

EFFECT OF MOISTURE ON THE ROOT TREATED CANALS' MICROLEAKAGE USING TWO TYPES OF RESIN BASED SEALERS

Razavian H,¹ Hanjani K²

1. Dental Materials Research Center, Departments of Endodontics, School of Dentistry, Isfahan University of Medical Sciences, Isfahan, Iran.

2. Dental Student, Isfahan University of Medical Sciences, School of Dentistry, Isfahan, Iran.

ABSTRACT

Aim: To estimate the stature of an individual using both clinical and radiological methods and to evaluate the most accurate method in estimating stature of an individual.

Materials & Method: 36 human canines were extracted and decoronated to achieve a 15 mm root length and were assigned to 4 groups randomly. All teeth were instrumented using the step-back technique and 2.5% sodium hypochlorite was used as an irrigation solution in this protocol.

The groups were organized to have 18 teeth for each sealer, half of which belonged to the group of moistened teeth and the other half to the group of the dried teeth. In this study, AH-plus and AH26 (Dentsply/Middle East and Africa) were used as two common resin sealers.

To evaluate the microleakage, the bacterial evaluation system was used. The roots were placed in 36 end-removed micropipettes, sealed and sterilized with ethylene oxide gas.

One ml of the solution containing *E. faecalis* (ATCC 29212) (the most prevalent and strongest bacteria in endodontics with the minimal size) was injected from the upper part of the system every three days for 90 days and microleakage was evaluated daily by observing turbidity in the BHI solution.

Results: The results showed microleakage in all the samples over 40 days and the survival test analysis demonstrated different times of microleakage in the groups. The longest mean time of microleakage was related to group 2 (AH26 dry) and the shortest mean time was related to group 3 (AH-plus wet).

Conclusion: The results of this experimental study showed that among the four groups of the study, the highest resistance to microleakage belonged to group 2 (AH26 dry) and the least resistance belonged to group 3 (AH-plus wet). This difference in resistance was not significant between other group.

Key words: *Microleakage, Moisture, Obturation, Resin sealers.*

Introduction

The main goal of root canal therapy (RCT) is to save the tooth and its function in the oral cavity. Root canal treatment is achieved through caries removal, access cavity preparation, debridement, shaping and obturation. The process would be completed by appropriate coronal restoration.

One critical factor is to seal the canal perfectly to prevent microleakage or any other harmful chemicals from passing through. To achieve this goal, sealers should be used.¹

One of the most common sealers used in endodontists is resin sealers which have been used for many years. The sealing ability of these sealers has been compared in different studies so far. However, the interaction between these resin sealers and dentin in the presence of moisture has not been evaluated properly.

In order to decrease microleakage, a sealer needs to penetrate into the dentinal tubules. What is important about resin sealers is that they form a hybrid layer by being bonded to the demineralized dentin. Now here, the moisture left in dentin is a source of concern.²⁻⁴

The root canal is ready to be filled when it is completely debrided and dried. However, in some cases in spite of all our efforts, there would be some moist left in the root canal. This might be the result of delayed bleeding, interstitial fluid or irrigation solution remnants, particularly sodium hypochlorite.⁵ These all have definitely happened

out of sight of the operator, interfering with the sealing ability of the sealers.^{6,7}

In this situation, retreatment may not be practical; therefore, complete elimination of moisture from the root canal would not be possible.⁸

However, the question is whether this negligible amount of moisture can affect the sealing ability or not.¹

Some experiments have shown that moisture leads to less microleakage, which also depends on the sealer type, irrigants and irrigation methods.^{9,10}

Some other studies have claimed, based on the amount of dye penetration, that the side effects of the remaining moisture in the root canal occur only when the sealer is initially placed, with no effect on the sealing ability over time.¹¹⁻¹⁷

In addition, according to some in vitro analyses, there have been no significant differences in microleakage between moistened root canals and the dry ones.¹³

Given all these contradictory documents mentioned above, the point is to determine what the real influence of moisture is on the microleakage of obturated root canals, using resin sealers.^{4,18-20}

The aim of this study was to assess the effect of moisture on two resin sealers, AH-plus and AH26, in obturated root canals.

Materials & Method

In this experimental study, 36 human canines extracted within one month were used. Initially, the teeth were debrided and all the calculi and remaining PDL tissues were removed. The samples were kept in normal saline solution at 25°C. The teeth were assigned to 4 groups (n=9). The tooth crowns were removed at CEJ using a water-cooled low-speed diamond disk to achieve a 15-mm root length.

Following apical patency verification using a #10 K-file, the teeth were instrumented using the step-back technique and shaped to file #80, establishing a #40 master apical cone for all the samples. In this protocol, 2.5% sodium hypochlorite was used as an irrigation solution.

The smear layer was removed by irrigating the root canals with 5.25% hypochlorite for 1 minute and then 17% EDTA for 1 minute and preferably normal saline in between, to prevent any unpredictable interaction.

The groups were organized to have 18 teeth for each sealer, half of which belonged to the group of moistened teeth and the other half to the group of the dried teeth. In this study, AH-plus and AH26 (Dentsply/Middle East and Africa) were used as two common resin sealers.

To prepare the dry group, the canals were flushed with normal saline solution to remove the smear layer after being irrigated by 25% NaOCl and 17% EDTA and after that, dried by using paper points with similar size and length as the master apical file until the moisture was eliminated thoroughly.

In the moistened group, however, the canals were not dried completely after irrigation with NaOCl, as only the moisture of the pulp chamber and the coronal two-thirds of the canal would be displaced using a paper point with greater size than the master apical file.

Next, in each group, the sealers were mixed according to manufacturers' instructions and inserted into the root canals coating all the walls by a spreader with the size and length of the master apical cone.

The root canals were filled and obturated using Dia-Dent (Korea) gutta-percha cones, applying cold lateral condensation.

After radiography procedure to make sure of proper obturation, the teeth were incubated at 37°C and 100% humidity for 7 days for the sealers to set.^{4,21}

Bacterial microleakage evaluation

The bacterial microleakage evaluation system in this study was the same as the system used in a study by Lima *et al.*²²⁻²⁶ The roots were placed in 36 end-removed micropipettes and the interface between the teeth and micropipettes was sealed with two layers of cyanoacrylate glue and one layer of varnish.

Before mounting the roots, except for the coronal two mm and the apical two mm, the rest of the root surfaces were

covered with two layers of nail polish to eliminate the effect of lateral canals. Then, to sterilize the system with ethylene oxide gas, it was transferred to anti-serum vials for 24 hours.²⁷

To make sure of the sterilization procedure, the system was transferred to anti-serum vials containing 10 mL of BHI (brain heart infusion) (Merck, Darmstadt, Germany) under aseptic conditions. The samples were kept in an incubator for 3 days. In case of any turbidity which indicated the presence of bacteria, the sample was retrieved and replaced. One mL of the solution containing *E. faecalis* (ATCC 29212) (the most prevalent and strongest bacteria in endodontics with the minimal size) was injected from the upper part of the system every three days and microleakage was evaluated daily by observing turbidity in the BHI solution.

The samples were checked daily for 90 days and the ones with microleakage were recorded and excluded from the study.

Data collected from the two resin sealers in dry and moistened groups were analyzed with survival test ($p < 0.05$).²⁸

Results

The results of the present study showed microleakage in all the samples of the four different groups over 40 days.

The survival test analysis demonstrated different times of microleakage in the groups as displayed in Table 1 and Diagram 1.

	Group	AH26 wet		AH26 dry		AH-plus wet		AH-plus dry	
		Chi-squared	Sig.	Chi-squared	Sig.	Chi-squared	Sig.	Chi-squared	Sig.
Log Rank (Mantel-Cox)	1. AH26 wet			.407	.524	.570	.450	.004	.951
	2. AH26 dry	.407	.524			4.739	.029	.125	.724
	3. AH-plus wet	.570	.450	4.739	.029			1.703	.192
	4. AH-plus dry	.004	.951	.125	.724	1.703	.192		

Table 1. Means and medians for survival time

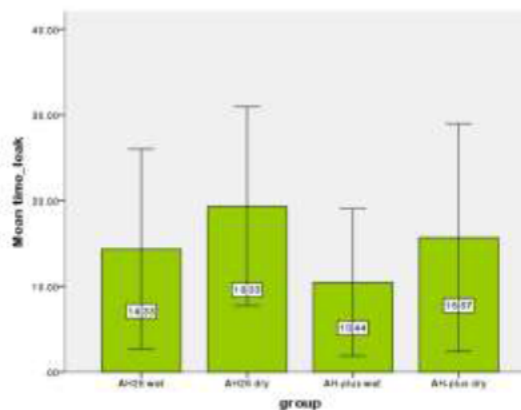


Diagram 1: Mean time-leak of the 4 groups

According to the Table, the longest mean time of microleakage was related to group 2 (AH26 dry) and the shortest mean time was related to group 3 (AH-plus wet).

The main point of this study was that there was a significant difference between groups 2 (AH26 dry) and 3 (AH-plus wet), with no significant differences between the other groups. [Table 2]

	Group	AH26 wet		AH26 dry		AH-plus wet		AH-plus dry	
		Chi-squared	Sig.	Chi-squared	Sig.	Chi-squared	Sig.	Chi-squared	Sig.
Log Rank (Mantel-Cox)	1. AH26 wet			.407	.524	.570	.450	.004	.951
	2. AH26 dry	.407	.524			4.739	.029	.125	.724
	3. AH-plus wet	.570	.450	4.739	.029			1.703	.192
	4. AH-plus dry	.004	.951	.125	.724	1.703	.192		

Table 2. Pair-wise comparisons

As indicated in Diagram 2 (survival functions), over a period of 40 days, the four groups showed microleakage in the following succession:

- 1) (AH-plus wet)
- 2) (AH26 wet)
- 3) (AH-plus dry)
- 4) (AH26 dry)

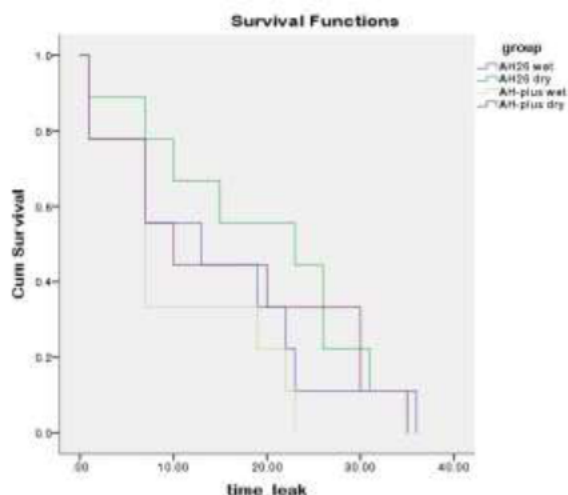


Diagram2: Cum survival over time-leak.

In this diagram, we can also inspect that the most resistant group to micro-leakage is the group (AH26 dry) and the least resistance relates to the group (AH-plus wet).

Discussion

Root canal therapy success depends on the presence of the microorganisms in the root canal. This presence might be the result of microleakage or microleakage might intensify the activity of residual bacteria. *E. faecalis* is the most common reason for root canal therapy failure because of its strength, small size and ability to grow independently from other microorganisms. Therefore, in this study we used *E.*

faecalis (ATCC 29212) which penetrated into all the samples over 40 days. Therefore, it is too difficult to achieve an impermeable three-dimensional seal.

One of the potential factors affecting microleakage is the presence of moisture in the root canal. This moisture might be due to remaining irrigants, interstitial fluid or delayed bleeding out of the sight of the operator.

The results of this study showed no significant difference between the two sealer types in the moist and dry groups. Although the dry group was somewhat more resistant to microleakage, this result is consistent with the results of previous experimental studies.^{11,13,15,17} The reason for this similarity might be the sealer type because resin sealers are able to bond to the root canal walls without being affected by a slight amount of moisture.

The only significant difference found in this study was between groups 2 (AH26 dry) and 3 (AH-plus wet). Consequently, it would be inferred that AH26 sealer is more resistant to microleakage than AH-plus sealer. The cause of this diversity might be differences in study methods or sample sizes.

In spite of the absence of any significant difference between these two types of resin sealers in dry and moist environments in this study, it is recommended that another similar study be undertaken with different microleakage evaluation method like fluid infiltration technique.

In this study, since microleakage occurred in all the samples, the main variable was the time of microleakage in the groups, which was analyzed with survival test ($p < 0.05$).

Evaluation of the effect of moisture on microleakage with the use of normal saline as a moisturizing agent was one of the limitations of this study because other moisturizing agents might affect microleakage such as irrigants and interstitial fluid or blood.

Conclusion

The results of this experimental study showed that among the four groups of the study, the highest resistance to microleakage belonged to group 2 (AH26 dry) and the least resistance belonged to group 3 (AH-plus wet). This difference in resistance was not significant between other groups.

References

1. Sheikh Rezaei MS, Ghazikhansari DM, Soltanpanah E, Danesh F. Effect of existence of liquid in canal on apical seal by AH26 and Rosen. *J Islamic Dent Assoc.* 2006;3:42-46.
2. Sevimay S, Kalayci A. Evaluation of apical sealing ability and adaptation to dentine of two resin-based sealers. *J Oral Rehabil.* 2005;32(2):105-110.
3. Kalra M, Iqbal K, Nitisusanta LI, Daood U, Sum CP, Fawzy AS. The effect of proanthocyanidins on the bond strength and durability of resin sealer to root dentine. *Int Endod J.* 2013;46(2):169-178.

4. Gibby SG, Wong Y, Kulild JC, Williams KB, Yao X, Walker MP. Novel methodology to evaluate the effect of residual moisture on epoxy resin sealer/dentine interface: a pilot study. *Int Endod J.* 2011;44(3):236-244.
5. Tay FR, Loushine RJ, Monticelli F, Weller RN, Breschi L, Ferrari M, *et al.* Effectiveness of resin-coated gutta-percha cones and a dual-cured, hydrophilic methacrylate resin-based sealer in obturating root canals. *J Endod.* 2005;31(9):659-664.
6. Morgental RD, Singh A, Sappal H, Kopper PM, Vier-Pelisser FV, Peters OA. Dentin inhibits the antibacterial effect of new and conventional endodontic irrigants. *J Endod.* 2013;39(3):406-410.
7. Bergmans L, Moisiadis P, De Munck J, Van Meerbeek B, Lambrechts P. Effect of polymerization shrinkage on the sealing capacity of resin fillers for endodontic use. *J Adhes Dent.* 2005;7(4):321-329.
8. Hargreaves KM, Berman LH, editors. *Cohen's pathways of the pulp.* 11th ed. St. Louis:Elsevier Health Sci;2015.
9. Vivacqua-Gomes N, Ferraz CC, Gomes BP, Zaia AA, Teixeira FB, Souza-Filho FJ. Influence of irrigants on the coronal microleakage of laterally condensed gutta-percha root fillings. *Int Endod J.* 2002;35(9):791-795.
10. Shokouhinejad N, Gorjestani H, Nasseh AA, Hoseini A, Mohammadi M, Shamshiri AR. Push-out bond strength of gutta-percha with a new bioceramic sealer in the presence or absence of smear layer. *Aust Endod J.* 2013;39(3):102-106.
11. Vilanova WV, Carvalho-Junior JR, Alfredo E, Sousa-Neto MD, Silva-Sousa YT. Effect of intracanal irrigants on the bond strength of epoxy resin-based and methacrylate resin-based sealers to root canal walls. *Int Endod J.* 2012;45(1):42-48.
12. Neelakantan P, Subbarao C, Subbarao CV, De-Deus G, Zehnder M. The impact of root dentine conditioning on sealing ability and push-out bond strength of an epoxy resin root canal sealer. *Int Endod J.* 2011;44(6):491-498.
13. Kuhre AN, Kessler JR. Effect of moisture on the apical seal of laterally condensed gutta-percha. *J Endod.* 1993;19(6):277-280.
14. Gonçalves L, Silva-Sousa YT, Raucci Neto W, Teixeira CS, Sousa-Neto MD, Alfredo E. Effect of different irrigation protocols on the radicular dentin interface and bond strength with a methacrylate-based endodontic sealer. *Microsc Res Tech.* 2014;77(6):446-452.
15. Costa JA, Rached-Júnior FA, Souza-Gabriel AE, Silva-Sousa YT, Sousa-Neto MD. Push-out strength of methacrylate resin-based sealers to root canal walls. *Int Endod J.* 2010;43(8):698-706.
16. Carneiro SM, Sousa-Neto MD, Rached FA Jr, Miranda CE, Silva SR, Silva-Sousa YT. Push-out strength of root fillings with or without thermomechanical compaction. *Int Endod J.* 2012;45(9):821-828.
17. Alfredo E, Silva SR, Ozório JE, Sousa-Neto MD, Brugnera-Júnior A, Silva-Sousa YT. Bond strength of AH Plus and Epiphany sealers on root dentine irradiated with 980 nm diode laser. *Int Endod J.* 2008;41(9):733-740.
18. Mamootil K, Messer HH. Penetration of dentinal tubules by endodontic sealer cements in extracted teeth and in vivo. *Int Endod J.* 2007;40(11):873-881.
19. 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(1):23-28.
20. Ballal NV, Tweeny A, Khechen K, Prabhu KN, Satyanarayan, Tay FR. Wettability of root canal sealers on intraradicular dentine treated with different irrigating solutions. *J Dent.* 2013;41(6):556-560.
21. Weller RN, Tay KC, Garrett LV, Mai S, Primus CM, Gutmann JL, *et al.* Microscopic appearance and apical seal of root canals filled with gutta-percha and ProRoot Endo Sealer after immersion in a phosphate-containing fluid. *Int Endod J.* 2008;41(11):977-986.
22. Lin S, Zuckerman O, Weiss EI, Mazor Y, Fuss Z. Antibacterial efficacy of a new chlorhexidine slow release device to disinfect dentinal tubules. *J Endod.* 2003;29(6):416-418.
23. Lima KC, Fava LR, Siqueira JF Jr. Susceptibilities of *Enterococcus faecalis* biofilms to some antimicrobial medications. *J Endod.* 2001;27(10):616-619.
24. Komorowski R, Grad H, Wu XY, Friedman S. Antimicrobial substantivity of chlorhexidine-treated bovine root dentin. *J Endod.* 2000;26(6):315-317.
25. Giardino L, Ambu E, Savoldi E, Rimondini R, Cassanelli C, Debbia EA. Comparative evaluation of antimicrobial efficacy of sodium hypochlorite, MTAD, and Tetraclean against *Enterococcus faecalis* biofilm. *J Endod.* 2007;33(7):852-855.
26. Abdullah M, Ng YL, Gulabivala K, Moles DR, Spratt DA. Susceptibilities of two *Enterococcus faecalis* phenotypes to root canal medications. *J Endod.* 2005;31(1):30-36.
27. Parker HH 4th, Johnson RB. Effectiveness of ethylene oxide for sterilization of dental handpieces. *J Dent.* 1995;23(2):113-115.
28. Razavian H, Barekatin B, Shadmehr E, Khatami M, Bagheri F, Heidari F. Bacterial leakage in root canals filled with resin-based and mineral trioxide aggregate-based sealers. *Dent Res J.* 2014;11(5):599-603.

Corresponding Author

Dr. Kiana Hanjani

Dental Student,
Isfahan University of Medical Sciences,
School of Dentistry,
Isfahan, Iran.
Email Id: - kianahanjani@gmail.com