

Scanning Electron Microscope Evaluation of Dentinal Tubules Penetration of Three Different Root Canal Sealers

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Abstract

Aim: The aim of this study was to evaluate tubular depth of penetration of three different types of sealers in the apical, middle, and coronal thirds in conjunction with a single cone condensation of gutta percha.

Methods: Thirty single rooted permanent anterior teeth were prepared then filled with 3 different root canal sealers and gutta-percha using single cone technique. Roots were then cross-sectioned and set for scanning electron microscope assessment. Sealer adaptation to root canal and tubular walls together with tubular penetration were evaluated.

Results: At the coronal level there was no statistically significant difference between the three tested groups, while at the middle Apexit showed the best penetration. However, at the apical level the only penetration was recorded with AH-Plus. There was no statistically significant difference between sealer penetration between the coronal and middle levels, while the apical level showed the least sealer penetration among the three tested groups.

Conclusions: Tubular penetration differs along with the difference in sealers properties; physical and chemical. Optimal tubular penetration was reported by AH Plus in the apical part of all sealers tested.

Keywords: Scanning Electron Microscope; Dentinal Tubule Penetration; AH Plus; Apexit Plus; Smartpaste Bio

Introduction

Microorganisms in root canals are considered main causes of apical periodontitis [1]. Regarding major objectives of root canal therapy, disinfecting root canal space and prevention of its reinfection are the most important. Bacteria can remain in areas like ramification, anastomosis and dentinal tubules, which act as shelter from all disinfecting irrigants as well as intracanal medicaments [2].

The perfect penetration, adaptation, together with adhesion properties of root canal sealers are counted as favorable effects. Sealing can be achieved by increasing the sealer-dentin contact then residual bacteria in the dentinal tubules can be entombed by sealer [3]. Since there is no adhesion with dentin the sealer penetration into dentinal tubules could be a perfect sign [4].

The extrusion of Bacteria engaged within root canals can be only avoided by achieving perfect seal. However, in order to completely eliminate residual microorganisms, root canal sealer with antimicrobial properties could have a role in disinfecting root canal [5].

Epoxy resin-based sealers as AH-Plus (Dentsply- Maillefer, Germany) were greatly used with recorded high bond strength values to dentin and adequate biological property [6,7].

Apexit Plus (Ivoclar-Vivadent, Schaan, Liechtenstein), one of the calcium hydroxide root canal sealers, was introduced to the market for periapical tissue stimulation. This in turn stabilizes health or enhance healing. Its antibacterial activity was also considered [8].

Smartpaste bio (Smartseal, UK) is a resin-based sealer containing with active polymer which swells for fill spaces or voids within root canal. The manufacturer claims that the polymer may also swell latterly for filling the voids which might develop [9].

Therefore, our goal is the assessment *in vitro* of penetration depth of 3 root canal sealers in the apical, middle, and coronal third while using single gutta percha cone technique.

Materials and Methods

Thirty freshly extracted single-rooted permanent human teeth of similar size and root shape were selected and stored in 3% formaldehyde solution. The crowns were sectioned at the cement-enamel junction using diamond bur mounted on high-speed hand piece (Alegra HS, WandH, Eckbolsheim, France) under continuous water spray. Size 10 K-file (Mani Inc, Japan) was introduced into each canal until it could be seen through the apical foramen and the length measured. Working length was established by subtracting 0.5 mm from this length. Glide path was established by using K-files from #10 to 20. The canals were then instrumented using a crown-down technique with rotary ProTaper Universal nickel-titanium files (Dentsply Maillefer, Ballaigues, Switzerland) to F3 taper 0.09 at the Working Length. Between each instrument, the canal was irrigated with 2 mL 5.25 % sodium-hypochlorite (NaOCl) solution with a syringe and a side vented needle (RC Twents, Prime Dental Products, Thane, India). At the end of the preparation, the canal was irrigated with 3 mL 17% EDTA (MD-cleanser, Meta Biomed) for 3 minutes. The final rinse was performed with 5 mL 3% NaOCl. The canal was dried with a sterile paper point size 30, taper 0.02. The specimens were randomly divided into 3 groups (n = 10) and filled with gutta percha using single cone technique and 3 different root canal sealers:

1. **Group 1 sealer:** AH Plus was prepared by following manufacturer's instructions.
2. **Group 2 sealer:** Apexit Plus was mixed with the mixing syringe supplied by the manufacturer.
3. **Group 3 sealer:** Smart Paste bio was supplied in premixed form.

Every sealer was delivered to assigned root canal with a K-file size 15 placed at working Length in anti-clockwise direction. Then, walls of every root canal were coated by sealer with F3 gutta percha (Dentsply Maillefer, Ballaigues, Switzerland) placed till working length. Each coronal part was restored using IRM [10]. All teeth were stored (37°C and 100% humidity) for two weeks, to ensure that sealers were fully set.

Roots were placed in resin. Then, all samples were sectioned at three, six, and nine mm from the root tip using a 0.3 mm disk thickness (Isomet 4000, Buehler, USA). To eliminate all inorganic debris, specimens are washed in a bath with 17% EDTA for two minutes. and then 3% NaOCl for two minutes. The specimens were mounted on a tub and gold sputtered. Assessment was done using Scanning Electron Microscope (SEM) (Hitachi science and technology, Japan) (Model number S-3400N) and each specimen was photographed at magnification 1000 X.

On every photomicrograph, sealer depth maximum and minimum penetration within tubules was assessed. Statistical analysis was done for comparing mean penetration depth among different sealers at all tested levels ($P < 0.05$).

Results

Comparison between coronal, middle and apical parts was done using ANOVA and Tukey post-hoc tests. No statistically significant difference was reported among all tested groups for coronal part. At the middle part no statistically, significant difference was reported among groups I and II, also between I and III while, II and III recorded statistically significant difference between each other. As for the apical level a statistically significant difference between I and II, and I and III, while there was no difference between II and III. The *p*-value was 0.10485, 0.039439, 0.00061 for the coronal, middle, and apical respectively.

Root Section	Sealer						P Value
	Group 1		Group 2		Group 3		
	Mean	S. D	Mean	S. D	Mean	S. D	
Coronal	61.3 ^a	± 38.779	73.7 ^a	± 38.422	40.2 ^a	± 22.939	0.10485
Middle	59.7 ^{a,b,c}	± 35.634	69.0 ^b	± 33.490	33.6 ^c	± 19.357	0.039439
Apical	10.2 ^a	± 10.272	0 ^b	0	0 ^b	0	0.00061

Table 1: Comparison between the three groups in coronal, middle and apical parts using Tukey post-hoc test.
*In each row, values with same lower-case letter were not statistically significant.

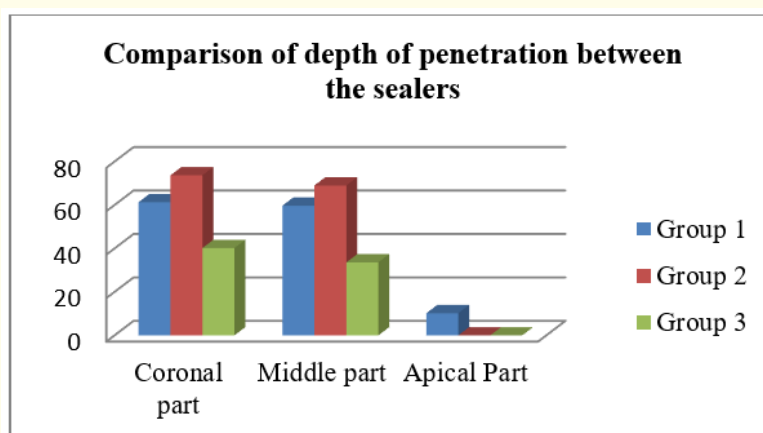
	Coronal	Middle	Apical
Group 1 vs Group 2	.007 (NS)	.774 (NS)	.002 (S)
Group 2 vs Group 3	.091 (NS)	.038 (S)	1.000 (NS)
Group 1 vs Group 3	.365 (NS)	.152 (NS)	.002 (S)

Table 2: Showing calculated *p* value comparison between the groups in coronal, middle and apical parts using Tukey post-hoc test.
*S: Significant, NS: Not Significant.

In the coronal part there were no significant difference. In middle part Group 2 vs Group 3 shows *p* = 0.038 which is significant. In apical part Group 1 vs Group 2 and Group 1 vs Group 3 both shows *p* = 0.002 which is significant.

Root Section	Sealer		
	Group 1	Group 2	Group 3
Coronal vs Middle	.993 (NS)	.932 (NS)	.675 (NS)
Coronal vs Apical	.003 (S)	.000 (S)	.000 (S)
Middle Vs Apical	.004 (S)	.000 (S)	.001 (S)

Table 3: Comparison of penetration of each sealer between different root sections.
Between coronal and middle parts all 3 groups shows no significant difference. Between coronal and apical all three groups show significant difference. Also, between middle and apical all three groups show significant difference.



Graph 1: Comparison of the mean values of sealer penetration.

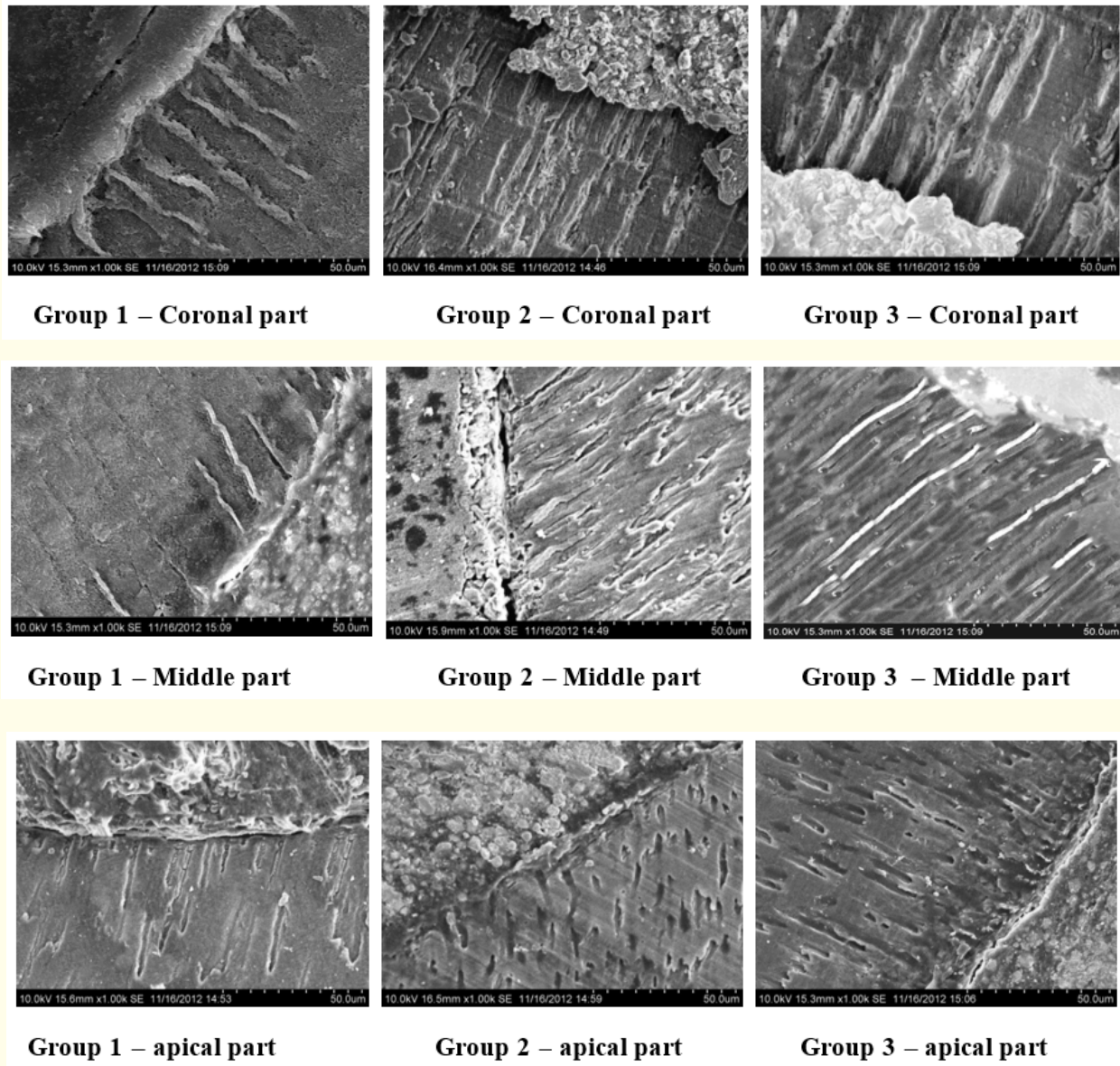


Figure 1: Scanning electron photomicrograph of different sealer penetration in the dentinal tubule at three levels of the root magnification (1000 X).

Discussion

In the current study, sealers penetration within dentinal tubules increased in a significant pattern in both coronal and middle thirds within the canal when compared to apical third in all three groups. This confirms what Balguerie., *et al.* [3] and Weis., *et al.* [11] reported where, Balguerie., *et al.* [3] stated that both coronal and middle thirds showed higher penetration than the apical part ($P < .0416$) with

the exception of Endobtur ($P > .055$) [3]. The study done by Weis., *et al.* has shown that sealer penetration showed increase in depth and occurred frequently at both 3 and 5 mm levels than the 1mm level ($P < .001$) [11].

This may be because apical dentine displays less tubule density as per Mjor., *et al* [12]. In his study, the tubules number decreased from 40,000 - 14,400 (coronal to apical), that meant lesser tubules were found coronally available for resin penetration. Either areas with irregular secondary dentin or those with no tubules might also point out that hybrid layer shall provide good platform for apical dentin adhesion. Observation of apical root dentine showed structural irregularity. This in turn show obstacles upon obturation when those sealer penetration-based techniques were developed. It is extremely hard to control inherent wetness of the hybrid layer apically: so that, 1 - 2 mm of the canal's most apical part can better remain unfilled so that biological healing gains the chance to occur in those parts which the obturating material could not reach [12].

The significantly lesser penetration of sealer in the apical part may also be due to the fact that techniques used to remove smear layer were less effective when approximating the apex as found by O' Connel., *et al* [13]. Also, it might result from presence of dentinal tubules with larger diameter at both cervical and middle parts when tested against the apical part according to Vijay Singh., *et al* [10].

In the present study mean penetration depth of AH Plus at coronal part was 61.3 μm and in the middle part was 59.7 μm which was comparable to results obtained by Balguerie., *et al.* [3] in which the mean penetration depth was 62.5 μm at coronal part and 53.3 μm at middle part. Also, the results came along with Piai., *et al.* [14] who didn't find any differentiation in penetration between AH Plus and sealer plus which is resin sealer. In another study by Akcay., *et al.* [15] no sign. difference between MTA Fillapex, AH Plus and Gutta flow Bioseal was reported, while all of them showed lesser penetration than iRoot SP. Also, our results came along with Abdul Khader [4] who found no difference in tubular penetration when comparing AH-Plus to Apexit Plus.

When compared to our study, greater sealer penetration of resin-based sealer was observed by Zapata., *et al* [16]. The mean penetration depth of resin base sealer was 237.84 μm . This difference may be due to the condensation technique used in their study. Lateral compaction technique was used in the study done by Zapata., *et al.* But this is in contradictory with the study done by Kokkas., *et al* [17]. The results of the study have shown a mean penetration depth of AH Plus of 54.6 μm which was condensed with lateral compaction technique. According to Weis., *et al.* [11] the difference in sealer penetration depth were found to be not related to obturation technique, concluding that sealer penetration is mainly associated with permeability of dentinal tubules together with different properties of the sealer utilized [11]. According to Messer., *et al.* [2] tubular penetration of resin-based sealers does not depend on hydraulic forces formed during obturation; rather the sealer is sucked into the tubules by capillary action.

The results of the study done by C Vijay Singh., *et al.* [10] has shown that mean penetration depth of AH Plus was 24 μm which is considerably lesser than the values obtained in the present study. This could be because, in the study by Chandra Vijay Sing., *et al.* the irrigating solution used was 2.5% NaOCl and EDTA was not used [10]. But in the present study, both NaOCl and EDTA were used as irrigating solution.

In the present study, AH Plus has shown a mean penetration depth of 10.2 μm at the apical part where as Apexit Plus and Smart Paste Bio did not show any penetration in apical part. This may be due to the fact that non-resin-based sealers have shown a granular like appearance inside the tubules, indicating that sealer penetration depth might be related to the size of its particles, flow and surface tension characteristics following the removal of the smear layer according to Weis., *et al* [11].

In the current study, there was no statistical significance between penetration depth of AH Plus and Apexit Plus in both coronal and middle part of the root. That resulted from findings of the study by McMichen., *et al* [6]. According to McMichen., *et al.* the mean flow time for both AH Plus and Apexit were similar which was 5.60 seconds and 5.40 seconds respectively. Also, the rate of flow for both AH Plus and Apexit were similar (0.04 mL/s) [18-21].

Conclusion

Sealer tubular penetration varies with the different physical and chemical characteristics of the used sealer. "AH Plus" have reported greater penetration in apical part of all sealers tested.

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