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Research Article

ASSESSMENT OF DENTINAL TUBULE PENETRATION OF AH-PLUS, MTA FILLAPEX AND GUTTAFLOW BIOSEAL ROOT CANAL SEALERS AFTER PASSIVE ULTRASONIC IRRIGATION – A CONFOCAL MICROSCOPIC STUDY

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A B S T R A C T

Aim: To evaluate the effect of three different root canal sealers on dentinal tubule penetration after ultrasonic passive irrigation and its assessment by confocal microscope.
Material and Method: A total of 30 single rooted extracted mandibular premolars were prepared upto F3 (Size 30, 0.06 taper) using rotary Protaper Universal system and irrigated with sodium hypochlorite along with passive ultrasonic irrigation. The specimens were randomly divided into 3 groups and obturated using gutta percha cone with single cone technique with sealers such as Group A: AH Plus, Group B: Gutta flow Bioseal,, Group C: MTA Fillapex. Specimens were sectioned at 2mm, 5mm and 8 mm from the apex and all sections were examined under confocal microscope. **Result:** AH Plus sealer showed significant difference in sealer penetration between 2mm and 5mm, 5mm and 8mm, and 2mm and 8mm . MTA Fillapex showed significant difference in sealer penetration between 2mm and 8mm, 5mm and 8mm. Guttaflow Bioseal showed a significant difference between 2mm and 8mm.
Conclusion: Within the limitations of the study it can be concluded that sealer penetration in dentinal tubules is highest for AH Plus sealer and is highest in coronal third of canal and lowest at the apical third.

INTRODUCTION

Success of endodontic treatment is mainly based on adequate biomechanical preparation and three-dimensional obturation of root canal system. Root canal sealers play a critical role in the success of endodontic therapy by eliminating the space between the root canal wall and core filling material.[1] However, microscopic gaps between the sealer and dentin and sealer and core material jeopardize the outcome of root canal treatment, and marginal leakage through these gaps continues to be a major reason for the failure of root canal therapy.[2]

To minimize marginal gaps between sealer and dentin, there has been continuous search for the alternative endodontic sealers which could adhere to dentin. In this study we have compared the dentinal tubule penetration of AH-Plus sealer, MTA Fillapex sealer and Gutta-flow Bioseal sealer and gutta-percha is used as a core obturating material.

AH-Plus, an epoxy resin-based sealer, has shown to produce high bond strength to the canal wall and adequate long term dimensional stability.[3] However, tubular penetration and adaptation to peritubular dentin of AH Plus sealer has been reported in some studies.[4,5]

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low solubility.[6] Because of its suitable physicochemical properties and excellent biocompatibility and bioactivity, it has attracted considerable attention. Hence this study was planned to evaluate the depth of dentinal tubule sealer penetration of MTA Fillapex sealer.

GuttaFlow 2 and later introduced Guttaflow Bioseal, a new formulation of Gutta flow is a silicone-based root canal sealer that combines sealer and guttapercha in powder form with a particle size of less than 30 micrometer.[7] It consists of a mixture of gutta-percha powder, poly-dimethylsiloxane, platinum catalyst, zirconium dioxide, and micro-silver. GuttaFlow 2 has been shown to be more biocompatible than AH Plus Jet sealer and less toxic to human gingival fibroblasts cells than AH Plus.[8,9]

The aim of this in vitro study was to assess the dentinal tubule penetration of the three various sealers after the application of passive ultrasonic irrigation technique. The null hypothesis was that there would be no difference among the sealer penetration after passive ultrasonic irrigation technique.

MATERIALS AND METHOD

A total sample of 30 single rooted extracted mandibular premolars without any defects were prepared upto F3 (size 30, 0.06 taper) using rotary Protaper Universal system (Dentsply

Maillefer, Switzerland) and irrigated with 2 ml of 5% NaOCl at each instrument change. Passive ultrasonic irrigation was carried out by using 0.5 ml of NaOCl & Irri-Safe ultrasonic tips (#25,25 mm; Acteon, France) placed 2 mm short of working length and using up and down movement with ultrasonic device for 1 minute. It was later rinsed with distilled water. Final irrigation was done using 5 ml of 17% EDTA for 1 minute followed by 5ml of distilled water. The specimens were randomly distributed into 3 groups (n=10) as follows: Group A: AH Plus (Dentsply, DeTrey, Konstanz, Germany), Group B: GuttaFlow Bioseal (Coltène/Whaledent AG, Altstatten, Switzerland) and Group C: MTA Fillapex (Angelus, Londrina, PR, Brazil). The root canals are then dried using paper points. The canals were coated with the respective sealers mixed with 0.1% fluorescent rhodamine B isothiocyanate, 1 mm short of working length using lentulospiral and obturated using single gutta-percha cone. Specimens were incubated at 37ºC for one week to simulate oral conditions. Specimens were sectioned horizontally at 2, 5 and 8 mm from anatomic apex and all sections were examined under confocal microscope to calculate dentinal tubule penetration area.

Digital images were imported into the Fiji program (Fiji, ImageJ software, NIH) to measure the total dentinal tubule penetration area. The dentinal tubule penetration area was measured as micrometers (mm) and converted to square millimeters (mm^2) for the statistical analysis.

Statistical Analysis

Statistical analysis was done within groups of sealer and between groups of sealer at 2mm, 5mm and 8mm using ANOVA test. If statistically significant difference was observed, further analysis was done using post hoc Tukey's test.

RESULTS

When comparison was made between the groups of sealers, statistically highly significant difference ($p<0.001$) was obtained at the 2 mm level for AH Plus, GuttaFlow Bioseal and MTA Fillapex. On further applying Post-hoc Tukey's test, MTA Fillapex showed a significant difference with AH Plus, Gutta Flow Bioseal sealers at 2mm. At 5mm & 8mm levels, the comparison was not significant between the three sealers.

When comparison was made within the groups, all sealers showed a highly significant difference within themselves at the 2 mm, 5 mm and 8 mm levels. On further applying Post-hoc Tukey's test, AH Plus showed a significant difference in sealer penetration between 2 mm and 5 mm, 5mm and 8 mm and 2 mm and 8 mm. MTA Fillapex showed a significant difference in sealer penetration between 2 mm and 8 mm, 5 mm and 8 mm. GuttaFlow Bioseal showed a significant difference in sealer penetration between 2 mm and 8 mm.

Table 1 Mean and standard deviation of sealer penetration area in μm^2 for AH PLUS, GUTTAFLOW BIOSEAL and MTA FILLAPEX AT 2 mm, 5 mm, 8 mm sections. Non-identical letters in superscript indicate statistically significant difference between groups.

	AT 2 mm (apical)	AT 5 mm (middle)	AT 8 mm(coronal)
AH plus	(226527.64±38144.67)^a	(661841.50±429830.67)^d	(1726630.24±350058.77)^e
Gutta flow	(205365.50±19401.90)^a	(545127.58±314470.97)^d	(1399982.26±929701.96)^e
MTA fillapex	(118411.92±17355.68)^b	(466148.11±218348.08)^d	(1544125.47±784912.64)^e

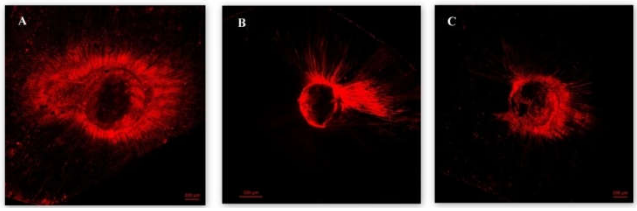


Figure 1 Representative images of sections scanned by confocal laser microscope for specimens obturated with AH sealer at (A) 8 mm, (B) 5 mm and (C) 2mm level from the root apex.

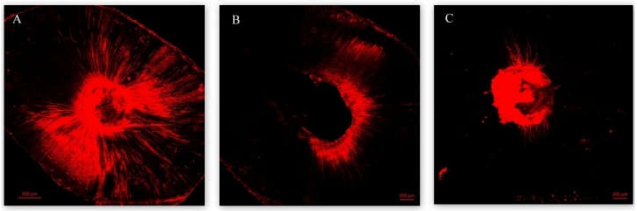


Figure 2 Representative images of sections scanned by confocal laser microscope for specimens obturated with GuttaFlow Bioseal sealer at (A) 8 mm, (B) 5 mm and (C) 2mm level from the root apex.

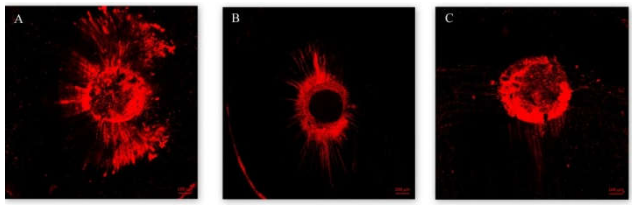


Figure 3 Representative images of sections scanned by confocal laser microscope for specimens obturated with MTA Fillapex sealer at (A) 8 mm, (B) 5 mm and (C) 2mm level from the root apex.

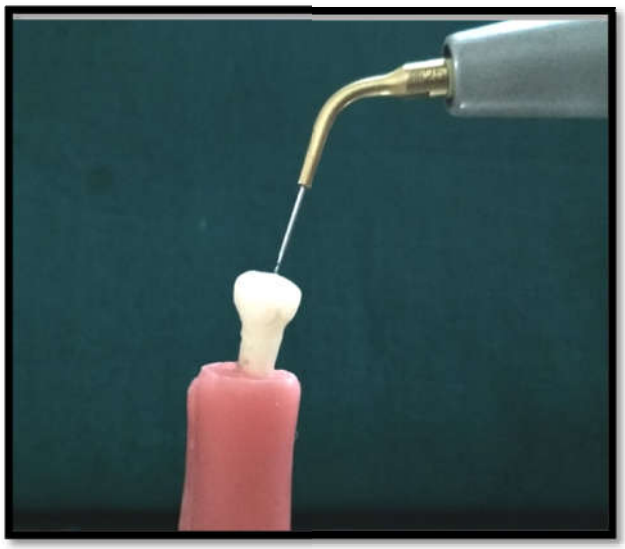
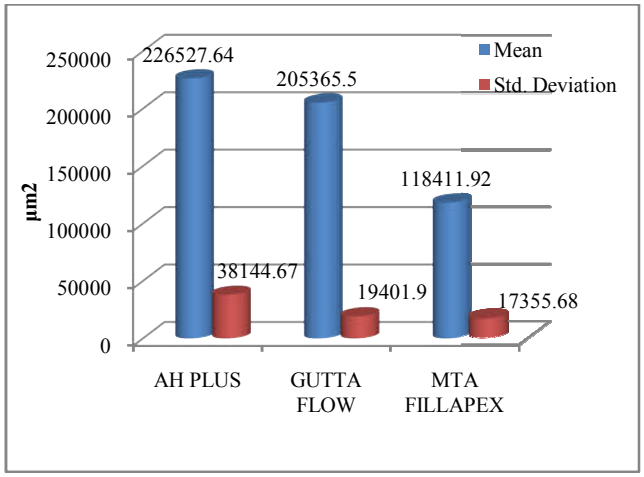
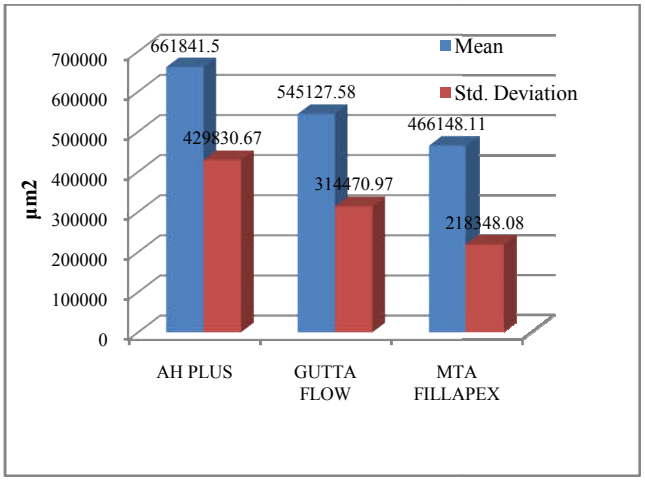


Figure 4 Passive Ultrasonic irrigation being performed with Irrisafe Tips.

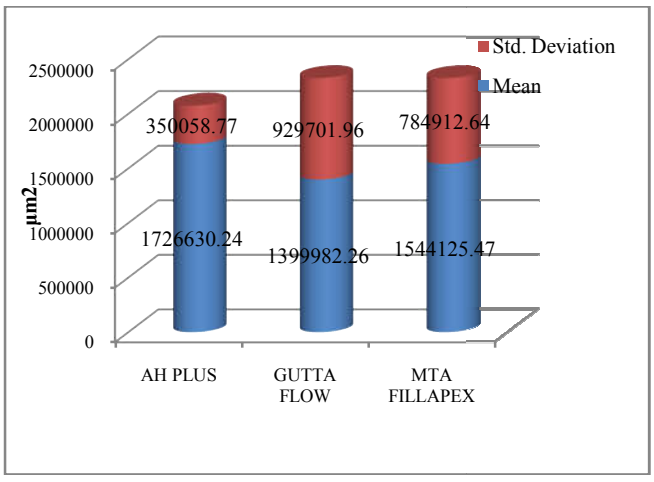
Figure 5 Specimens after transverse sectioning at 2mm, 5mm and 8 mm from the apex.



Graph 1 Comparison of sealer penetration area between AH Plus, GuttaFlow Bioseal and MTA Fillapex at 2mm



Graph 2 Comparison of sealer penetration area between AH Plus, Gutta Flow Bioseal and MTA Fillapex at 5mm



Graph 3 Comparison of sealer penetration area between AH Plus, GuttaFlow Bioseal and MTA Fillapex at 8mm

DISCUSSION

Adequate sealing of the root canal system is influenced by the properties of flow and adhesiveness of the sealers. Flow is essential to allow adequate time for the sealer to penetrate the dentinal tubules and to set and whereas adhesiveness confirms stability of the filling mass to the walls of the dentin.[10] Penetration of sealers in tubules confines the microbes by entombing them and helps as an antiseptic.[11]

Removal of smear layer is a pre-requisite to allow for opening of tubules to allow for ingress of the sealer.[12] Irrisafe tips (Satelec Acteon Group, Merignac, France) were used for passive ultrasonic irrigation (PUI) in our study. They are attached to an ultrasonic device operating in the range of 25–30 kHz that activates the irrigant solution. This occurs due to acoustic streaming and microcavitation and it allows the delivery of irrigants up to the working length of the root canal unlike conventional endodontic needle.[13] Its efficacy has been proved in a study by Paragliola *et al* who showed that the use of an ultrasonic agitation exhibited significantly more penetration of irrigants in tubules than sonic agitation.[14]

The current study used the single cone obturation technique as it is the common, simplest technique and ensures uniformity. The use of a single-cone filling technique is often considered inferior to more sophisticated three-dimensional compaction techniques.[15] However, it must be noted that the concept of the single-cone technique has been recently re-visited by Wu *et al*[16] and the volume of the sealer used in the present study was minimized because calibrated gutta-percha cones were used in the prepared canal. Also a study conducted by Kok *et al* showed that sealer penetration was not affected when different filling techniques were used along with an epoxy resin sealer.[17]

CLSM was used in our current study as opposed to SEM analysis as it presents with many advantages. CLSM uses high contrast points to identify sealer distribution within dentinal tubules.[17] It works even with thick section where it collects serial optical sections by making possible control over depth and reducing background information. It also cause less artifact as opposed to SEM which is notorious about producing artifact during processing along with additional disadvantages of the time consuming gold-sputtering and vacuum stages.[18]

Physical properties of sealers like flow, surface tension, solubility, viscosity affect their penetration into critical areas. The current study shows that greatest penetration was observed for AH plus at all the three levels of examination. AH plus belongs to group of epoxy resin based sealers. These sealers have been known for their desirable properties of greater dimensional stability, low rates of solubility, great radiopacity and optimal adhesiveness to the root dentin than others endodontic sealers and currently serve as gold standards for comparison.[19,20,21] The pseudoplastic behaviour of AH Plus is a benefit which allow increased flow by reduction of viscosity during filling procedures thus enhancing its penetration in tubules.[22]

Guttaflow Bioseal is a newer product of the Roeokoseal, Guttaflow and Guttaflow 2 family. It is a cold flowable system that contains gutta percha particles in silicone based polymethyl hydrogen siloxane. This system is known to provide a linear setting expansion of 0.16% on obturation.[23] It is rarely studied for its dentinal tubule penetration uptil date. Another silicone-based sealer, RoekoSeal, was showed to have a similar dentinal penetration as that of AH Plus sealer in a confocal laser scanning microscope study by Chandra *et al*.[24]

MTA Fillapex is a based on tricalcium silicate cement impregnated in a matrix of salicylate resin. They are known for their biocompatibility, adhesiveness, solubility and antibacterial activity.[25] The high solubility and presence of hydrophilic characteristics may be the reason for inferior results than other sealers used in this study. However this

sealer has shown superior result in a study conducted by Kuci *et al* where it was compared with AH 26 sealer using both cold lateral and warm vertical compaction techniques. The result concluded higher penetration for MTA Fillapex with cold lateral compaction and for AH plus with warm vertical compaction.[26] Absence of consensus with this result may be because of decreased viscosity and greater flow of MTA Fillapex under compaction pressure which was absent in our study due to single cone obturation technique used.

As previously investigated by Kok, the sealer penetration of epoxy resins is not affected by the technique of obturation used.[17] This implies that the hydraulic forces have no role in pushing the sealers into tubules and the sealer penetrates the tubule by virtue of capillary forces that draw it in.[24] This may also explain how AH Plus and Guttaflow Bioseal with higher setting time are available for more penetration when in contact with tubules as stated by Chandra *et al*.[24]

The results of this study showed that the maximum penetration of all the sealers was seen at the coronal third, followed by the middle third and least in the apical third. This is attributed to variations in properties of radicular dentin majorly relating first to its tubule density, size of tubules, and metamorphic changes that undergo over time. Coronal dentin has the highest tubule density and it decreases as one moves further, the least found apically. The mean diameter of tubules is also greatest in the coronal third and least occurring in the apical third of root. This favours high penetration coronally. The tubules in the apical third happen to show obliteration due to sclerosis as a result of constant functional loading or ageing. Poor delivery of irrigants and ineffectiveness of smear layer removal techniques in the apical third is also a reasonable explanation for the sealer unable to acquire the desired space.[26,27] All these predispose to the obtained pattern of result also demonstrated by various other studies.[28,29,30]

For the dentinal tubule penetration evaluation in the confocal laser scanning microscopy, generally, two parameters have been measured, the maximum depth of penetration and the percentage of the sealer penetration. [31] However, these methods have some limitations. Using one or multiple measurements affected the overall depth, and the thick/loose penetration changed the percentage. Therefore, in the present study, the ImageJ program was used to measure the total dentinal tubule penetration area as previously done by Ackay *et al*.[32] The program calculated the area of both user-defined selections and intensity-threshold objects.

CONCLUSION

Within the limitations of the study it can be concluded that the sealer penetration in dentinal tubules highest for AH Plus sealer. It is highest at coronal third of the canal and lowest at apical third. It is governed by anatomic features of dentin pertaining to coronal, middle or apical thirds. It is also dependent on the physical properties of the sealer.

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References

1. Deus GD, Gurgel Filho ED, Ferreira CM, Coutinho Filho T. Penetração intratubular de cimentos endodônticos. Pesquisa Odontológica Brasileira. 2002 Dec;16 (4):332-6.
2. Orstavik, D. Materials used for root canal obturation: technical, biological and clinical testing. Endod Top 2005;12:25-38.
3. Duarte MA, Ordinola-Zapata R, Bernardes RA, Bramante CM, Bernardineli N, Garcia RB, De Moraes IG. Influence of calcium hydroxide association on the physical properties of AH Plus. J Endod. 2010 Jun 1;36 (6):1048-51.
4. 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.
5. 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.
6. Torabinejad M, Parirokh M. Mineral trioxide aggregate: a comprehensive literature review–part II: leakage and biocompatibility investigations. J Endod 2010;36: 190–202.
7. Bouillaguet S, Wataha JC, Tay FR, *et al*. Initial in vitro biological response to contemporary endodontic sealers. J Endod 2006;32:989–92.
8. Accardo C, Himel VT, Lallier TE. A novel GuttaFlow sealer supports cell survival and attachment. J Endod 2014;40:231–4.
9. Mandal P, Zhao J, Sah SK, *et al*. In vitro cytotoxicity of guttaflow 2 on human gingival fibroblasts. J Endod 2014;40:1156–9.
10. De-Deus G, Coutinho-Filho T, Reis C, Murad C, Paciornik S. Polymicrobial leakage of four root canal sealers at two different thicknesses. J Endod 2006;32:998-1001.
11. Silva RV, Silveira FF, Horta MC, Duarte MA, Cavenago BC, Morais IG, Nunes E. Filling effectiveness and dentinal penetration of endodontic sealers: a stereo and confocal laser scanning microscopy study. Braz dent J. 2015 Oct;26(5):541-6.
12. Bacterial penetration through canals of endodontically treated teeth in the presence or absence of the smear layer. Clark-Holke D, Drake D, Walton R, Rivera E, Guthmiller JM. J Dent. 2003 May; 31(4):275-81
13. Munoz HR, Camacho-Cuadra K. In vivo efficacy of three different endodontic irrigation systems for irrigant delivery to working length of mesial canals of mandibular molars. J Endod 2012;38:445–8.
14. Paragliola R, Franco V, Fabiani C, Mazzoni A, Nato F, Tay FR, Breschi L, Grandini S. Final rinse optimization: influence of different agitation protocols. Journal of endodontics. 2010 Feb 1;36(2):282-5.
15. Kontakiotis EG, Tzanetakis GN, Loizides AL. A l2-month longitudinal in vitro leakage study on a new silicon-based root canal filling material (Gutta-Flow). Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2007;103:854–9.
16. Wu MK, Bud MG, Wesselink PR. The quality of single cone and laterally compacted gutta-percha fillings in small and curved root canals as evidenced by

bidirectional radiographs and fluid transport measurements. Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology. 2009 Dec 1;108 (6):946-51.
17. Kok D, H_ungaro Duarte MA, Abreu Da Rosa R, *et al*. Evaluation of epoxy resin sealer after three root canal filling technique by confocal laser scanning microscopy. Microsc Res Technique 2012;75:1277–80.
18. Mamootil K, Messer HH (2007) Penetration of dentinal tubules by endodontic sealer cements in extracted teeth and in vivo. Int Endod J 40:873–881. doi:10.1111/j.1365-2591.2007.01307.x
19. Leonardo MR, Silva LA, Filho MT, *et al*: Release of formaldehyde by 4 endodontic sealers. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 1999; 88: 221–225.
20. Orstavik D, Nordahl I, Tibballs JE: Dimensional change following setting of root canal sealer materials. Dent Mater 2001; 17: 512–519.
21. Duarte MA, Ordinola-Zapata R, Bernardes RA,Bramante CM, Bernardineli N, Garcia RB *et al*.Influence of calcium hydroxide association on the physical properties of AH Plus.J Endod 2010;36:1048-1051.
22. Zhou HM, Shen Y, Zheng W, *et al*. Physical properties of 5 root canal sealers.J Endod 2013;39:1281–6.
23. Hammad M, Qualtrough A, Silikas N. Extended setting shrinkage behavior of endodontic sealers. JEndod. 2008; 34:90–3.
24. Chandra SS, Shankar P, Indira R. Depth of penetration of four resin sealers into radicular dentinal tubules: a confocal microscopic study. *Journal of endodontics*. 2012 Oct 1;38 (10):1412-6.
25. Torabinejad M, Parirokh M. Mineral trioxide aggregate: A comprehensive literature review-part II: Leakage and biocompatibility investigations. J Endod 2010;36 (2):190–202.
26. Kuci A, Alacam T, Yavas O, Ergul-Ulger Z, Kayaoglu G. Sealer penetration into dentinal tubules in the presence or absence of smear layer: A confocal laser scanning microscopic study. J Endod 2014;40(10):1627–1631.Teixeira CS, Felippe MC, Felippe
27. WT. The effect of application time of EDTA and NaOCl on intracanal smear layer removal: An SEM analysis. Int Endod J 2005;38 (5):285–290.
28. Paque F, Luder HU, Sener B, Zehnder M. Tubular sclerosis rather than the smear layer impedes dye penetration into the dentine of endodontically instrumented root canals. Int Endod J 2006;39(1):18–25.
29. Moon YM, Kim HC, Bae KS, Baek SH, Shon WJ, Lee W. Effect of laser-activated irrigation of 1320-nanometer Nd:YAG laser on sealer penetration in curved root canals. J Endod 2012;38 (4):531–535.
30. Gharib SR, Tordik PA, Imamura GM, Baginski TA, Goodell GG. A confocal laser scanning microscope investigation of the epiphany obturation system. J Endod 2007;33 (8):957–961.
31. Akcay M, Arslan H, Mese M, Durmus N, Capar ID. Effect of photon-initiated photoacoustic streaming, passive ultrasonic, and sonic irrigation techniques on dentinal tubule penetration of irrigation solution: a confocal microscopic study. Clinical oral investigations. 2017 Sep 1;21 (7):2205-12.
32. Kara Tuncer A, Tuncer S. Effect of different final irrigation solutions on dentinal tubule penetration depth and percentage of root canal sealer. J Endod 2012;38 (6):860–863.

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