrate in the tubules. Gaps between the filling material and the root canal wall are frequently seen (1,800×). (*E*) AH Plus is very well adapted to the root canal wall and peritubular dentin. No gaps are seen between the gutta-percha (G) and sealer (S) or sealer (D) and root canal wall (850×). (*F*) The structure of RSA was granular. It is unable to completely fill the dentinal tubules (1,000×).

are irregular in direction and density; some areas are devoid of tubules. Also, cementum-like tissue can line the apical root canal wall, occluding any tubules. In this study, none of the sealers penetrated in the apical root canal with the exception of 2 samples from the AH group.

In this study, the results for RSA (Roeko Dental Product, Langenau, Germany) were worse than in another study (27), which can be related to the filling technique used. We used a warm technique, and the manufacturer advises to use cold filling techniques in combination with RSA. An increase in temperature can cause clustering of the sealer and could explain less tubular penetration and the granular aspect (30).

Conclusion

The tubular penetration depth varies with the different physical and chemical properties of the sealers used. AH Plus showed the most optimal tubular penetration and adaption to the root canal wall of the sealers tested. The sealer penetration depth in the apical root canal is less because of the different properties of the apical root canal.

Acknowledgments

The authors deny any conflicts of interest related to this study.

References

1. Sjögren U, Figdor D, Persson S, Sundqvist G. Influence of infection at the time of root filling on the outcome of endodontic treatment of teeth with apical periodontitis. Int Endod J 1997;30:297–306.
2. Evans JT, Simon JHS. Evaluation of the apical seal produced by injected thermoplasticized gutta-percha in the absence of smear layer and root canal sealer. J Endod 1986;12:101–7.
3. Wu MK, Özok AR, Wesselink PR. Sealer distribution in root canals obturated by three techniques. Int Endod J 2000;33:340–5.
4. Wu MK, Fan B, Wesselink PR. Diminished leakage along root canals filled with gutta percha without sealer over time: a laboratory study. Int Endod J 2000;33:121–5.
5. Hata G, Kawazoe S, Toda T, Weine F. Sealing ability of Thermafill with and without sealer. J Endod 1992;18:322–6.
6. Wu MK, De Gee AJ, Wesselink PR. Effect of tubule orientation in the cavity wall on the seal of dental filling materials: an in vitro study. Int Endod J 1998;31:326–32.
7. Heling I, Chandler NP. The antimicrobial effect within dentinal tubules of four canal sealers. J Endod 1996;22:257–9.
8. Vassiliadis LP, Sklavounos SA, Stavrianos CK. Depth of penetration appearance of Grossman sealer in the dentinal tubules: an in vivo study. J Endod 1994;20:373–6.
9. Mamootil K, Messer HH. Penetration of dentinal tubules by endodontic sealer cements in extracted teeth and in vivo. Int Endod J 2007;40:873–81.
10. Niu W, Yoshioika T, Kobayashi C, Suda H. A scanning electron microscopic study of dentinal erosion by final irrigation with EDTA and NaOCl solutions. Int Endod J 2002;35:934–9.
11. Teixeira CS, Felippe MCS, Felippe WT. The effect of application time of EDTA and NaOcl on intracanal smear layer removal: an SEM analysis. Int Endod J 2005;38: 285–90.
12. Yamada RS, Armas A, Goldman M, Sun Lin P. A scanning electron microscopic comparison of a high volume final flush with several irrigating solutions: part 3. J Endod 1983;9:137–42.
13. Allan NA, Walton RE, Schaffer M. Setting times for endodontic sealers under clinical usage and in-vitro conditions. J Endod 2001;6:421–3.
14. Sen BH, Piskin B, Baran N. The effect of tubular penetration of root canal sealers on dye microleakage. Int Endod J 1996;29:23–8.
15. Kokkas A, Boutsioukis A, Vassiliadis L, Stavrianos CK. Stereomicroscopique and scanning electron microscopic study of roots obturated with vertically condensed gutta-percha, epoxy resin cement, and dentin bonding agent. J Endod 2004;30: 100–2.
16. Lee KW, Williams MC, Camps JJ, Pashley DH. Adhesion of endodontic sealers to dentin and gutta-percha. J Endod 2002;28:684–8.
17. Siqueira JF, Favieri A, Gahyva SMM, Moraes SR, Lima KC, Lopes HP. Antimicrobial activity of flow rate of newer and established root canal sealers. J Endod 2000;28: 274–7.
18. Peters LB, Van Winkelhoff AJ, Buijs JF, Wesselink PR. Effects of instrumentation, irrigation and dressing with calcium hydroxide on infection in pulpless teeth with periapical bone lesions. Int Endod J 2002;35:13–21.
19. De Deus GA, Gurgel-Filho ED, Maniglia-Ferreira C, Coutinho-Filho T. Intratubular penetration of root canal sealers. Pesq Odonto Bras 2002;16:332–6.
20. De Deus GA, Gurgel-Filho ED, Maniglia-Ferreira C, Coutinho-Filho T. The influence of filling technique on depth of tubule penetration by root canal sealer: a study using light microscopy and digital image processing. Aust Endod J 2004;30:23–8.
21. Weis MV, Parashos P, Messer HH. Effect of obturation technique on sealer cement thickness and dentinal tubule penetration. Int Endod J 2004;37:653–63.
22. Ørstavik D. Materials used for root canal obturation: technical, biological and clinical testing. Endod Topics 2005;12:25–38.
23. Bernardes AR, de Amorin Campelo A, Silva S, et al. Evaluation of the flow rate of 3 endodontic sealers: sealer 26, AH Plus and MTA obtura. Oral Surg Oral Med Oral Pathol OralRadiol Endod 2010;109:47–9.
24. Kaqueler JC, Decombas M, Le May O. Structures bucco-dentaires en microscopie électronique à balayage. Masson: Paris; 1989:39–76.
25. Calt S, Serper A. Dentinal tubule penetration of root canal sealers after root canal dressing with calcium hydroxide. J Endod 1999;25:431–3.
26. Katoaka H, Yoshioka T, Suda H, Imai Y. Dentin bonding and sealing ability of a new root canal sealer. J Endod 2000;26:230–5.
27. Sevimay S, Dalat D. Evaluation of penetration and adaptation of three different sealers: a SEM study. J Oral Rehab 2003;30:951–5.
28. Carrigan PJ, Morse DR, Furst ML, Sinai IH. A scanning electron microscopic evaluation of human dentinal tubules according to age and location. J Endod 1984; 10:359–63.
29. Mjör IA, Smith MR, Ferrari M, Mannoci F. The structure of dentine in the apical region of human teeth. Int Endod J 2001;34:346–53.
30. Wu MK, van der Sluis LW, Ardila CN, Wesselink PR. Fluid movement around the coronal two-third of root fillings placed by three different gutta-percha techniques. Int Endod 2003;36:533–40.