



RELATIVE ASSESSMENT AND COMPARISON OF MARGINAL ADAPTATION AND TUBULAR PENETRATION WITH VARIOUS OBTURATING MATERIALS: AN OBSERVATIONAL SCANNING ELECTRON MICROSCOPIC STUDY.

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ABSTRACT: prevention of persistent infection and re-infections of root canal is an ideal treatment usually achieved by counter-balancing the bacterial invasions, destruction of bacterial biofilms, thorough debridement and complete sealing of the root canal anatomy. Among all the most widely accepted and successful anticipations in a root canal therapy is a perfect hermetic three dimensional seal. Our study thus

AIMED to evaluate and compare the accuracy of marginal adaptation and tubular penetration of three different sealers with respect to the cervical, middle and apical third of the root canal.

MATERIAL AND METHODOLOGY: 45 single rooted extracted teeth were obtained from the oral surgery department and stored in plastic vials containing artificial saliva. The teeth were then randomly divided into three groups with 15 specimens in each group. Root canal therapies were completed according to the single visit protocol and the canal were eventually filled using a specific sealer. In Group A, bio-root RCS, in Group B, AH plus and in Group C, Sealapex sealers were with single cone technique. The specimens were then sectioned at apical, middle and cervical third and evaluated using electron microscopy.

RESULTS: the data collected was analyzed statistically using means and inter and intragroup comparisons were done using two way ANOVA test. On evaluating the results, statistically significant results were obtained among all the three groups.

CONCLUSION: the results of our study concluded that experience, proper root canal preparations

and genuine obturation techniques can provide an absolute marginal adaptability irrespective of the sealers used.

KEYWORDS: MARGINAL ADAPTATION, OBTURATING MATERIALS, ROOT CANAL TREATMENT, SCANNING MICROSCOPY, TEETH.

INTRODUCTION: One of the fundamental convictions of root canal therapy is the elimination of irritants from the canals with the preservation of natural dentition and filling of the canal with a leak proof material.¹ Newer methods, instruments, materials as well as a deeper comprehension of the structure and anatomy of teeth have completely transformed endodontics.² Even though with the most effective method of root canal therapies, there may still be a trace amount of infectious material in the system. Due to their permeability and siphon effect, the remaining bacteria with their products and the dentinal chips frequently enter the dentinal tubule. The adhesive capacity of bacteria has found to be beneficial when its adhesion has been done to the peri and inter-tubular dentin by fostering bacterial reproduction and proliferation, resulting in the re-infection of the root canal system.³ As a result, an efficient approach to resolving this issue is to obturate the intricate root canal system with a filling material having superior sealing capacity. These materials are capable of embedding the remaining bacteria and preventing their transportation between the apical tissues and the root canal.⁴

In the past, too much emphasis was placed on whether the filling material reached the radiographic apex or not, but the long-term success of root canal treatment depends only on three-dimensional obturation of the canal.⁵ Apart from cervical and apical micro-leakage, a significant factor in the clinical outcome of root canal therapy is the micro-gap present between the sealer and the wall of the root along with its tubular penetration depth. The advantages of a deep penetration are evident with significant increase in the breaking strength of the root canal and good marginal adaptations between the sealer and the root canal wall.^{6,7} It expands the area of contact that exists between the dentin and root canal fillings, which has the potential to improve the sealing capacity of the entire canal system. In addition, it may prevent the entrance of bacteria into the dentinal tubule, and its antimicrobial activity will enhance its effect on contact with the bacteria.⁸ Deep sealer penetration also improves fracture resistance of the root canal. As a result, the ideal sealer must have good marginal adaptability and efficient tubular penetration depth.

One of the most accepted obturating materials of choice with promising results was gutta percha when used in combination with different set of sealers, but because of its hydrophobic and pulling away effect, its usage had to be re-thought.¹⁰ To conquer these defects a number of newer material have been invented with better water resistance, enhanced sealing abilities, good marginal adaptability and effective bond strength. A wide variety of commercially available endodontic sealers are known which are categorized according to their chemical composition, for instance, MTA-based, eugenol-based, epoxy resin-based zinc oxide, bioceramics sealers.¹¹ Thus, the main of our study was to assess and compare the marginal adaptability and tubular penetration of three endodontic sealers (BIO ROOT RCS, AH PLUS and SEALAPEX) in apical, cervical and middle third of the root using scanning electron microscopy.

MATERIAL AND METHODOLOGY: the study was conducted in the Endodontic department of Hazaribag College of Dental sciences and Hospital, Jharkhand. Being an in-vitro study no ethical justification was required except for the departmental permission for allowance of the smooth conductance of the same. 45, single rooted teeth which were extracted for orthodontic reasons were obtained from the department of oral surgery of the same college and stored in plastic vials containing artificial saliva till any procedure was conducted onto them. Non-carious teeth with intact apical foramen and root were selected for the study while restored, fractured, deeply carious, and root canal treated teeth were excluded from the study.

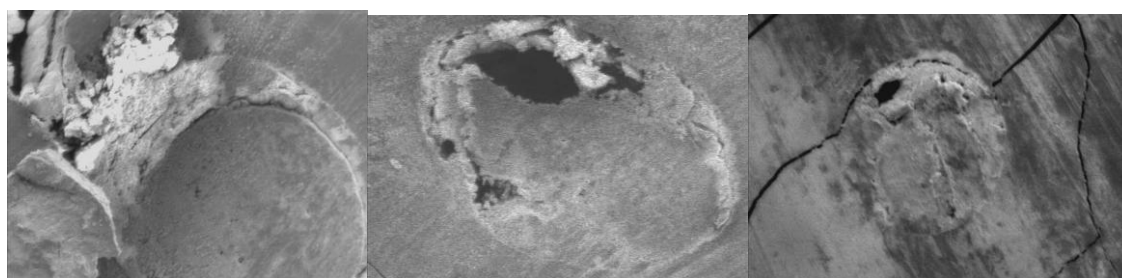
Before starting any procedure on the tooth, the stored teeth were cleaned and any extraneous tissue was removed from them. Conventional endodontic access of the selected tooth was done using a diamond bur (BR-45, 46), followed by copious irrigation using 5% of sodium hypochlorite. An initial working length of the tooth was done using apex locators and confirmed via RVGs. The working length was taken to be 0.5mm short of the radiographic apex. Pro-taper gold rotary files at a speed of 275-300 rpm, torque of 52g.cm and with in and out pecking motion were used in the apical direction for shaping, cleaning (S1, S2 and S3) and finishing (F1, F2 and F3) of the canals. 17% EDTA (Avue prep) was used as a lubricating material followed by 5% sodium hypochlorite and saline. Final obturation of the tooth canal was done using single cone technique with Aurum-pro cones of gutta percha along with the sealers among the three. The whole procedure was completed in a single day by the single operator to avoid any intra-operator error. The teeth were divided into three groups on the basis of the sealer used:

Group A: - 15 samples with Bio root RCS sealer (bio-ceramic).

Group B: - 15 samples with AH plus (resin based).

Group C: - 15 samples with Sealapex (calcium hydroxide based).

All the samples (N=45) prepared were mounted on a block made up of acrylic and 1mm thin sections were created using water-cooled precision low-speed diamond saw (CL-40, BESDIA). The thin slices of the sample were then dehydrated in ascending order of alcohol and mounted on aluminum slabs, where they were sputter coated with gold and observed under scanning electron microscope with the magnification power of 20X, 45X and 400X respectively. For assessing the tubular penetration, the most representative interface of the root-sealer i.e. cervical, middle and apical third of each sample was analyzed. The slices were first subjected to hydrochloric acid (6mol/L) for demineralization followed by soaking in sodium hypochlorite (2.5%) for deproteinization. Figure 1a, 1b and 1c represents the images obtained via scanning electron microscopy using Bioroot RCS sealer. Figure 2a, 2b, and 2c represents the images with AH Plus sealer in cervical, middle and apical third of the root while 3a, 3b and 3 c represent the images with Sealapex sealer respectively.

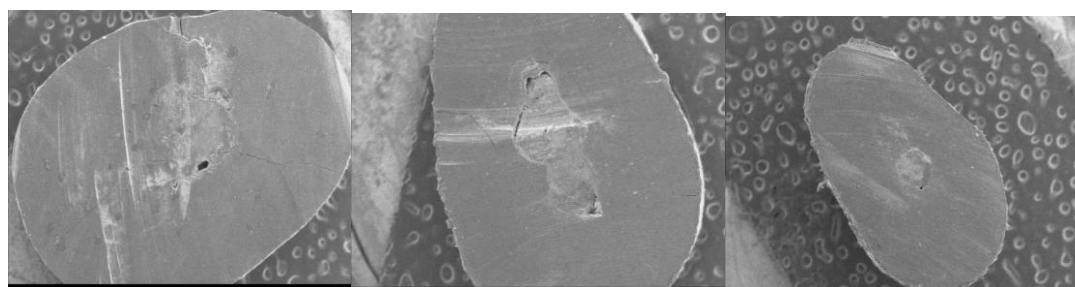


1a (cervical)

1b (middle)

1c (apical)

Figure 1: representing the images of Bioroot RCS sealer



2a (cervical)

2b (middle)

2c (apical)

Figure 2: representing the images of AH Plus sealer

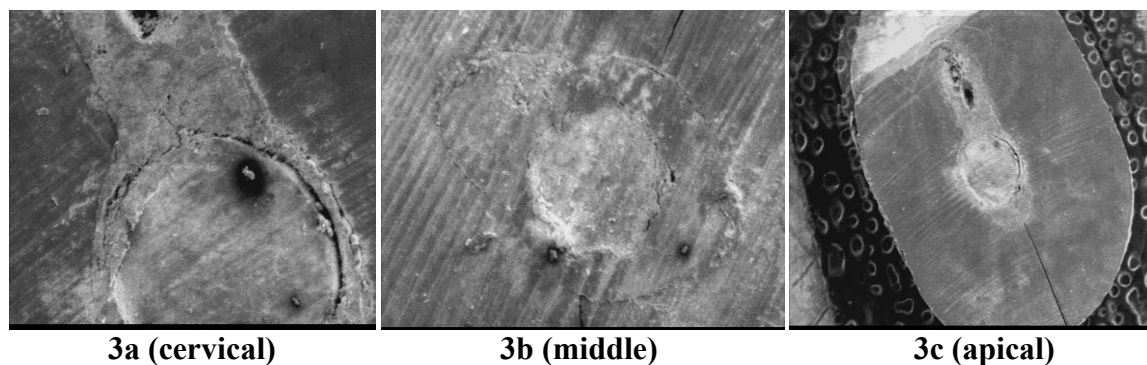


Figure 3: representing the images of Sealapex sealer

STATISTICAL ANALYSIS: the measurements obtained while using scanning electron microscopy were documented, tabulated and transferred to the Microsoft excel sheet for further analysis. SPSS software version 20 was used for further investigation of the compiled data. Descriptive analysis of the data was done using mean, standard deviation and standard error. One way ANOVA test and Post hoc tukey test was used for inter and intragroup comparison of the mean obtained. The significance level of 0.05 was taken as to be statistically significant. The results obtained were then represented using a bar diagram.

SAMPLE SIZE ESTIMATION: the sample size (N) of the study was calculated using $N = z^2 \cdot p \cdot (1-p) / e^2$, where $z=1.96$ for 95% confidence interval, $p=0.03$ as proportion and $e=0.05$ as the margin of error. The estimated value was $44.716 \approx 45$.

RESULTS: table 1, graph 1 represents the mean and standard deviation of the three group at cervical, middle and apical third of the root. On evaluation a higher mean and standard deviation was obtained in group A where Bio-root RCS was used as a sealer along with gutta percha cones followed by group B where AH Plus was used and ultimately group C where sealapex was used as a sealer.

Table 2, 3 and 4 represents the intra and inter group comparison between group 1(bio root rcs), group 2 (AH plus) and group 3 (sealapex). Highly statistically significant results were obtained among all the groups, representing that the difference between the averages of three groups is small enough to be statistically significant.

Table 5, graph 2 represents the mean and standard deviation of three groups while evaluating the penetrating depth at cervical, middle and apical third of the tooth. All the measurements were done in μm . On evaluating a higher values of mean and standard deviation was in the order of group 1 < group 2 < group 3 respectively.

Table 6, 7 and 8 represents the intergroup comparison between the three groups, while evaluating the penetrating depth of the three sealers. A highly statistically significant result were obtained in all which represents that the average difference between the means of all the group was less with significant results obtained only when a comparison of group 2 vs group 3 was done at the cervical and apical third of the involved tooth.

TABLE 1: REPRESENTING THE MEAN VALUES OF THREE GROUPS AT CERVICAL, MIDDLE AND APICAL THIRD OF THE ROOTS						
GROUPS	CERVICAL (n=15)		MIDDLE (n=15)		APICAL (n=15)	
	MEAN	STANDARD DEVIATION	MEAN	STANDARD DEVIATION	MEAN	STANDARD DEVIATION
GROUP A (Bio-root RCS)	226.958	25.935	195.637	34.962	139.012	36.596
GROUP B (AH Plus)	230.197	24.956	110.753	19.253	126.885	23.245
GROUP C (Sealapex)	102.693	42.569	98.553	44.902	87.267	38.489

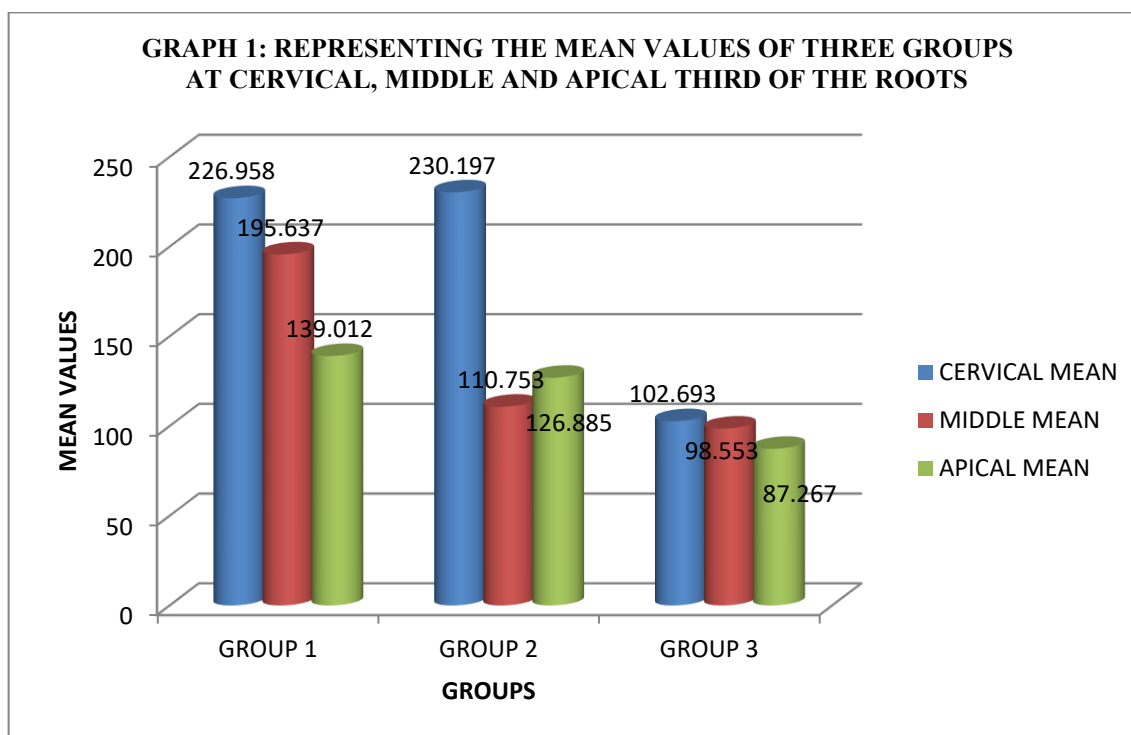


TABLE 2: REPRESENTING THE INTRA AND INTER GROUP COMPARISON BETWEEN GROUP 1, GROUP 2 AND GROUP 3 AT CERVICAL THIRD OF ROOT USING SCANNING ELECTRON MICROSCOPY

GROUPS	SUM OF SQUARES	df	MEAN SQUARE	F-VALUE	SIG.
BETWEEN THE GROUPS	147659.643	2	78329.5	58.559	0.000
WITHIN GROUPS	26464.371	42	630.1040		
TOTAL	174124.014	44			

TABLE 3: REPRESENTING THE INTRA AND INTER GROUP COMPARISON BETWEEN GROUP 1, GROUP 2 AND GROUP 3 AT MIDDLE THIRD OF ROOT USING SCANNING ELECTRON MICROSCOPY

GROUPS	SUM OF SQUARES	df	MEAN SQUARE	F-VALUE	SIG.
BETWEEN THE GROUPS	36691.824	2	18345.912	40.159	0.000
WITHIN GROUPS	69019.912	42	1643.331		
TOTAL	105711.736	44			

TABLE 4: REPRESENTING THE INTRA AND INTER GROUP COMPARISON BETWEEN GROUP 1, GROUP 2 AND GROUP 3 AT APICAL THIRD OF ROOT USING SCANNING ELECTRON MICROSCOPY

GROUPS	SUM OF SQUARES	df	MEAN SQUARE	F-VALUE	SIG.
BETWEEN THE GROUPS	26718.782	2	13359.391	12.047	0.000
WITHIN GROUPS	28369.667	42	675.468		
TOTAL	55088.449	44			

TABLE 5: REPRESENTING THE PENETRATING DEPTH OF THREE SEALERS AT CERVICAL, MIDDLE AND APICAL THIRD OF ROOT USING SCANNING ELECTRON MICROSCOPY

GROUPS	CORONAL MEAN±SD	MIDDLE	APICAL
GROUP 1	112.20±25.26	95.26±14.16	89.39±18.06
GROUP 2	79.28±20.97	62.82±11.91	52.36±12.56
GROUP 3	66.03±25.63	45.25±11.08	40.89±7.89

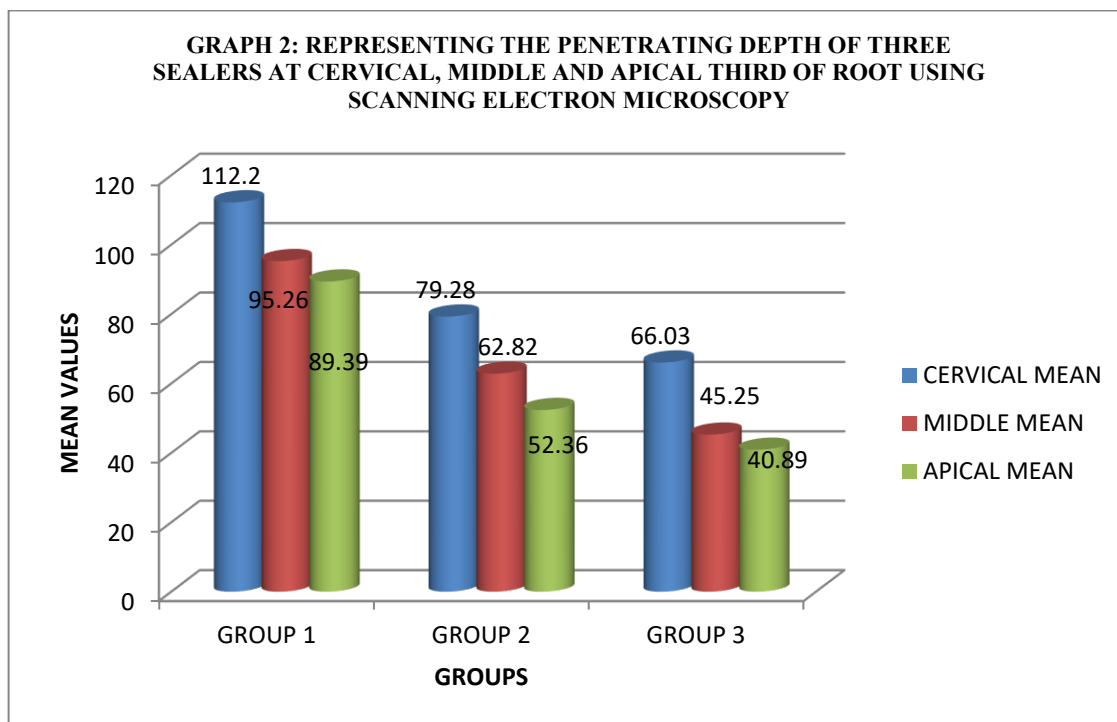


TABLE 6: REPRESENTING THE COMPARISON BETWEEN THE PENETRATING DEPTH OF THREE SEALERS AT CERVICAL THIRD OF ROOT USING SCANNING ELECTRON MICROSCOPY

GROUPS	DIFFERENCES	STANDARD ERROR	95% CI	T-TEST	DF	SIGNIFICANCE
GROUP 1 VS GROUP 2	-33.000	8.477	-50.3637 -15.6363	-3.893	28	0.000 (HS)
GROUP 2 VS GROUP 3	-13.250	8.550	-30.7647 4.2647	-1.550	25	0.03 (S)
GROUP 3 VS GROUP 1	46.170	8.550	28.6553 63.6847	5.400	28	<0.0001 (HS)

TABLE 7: REPRESENTING THE COMPARISON BETWEEN THE PENETRATING DEPTH OF THREE SEALERS AT MIDDLE THIRD OF ROOT USING SCANNING ELECTRON MICROSCOPY

GROUPS	DIFFERENCES	STANDARD ERROR	95% CI	T-TEST	DF	SIGNIFICANCE
GROUP 1 VS GROUP 2	-32.440	4.777	-42.2261 -22.6539	-6.790	28	<0.0001 (HS)
GROUP 2 VS GROUP 3	-17.570	4.200	-26.1735 -8.9665	-4.183	28	0.0003 (HS)
GROUP 3 VS GROUP 1	50.010	4.642	40.5006 59.5194	10.773	28	<0.0001 (HS)

TABLE 8: REPRESENTING THE COMPARISON BETWEEN THE PENETRATING DEPTH OF THREE SEALERS AT APICAL THIRD OF ROOT USING SCANNING ELECTRON MICROSCOPY

GROUPS	DIFFERENCES	STANDARD ERROR	95% CI	T-TEST	DF	SIGNIFICANCE
GROUP 1 VS GROUP 2	-37.030	5.680	-48.6647 -25.3953	-6.519	28	<0.001 (HS)
GROUP 2 VS GROUP 3	-11.470	3.830	-19.3149 -3.6251	-2.995	28	0.005 (S)
GROUP 3 VS GROUP 1	48.500	5.089	38.0764 58.9236	9.531	28	<0.0001 (HS)

DISCUSSION: Due to the compatibility with the root canal, a sealer can be used to minimize the inability of gutta percha to connect to the root dentin during the treatment of a root canal. Creating a three-dimensional seal using a sealer can prevent re-infection of the root canal and periapical tissues. The lack of complete filling of the root canal space accounts for nearly two-thirds of

unsuccessful root canal treatments. As a result, new materials that are suitable for root canal treatment have been developed.¹⁰ Among them are bioceramics are biocompatible, inorganic, nonmetallic materials with mechanical properties comparable to those of dental hard tissues. They interact well with the organic tissue, are chemically stable and non-corrosive in nature. By forming hydroxyapatite crystals, modern bioceramic sealers have a very strong bond with dentin walls.¹² Their excellent performance was based on the small size of the particle, low contact angle along with the deliquescent nature which warrants the cement to spread easily and evenly all along the dentinal walls of the canal including the inclusion into the lateral micro-canals. In addition to their corresponding active bio-silicate technology, bio-ceramic sealers exhibit a strong chemical bond to the root wall along with a significant expansion of 0.20%. Because of all these characteristics, these sealers are known to form a strong and efficient chemical bond with the dentinal wall without any gap.¹³ Schroeder first introduced epoxy resin-based sealers to endodontics, and current variations of the original formula are widely used today. Because of their reduced solubility, improved apical seal, and micro-retention to root canal dentin, epoxy resin sealers have been utilized. Epoxy resin is one of the sealers that are widely used in root canal filling because of its strength and resistance to adsorption.¹⁴ AH Plus sealers are commercially available as a two paste system which contains epoxide and amine as a base and a catalyst. Despite excellent bio-compatibility, tissue tolerance and long term dimensional stability of the material is sometimes overcome by the increased surface tension and shrinkage at sealer-dentin interface due to presence of silicon.¹⁵ The third sealer Sealapex differs from other root canal sealers as it contains calcium hydroxide as the main ingredient. This material has low solubility in tissue fluids and very low setting shrinkage.

In the present study, marginal adaptability and penetration depth of three sealers were evaluated and assessed using scanning electron microscopy. A highly statistically significant results were obtained when group 1, group 2 and group 3 were compared making all the sealers effective in providing a good hermetic seal with deeper penetration. Our study had similar results with that of the study conducted by Chen et al,¹⁶ Lee et al,¹⁷ Borges et al¹⁸ and Pius and Mathew.¹⁰ The interaction between the sealer type and the obturation methods had no effect on the degree of marginal adaptation or the depth of tubular penetration while its chemical composition can pose a significant impact on tubular penetration. The smear layer being an organic compound layer enclosed in mineral dentin plays a major role in the penetration of sealers, particularly in the apical region.¹⁹ Not only this a significant role is played by the experience, technique followed, amount and type of irrigate used by the operator.

LIMITATIONS: the major drawback being sample size followed by the type of technique used. Other techniques like computed CT, dyes, electrical methods, radio-isotope and fluid filtration techniques can be used in combination to confirm the validity of the tests. The only advantage of using scanning electron microscopy was the detection of defects at the sub-micron level while preserving the microphotographs of the specimens.

CONCLUSION: though with number of modifications present in the type of sealer, operator's technique remains the major factor for affecting the marginal adaptability along with the tubular penetration. Within the limitations, it can be concluded that the three sealers, Bioroot RCS, AH Plus and sealapex proved to be equally efficient and effective in reducing the micro-gaps while enhancing the tubular penetration.

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