ORIGINAL ARTICLE

Comparative analysis of microleakage associated with various endodontic sealers: An in-vitro study

ABSTRACT

Introduction: The purpose of the root canal obturation is to provide a hermetic seal, that inhibit the microleakage and canal reinfection, thus health of the periapical tissues can be preserved. It has been observed that the sealer used during endodontic therapy has a direct link with the longevity of the treatment and occurrence of post-operative pain.

Aim: The goal of present study was to assess microleakage in extracted teeth of four separate endodontic sealers (AH Plus, Sealapex, MTA Fillapex and Ceraseal) using dye penetration method.

Materials and Method: In this in-vitro study, 84 freshly extracted single rooted teeth were endodontically treated and obturated using four different sealers and divided into four groups alongwith two control groups based on the sealer used. Microleakage was checked in apical sections using dye penetration method under stereomicroscope.

Results: The maximum dye penetration was seen in Sealapex whereas least microleakage was shown by Ceraseal endodontic sealer.

Conclusion: Maximum microleakage was seen with calcium hydroxide based (Sealapex) while least microleakage was noticed in bioceramic based (Ceraseal) sealer group.

Keywords: Microleakage; Dye penetration; Bioceramic sealer, Resin sealer, Calcium hydroxide sealer.

INTRODUCTION

In endodontics, microleakage is indicated as clinically undetectable motion of ions, microorganisms, fluids and molecules between the tooth and obturation material or in spaces within the filling material. This long-term success of endodontic therapy is influenced by leakage as it lead to the failure of root canal treatment[1]. The main role of a root canal sealer is to fill irregularities and enhances adaptation of the obturating material to the root canal walls which prevents the possibility of leakage. Thus, improves the sealing capability and facilitates the retention of the core material. [2]

Muliyar S et al (2014)[1] stated that an endodontic sealer alongwith the core material provides a three-dimensional seal to the endodontic therapy that inhibits seepage and microleakage into the root canal space coming from periapical exudates, prevents reinfection, and make a beneficial biological environment for healing to take place.

Ballullaya SV et al. (2017)[2] compared the microleakage in six root canal sealers and found that microleakage was maximum in ZOE sealer and minimum in Endosequence BC sealer due to their chemical bond with dentin. Also, **Remy V et al. (2017)**[3] compared AH Plus, Endofill and MTA Fillapex sealer for their marginal adaptation and sealing ability and observed that AH Plus has better marginal adaptation compared to other sealers used.

Jasrotia A et al. (2021)[4] compared the apical sealing ability of three dissimilar endodontic sealers using dye penetration method under stereomicroscope and found that dye penetration was least for Ceraseal and Epiphany sealer and highest for AH Plus suggesting that better sealing were observed with Ceraseal bioceramic sealer and epiphany.

Mezaal ZS, Abdulkareem S and Shareef LG (2022)[5] evaluated the apical sealability of four different root canal sealers using a single cone technique by spectrophotometric analysis and found that TotalFill BC HiFlow showed highest mean values followed by AH Plus , GuttaFlow 2 and GuttaFlow bioseal.

There are various methods to evaluate the microleakage such as bacterial penetration, fluid transport, dye penetration or clarification but dye penetration is the most commonly used as it is relatively easier, faster and less complex than that of other techniques.

Since the apical seal, from irritants and bacteria, is an important determinant in the success of endodontic therapy, therefore, more research needs to be done on the sealing ability of the newly introduced sealers. The objective of the present study was to assess microleakage in extracted teeth using dye penetration, of different endodontic sealers (AH Plus, Sealapex, MTA Fillapex and Ceraseal).

MATERIAL AND METHODS

For this in-vitro study, freshly extracted teeth were obtained from Department of Oral and Maxillofacial Surgery, Sri Guru Ram Das Institute of Dental Sciences and Research, Sri Amritsar, to check the apical sealing ability of four different root canal sealers (AH Plus, Sealapex, MTA Fillapex and Ceraseal) using stereomicroscope.

SAMPLE SIZE CALCULATION

The sample size was calculated by using the following formula as suggested by Charan and Biswas (2013)[6]

n=2*Z1-α/2 2 X SD2 /d2

Z1-α/2=Power of the study

SD: Assumed standard deviation of the study variable

d: Mean value n=2*1.96*1.96*(0.15*0.15) / (0.157*0.157) = 7 in each group

Taking into consideration the various factors, the sample size for the study was increased to 14 in each group. The results were depicted in frequencies, percentage and mean±SD. The Chi-square test was used to compare categorical variables. The Unpaired t-test/One way analysis of variance (ANOVA) test had been used to compare continuous variables. The p-value <0.05 was considered significant.

The eighty four extracted teeth then were randomly divided into the following groups (FIGURE-I):

GROUP-1(n=14):- AH Plus sealer (Dentsply, Konstanz, Germany)

GROUP-2(n=14):- Sealapex (Sybron Endo, USA)

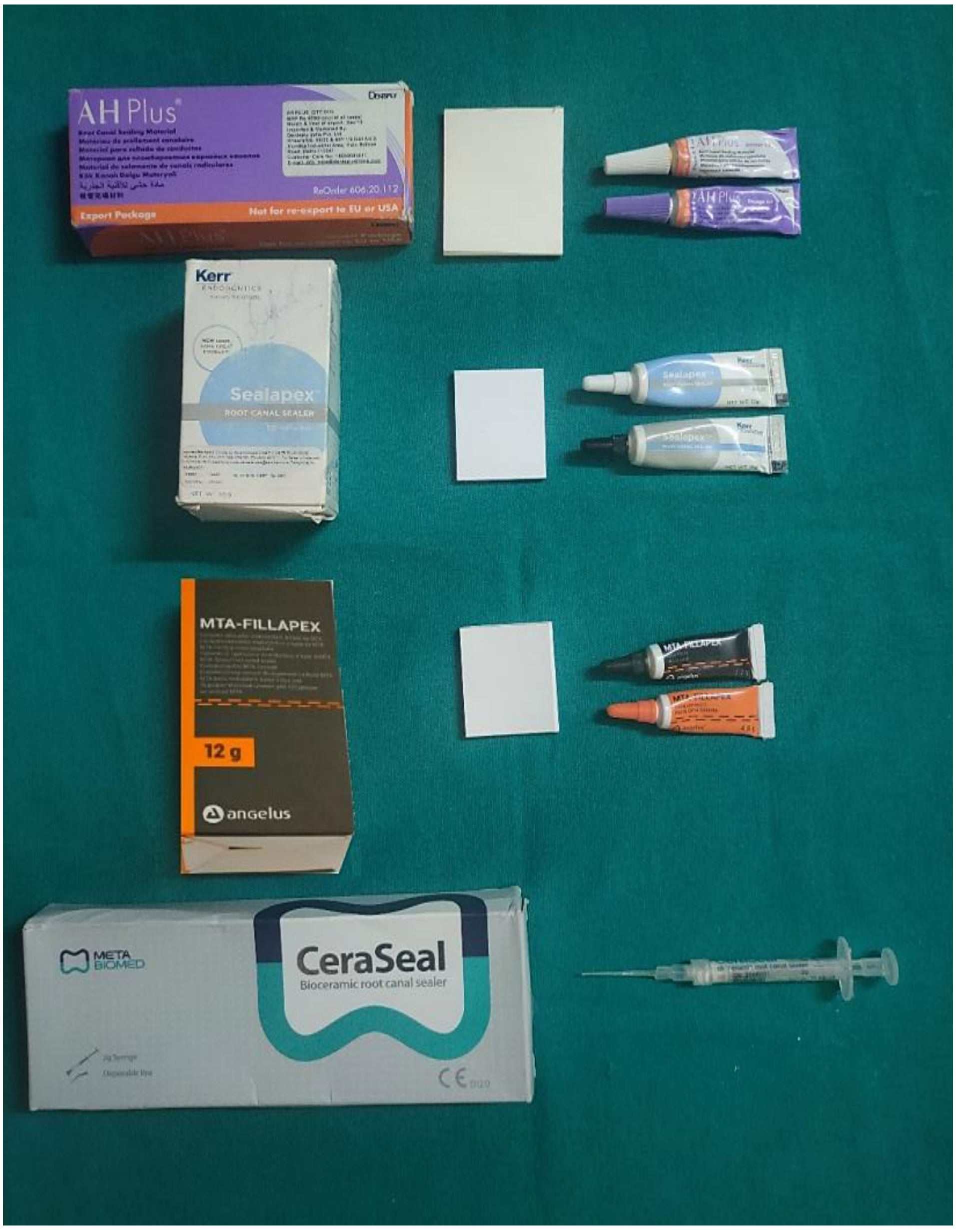
GROUP-3(n=14):- MTA Fillapex (Angelus, Londrina-Parana, Brazil)

GROUP-4(n=14):- Ceraseal (Meta Biomed Co., Cheongju, Korea)

GROUP-5(n=14):- Negative control group (access opening and biomechanical preparation was done but no obturation was done)

GROUP-6(n=14):- Positive control group (unprepared sound teeth in which the apical part was also coated with nail varnish)

FIGURE- I



SEALERS USED IN THE STUDY

INCLUSION CRITERIA:

1. Single rooted teeth with single root canal configuration.

2. Permanent teeth with complete root development.

3. Teeth free of cracks and restorations.

4. Single rooted teeth with degree of curvature between 10-20 degree.

EXCLUSION CRITERIA:

1. Previously root canal treated teeth

2. Teeth with evidence of resorption, craze lines, severe curvatures

3. Calcified canals

4. Teeth with root fracture

Procedure:

The extracted teeth were cleaned of organic debris and hard deposits were removed by an ultrasonic scaler. Teeth were then rinsed with distilled water to free from any soft tissue debris and stored in 10% formalin until used.

Endodontic Preparation:

Using a diamond disc mounted in a straight handpiece with micromotor, the teeth were decoronated at CEJ. After sectioning, 10 No. K-file (Dentsply Maillefer) was inserted into the canal until the tip of instrument was first visible at apical foramen and then working length was established by 15 No. K file (Dentsply Maillefer). The canals were prepared using rotary instrumentation (neo endo system) via crown down technique in the sequential order of 17/4%, 20/4%, 25/4%, 20/6% and 25/6%. After each instrument, canals were copiously irrigated using 30-gauge needle (Orikam Healthcare Pvt. Ltd., India) with 5.25% sodium hypochlorite solution (Parcan, Septodont Healthcare India Pvt. Ltd., India) and then 17% ethylenediamine tetra acetic acid gel (Endo-L, Maarc Dental, Maharashtra, India) by coating it over the endodontic file. Normal saline was used as the final irrigant (FIGURE-II).

FIGURE- II

ARMAMENTARIUM USED IN THE STUDY

(1)- Sodium hypochlorite; (2)-Chlorhexidine gluconate; (3)-Saline; (4)-2% Methylene blue dye; (5)-Neo endo rotary file system; (6)- NSK Endomotor; (7)-Micromotor; (8)-Spirit lamp; (9)- Lighter; (10)- LED Curing light; (11)-Applicator tip; (12)- Composite; (13)- Bonding agent; (14)-10 & 15 no. hand K file; (15)- Spreader; (16)-Nail varnish; (17)- Endogauge; (18)-EDTA gel; (19)-Paper points; (20)-2% gutta percha; (21)- 4% Gutta percha; (22)- BP blade and knife; (23)- Diamond disc mounted in straight handpiece; (24)- Irrigation needles

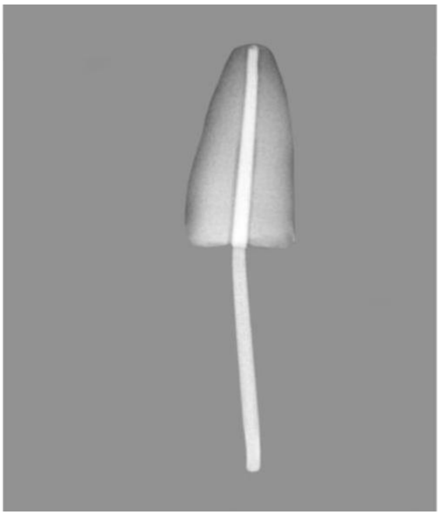
Obturation Technique:

After thorough cleaning and shaping, obturation with Cold Lateral Condensation Technique was done after canals were dried using paper points. The master gutta percha cone were selected according to the last file used at the working length which was confirmed by taking radiograph with dental RVG unit (Carestream) and its apical portion adjusted till its tug back was achieved. The apical half of primary gutta percha cone and the root canal walls were coated with sealers. Spreader was inserted alongside the primary cone one mm short of working length to compact the apical part of canal. This process was repeated with secondary gutta percha cones until the entire canal filled with a well condensed gutta percha (FIGURE-III).

FIGURE- III

III-a

III-b



III-c

III-d

III-e

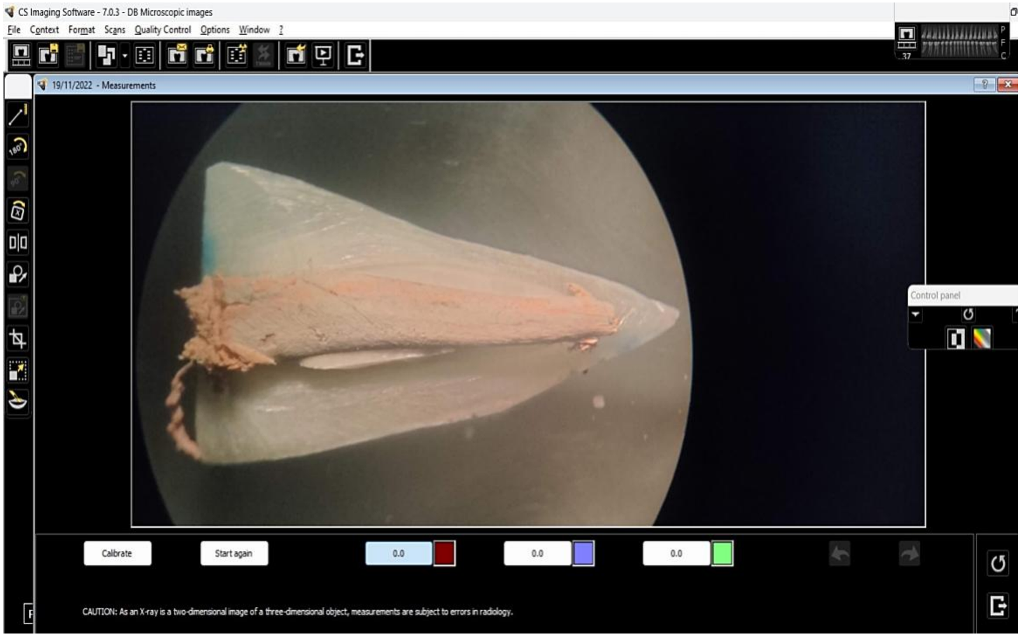
FIGURE-III (a): Sample decoronated at CEJ (b): Working length determination(c): Master Cone (d): Obturation (e): Application of nail varnish

Microleakage Evaluation:

The prepared samples were stored in a sealed container at 37^{o}C, 100% air humidity for 72 hours. Except for apical 2-3 mm, nail varnish was applied to the samples. The layer of varnish was allowed to dry and then immersed in dye for 72 hours, in different specimen containers. Samples were rinsed with running water and nail varnish was scrapped off with a surgical blade. All specimens were longitudinally sectioned in bucco-lingual direction using a diamond disc and straight hand-piece and the depth of dye penetration was analysed in millimetres. Microleakage was evaluated using dye penetration method (2% methylene blue) under stereomicroscope at 30X magnification (Motic, Hong-Kong) shown in FIGURE-IV and the length of dye penetration was evaluated in millimeters using CS imaging software (Carestream Dental LLC,NY) . The data calculated was sent for statistical analysis.

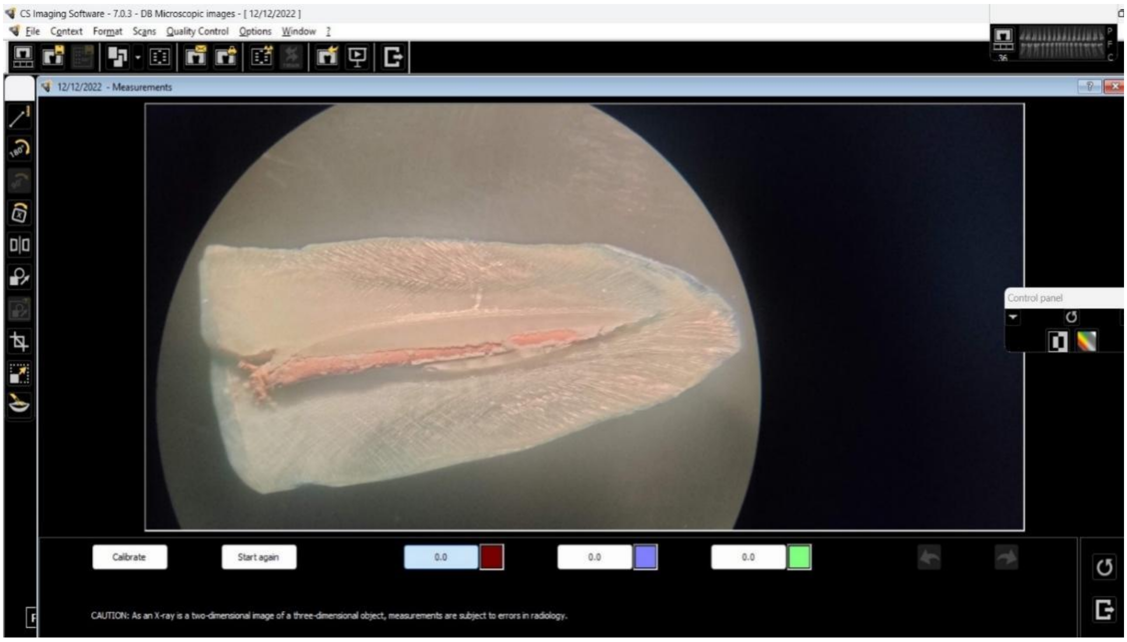
FIGURE- IV

FIGURE- V



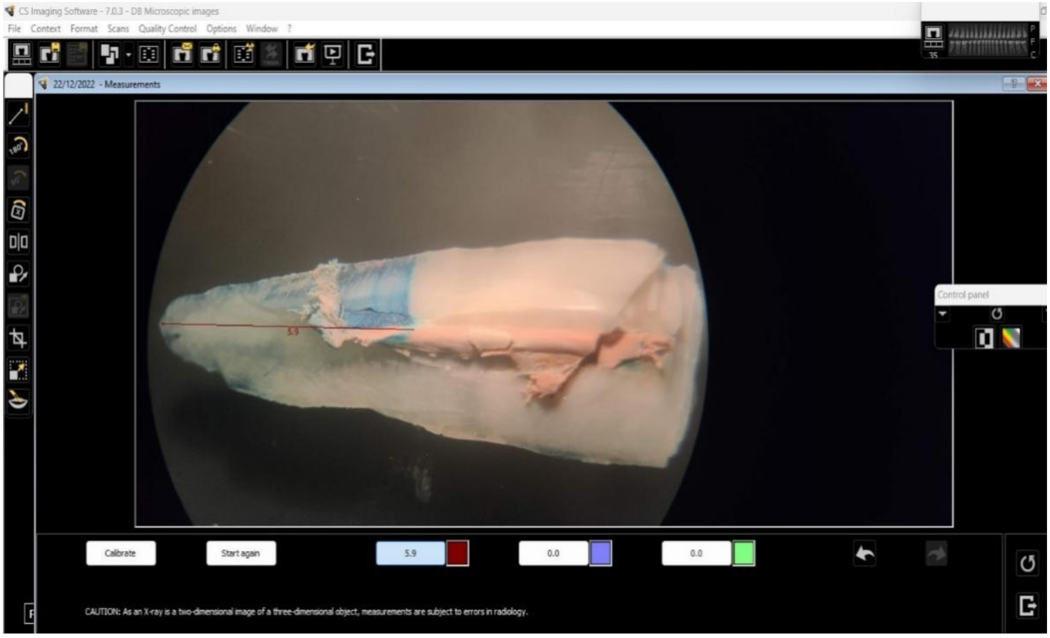
Group-I : Measurements of dye penetration in AH Plus samples showing maximum and minimum values.

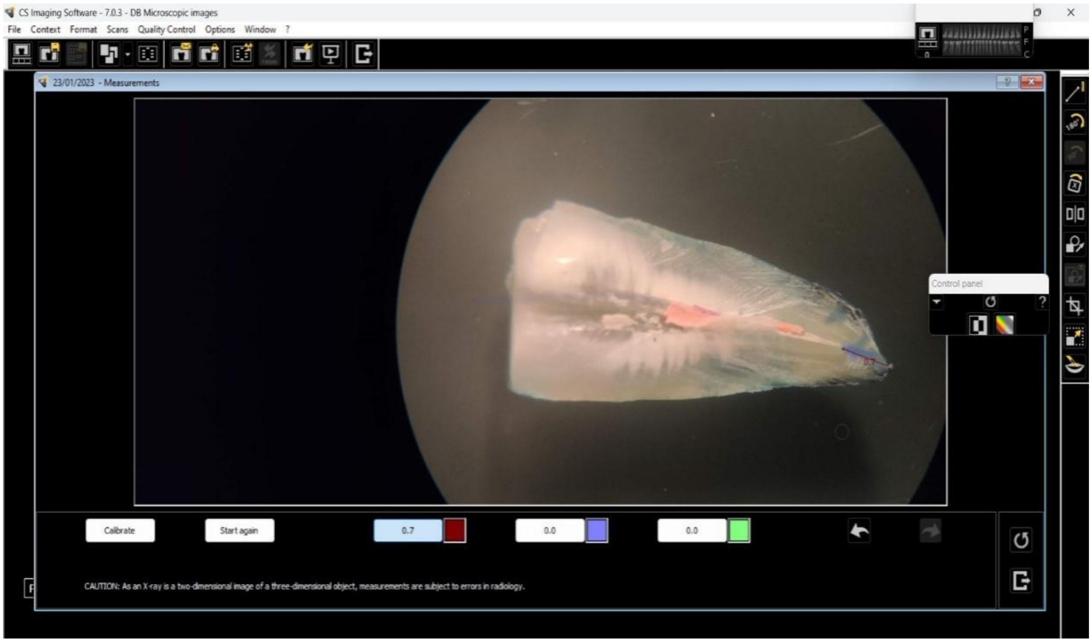
FIGURE- VI



Group-II : Measurements of dye penetration in Sealapex samples showing maximum and minimum values.

FIGURE- VII



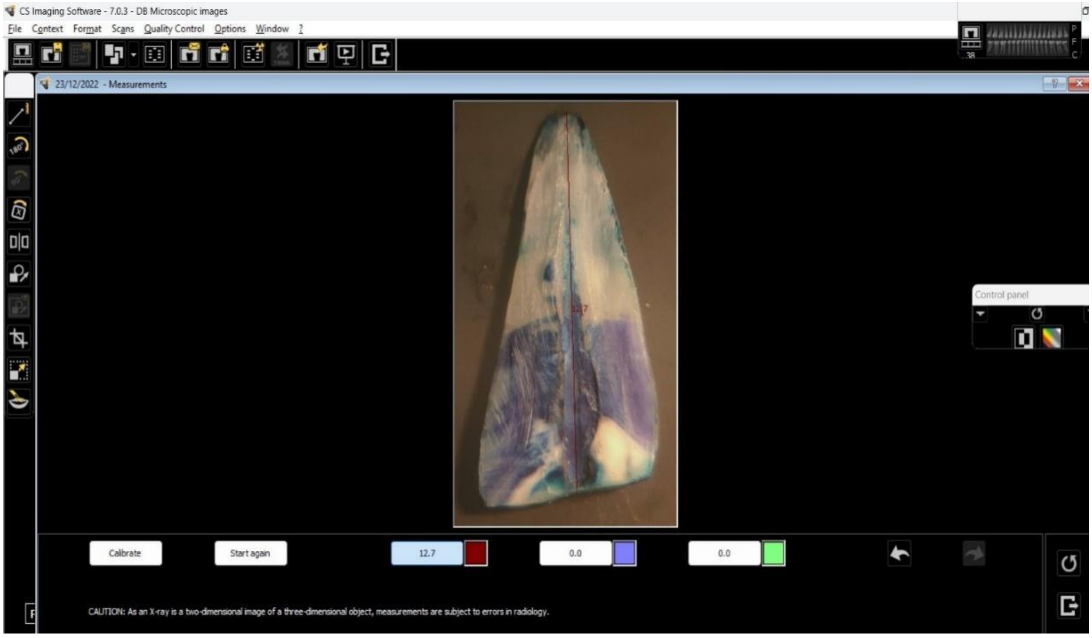


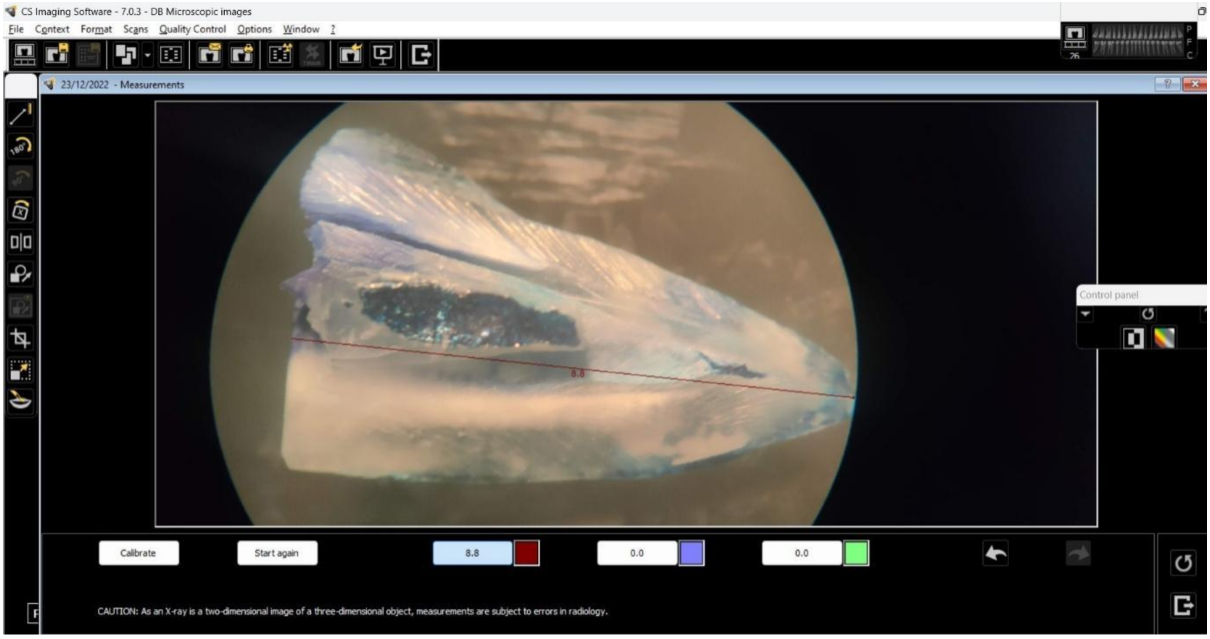
Group-III : Measurements of dye penetration in MTA Fillapex samples showing maximum and minimum values.

FIGURE- VIII

Group-IV: Measurements of dye penetration in Ceraseal samples showing maximum and minimum values.

FIGURE- IX





Group-V: Measurements of dye penetration in Negative Control Group samples showing maximum and minimum values.

FIGURE- X

Group-VI: Measurements of dye penetration in Positive Control Group samples showing maximum and minimum values.

RESULTS

1) Mean and standard deviation of depth of dye penetration for microleakage evaluation among different groups from Group-I to Group-VI was found to be 0.89, 3.13, 1.86, 0.82, 10.80 and 0.00 respectively.
2) Among various groups, Group-V showed the highest (10.8) mean depth of dye penetration. In regard to endodontic sealers used in the study, Group-IV (Ceraseal) showed the least dye penetration followed by Group-I (AH Plus), Group-III (MTA Fillapex) and Group-IV (Sealapex) as depicted in TABLE-1.
3) Statistical comparative analysis of mean dye penetration among various groups for evaluation of microleakage showed that the highly significant difference was seen between various groups with Group-V resulting in highest mean penetration in Group-V and significant difference was also seen in Group-II and Group-IV & between Group-I and Group-I as depicted in TABLE-2. (FIGURE-V-X).

TABLE-1: Mean and Standard Deviation (SD) of different groups for microleakage after dye penetration

Group	N	Mean	SD
Group-I (AH Plus)	14	0.893	1.43015
Group-II (Sealapex)	14	3.136	1.50721
Group-III (MTA Fillapex)	14	1.864	1.4026
Group-IV (Ceraseal)	14	0.828	1.0794
Group-V (Negative Control)	14	10.8071	2.1120
Group-VI (Positive Control)	14	0.000	0.000
Total	84	2.9213	2.0327

TABLE-2: Statistical comparative analysis of mean dye penetration among various groups

Comparison	Mean Difference	p-value
Group I vs II	2.243	0.001*
Group I vs III	1.021	0.881
Group I vs IV	0.064	1.000
Group I vs V	9.914	<0.001**
Group I vs VI	0.893	1.000
Group II vs III	1.221	0.386
Group II vs IV	2.307	0.001*
Group II vs V	7.671	<0.001**
Group II vs VI	3.136	<0.001**
Group III vs IV	1.085	0.673
Group III vs V	8.938	<0.001**
Group III vs VI	1.914	0.009*
Group IV vs V	9.978	<0.001**
Group IV vs VI	0.828	1.000
Group V vs VI	10.807	<0.001**

DISCUSSION

Root canal therapy not only aims to eradicate periapical inflammation and pathologies, but also to prevent the post-operative pain and its recurrence**. Muliyar S (2014)**[1] reported that endodontic failures that are due to inappropriate filling of root canal space accounts for about 60%, which further causes microleakage and hence failure.

Therefore, it's important to assess leakage tests for the excellence of the root canal treatment. In current study, microleakage evaluation has been done to check the sealing ability of endodontic sealers using dye penetration method with 2% Methylene Blue which is based on the linear measurement of the dye penetration which uses passive diffusion and capillary action in order to penetrate through any spaces between the canal walls and filling material[7]. Also, it is very sensitive, convenient, easy to use and dyes used are readily available. This stain is a standard material because it is economical, feasible and the identical size as organic products such as butyric acid produced by endodontic pathogen[8].

Another important consideration is the reduced effectiveness of smear layer removal techniques and irrigants in the apical region. Hence, in the present study, dye penetration from apical one-third of root was checked among the tested groups[9]. A digital software (Carestream Dental LLC,NY) was used to measure the amount of dye penetration in order to further decrease the chances of operator error.

The sealers used in this study belong to different categories due to which overlapping of results was avoided.

AH Plus (0.893mm) performed significantly better than Sealapex (3.136mm) with less depth of dye penetration due to its low solubility, dimensional stability and adhesiveness of the sealer that provide sufficient seal and adequate sealing ability than that of Sealapex group[10].

More microleakage was seen in specimens of MTA Fillapex than that of AH Plus (TABLE-1). Nevertheless, statistically insignificant difference (p-value=0.881) was found between the two groups (TABLE-2) due to the increased adhesiveness and adaptation to the canal walls of both the groups[11].

In the current study, Ceraseal, a calcium silicate bioceramic sealer, performed slightly better (0.828mm) than AH Plus (0.893mm) with statistically insignificant difference (p-value=1.00) between the two groups. This could be due to the similar water sorption and solubility of both the sealers that seemed to correlate with their sealing performance. So, it can be mentioned that both the sealers are comparable to each in terms of microleakage as both of them has minute particle size and can flow readily into the dentinal tubules[12].

However, contrasting results were seen in a study done by Asawaworarit W et al (2019)[13] 'who stated that bioceramic sealers have better sealing ability than epoxy resin based sealer due to the hydrophillic nature, small particle size and high flowability of these sealers that allows for greater penetreation into the dentinal tubules.

Highest mean dye penetration for microleakage (3.136mm) was seen with Sealapex endodontic sealer and lowest in Ceraseal (0.828mm) with significant difference between the two groups (TABLE-2). It was deduced that bioceramic sealer (Ceraseal) due to their small particle size and early formation of hard calcific barrier showed less microleakage than calcium hydroxide based sealer (Sealapex) [14].

Another conclusion can be drawn from results of the present study was that MTA Fillapex performed similar to Sealapex (TABLE-1) due to the ability of both the sealers to produce hard tissue formation by releasing calcium ions which further favoured the sealability[15].

Also, no significant difference was seen in dye penetration between Ceraseal and MTA Fillapex group due to their comparable viscosity and flow to penetrate the canal irregularities alongwith their property to form hard tissue that resulted insignificant difference between the two groups in terms of their sealing ability[16].

Therefore, the least microleakage found in bioceramic based sealers , in our study, was attributed to their small particle size, hydrophillic nature, high flowability and ability to leave least marginal gaps makes them ideal to use as an endodontic sealer[2,17].

LIMITATIONS OF THE STUDY

1. There were chances that operator bias could be seen in regard to sealer placement.
2. All the clinical conditions like presence of moisture, change of temperature, presence of variable masticatory forces etc. could not be simulated in an in-vitro study.

CONCLUSION

With reference to microleakage among sealers used in the study, Sealapex showed maximum (3.136) and Ceraseal (0.828) the least mean dye penetration depicting highest and lowest microleakage respectively. However, statistically significant difference was seen only between Sealapex & AH Plus and Sealapex & Ceraseal whereas other groups showed no significant differences. It was concluded that the apical sealing ability of AH Plus was almost similar to MTA Fillapex sealer, Ceraseal has the best and Sealapex the worst sealing ability.

Authors Contribution

This research work was completed by the equal contribution of all the authors and all the authors have read and agreed to the published version of the manuscript.

Funding

No external funding was received for this research.

REFERENCES

1) Muliyar, S, Shameem, KA, Thankachan, RP, Francis, PG, Jayapalan, C.S. and Hafiz, KA 2014, 'Microleakage in endodontics', Journal of International Oral Health, vol.6, no.6, pp.99-104.
2) Ballullaya, SV, Vinay, V, Thumu, J, Devalla, S, Bollu, IP and Balla, S 2017, 'Stereomicroscopic dye leakage measurement of six different root canal sealers', Journal of Clinical and Diagnostic Research, vol.11, no.6, pp.ZC65-68.
3) Remy, V, Krishnan, V, Job, TV, Ravisankar, MS, Raj, CR and John, S 2017, 'Assessment of Marginal Adaptation and Sealing Ability of Root Canal Sealers: An in vitro Study', The Journal of Contemporary Dental Practice, vol.18, no.12, pp.1130-34.

4) Jasrotia, A, Bhagat, K and Sharma, N 2021, 'Comparative evaluation of sealing ability of bioceramic sealer, AH plus and epiphany sealer: an in vitro study', Journal Advanced Medical Dental Sciences and Research, vol.9, no.4, pp.121-24.

5) Mezaal ZS, Abdulkareem, S and Shareef, L.G 2022, 'In vitro apical microleakage evaluation for different endodontic sealers by spectrophotometric analysis: an observational study' *F1000Research*, vol.11, no.1396, p.1396.

6) Charan, J and Biswas, T 2013, 'How to calculate sample size for different study designs in medical research?', Indian Journal of Psychological Medicine, vol.35, no.2, pp.121-26.

7) Raghuvanshi, R, Dubey, M.S.S.D.C. and Pushkar, AP 2018, 'A Comparative Evaluation of the Apical Microleakage in Root Canals Sealed with Bioceramic, Resin Based and Glass Ionomer Root Canal Sealers: An Ex-Vivo Study', Journal of Research and Advancement in Dentistry, vol.8, no.3, pp.46-55.

8) Al-Kadhi, A.M.A.A, Al-Ani, Z.B.M.A. and Al-Eanizi, J.A.J 2019, 'Comparison of apical microleakage of four contemporary endodontic sealers by dye penetration method', International Medicine Journal, vol.26, pp.237-40.

9) Gutmann, J.L. and Fan, B 2016, Cohen's Pathways of the Pulp, Elsevier publications, St. Louis.

10) Lone, MM, Khan, FR and Lone, M.A 2018, 'Evaluation of microleakage in single-rooted teeth obturated with thermoplasticized gutta-percha 127 using various endodontic sealers: an in-vitro study', Journal of the College of Physicians and Surgeons Pakistan, vol.28, no.5, pp.339-42.

11) Meidyawati, R and Suprastiwi, E 2017, 'Comparison sealing ability of MTA sealer and resin epoxy sealer', Journal of International Dental and Medical Research, vol.10, no.1, pp.134-38.

12) Hasnain, M, Bansal, P and Nikhil, V 2017, 'An in vitro comparative analysis of sealing ability of bioceramic-based, methacrylate-based, and epoxy resin-based sealers', Endodontology, vol.29, no.2, pp.146-50.

13) Asawaworarit, W, Pinyosopon, T and Kijsamanmith, K 2020, 'Comparison of apical sealing ability of bioceramic sealer and epoxy resin-

based sealer using the fluid filtration technique and scanning electron microscopy', Journal of Dental Sciences, vol.15, no.2, pp.186- 92.

14) Lin, G.S.S., Ghani, N.R.N.A., Noorani, TY and KAMARUDİN, A 2020, 'Apical sealing ability of different endodontic sealers using glucose penetration test: a standardized methodological approach', Cumhuriyet Dental Journal, vol.23, no.2, pp.79-87.

15) Jain V, Chawdhary P, Kaushik M, Roshni R, Mehra N 2017, 'Qualitative and quantitative analysis of apical microleakage of different endodontic sealers', International Journal of Advanced Research, vol.7, no.6, pp.476-81.

16) Zhou, HM, Shen, Y, Zheng, W, Li, LI, Zheng, YF and Haapasalo, M 2013, 'Physical properties of 5 root canal sealers', Journal of Endodontics, vol.39, no.10, pp.1281-86.

17) Muharsya, Y, Usman, M and Suprastiwi, E, 2017, 'Comparison sealability of root canal obturation using bioceramic sealer and methacrylate resin-based sealer', Journal of Physics: Conference Series, vol. 884, no. 1