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Comparative Evaluation of Apical Sealing Ability of Hydrophilic and Hydrophobic Obturating Materials -A Sem Study

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KEYWORDS
Sealers, Hydrophobic, Hydrophilic, Apical seal, obturation

ABSTRACT:

AIM- This in-vitro study was designed to evaluate the sealing ability of hydrophilic and hydrophobic obturation materials in the apical one third of the root canal which is the most vulnerable area to microleakage using scanning electron microscope.

METHODOLOGY- A total number of 60 undamaged human mandibular premolar teeth with single root canal and apical foramen were included in this study The root canals were prepared till WaveOne Gold medium medium file i.e 35 size which is of 6% and irrigated with 5 mL of 5.25% sodium hypochlorite before placement of 4 different kinds of sealer using GP cones. The sealers used were namely- Group A - Hydrophobic sealer (AH Plus),Group B – Hydrophobic sealer (GuttaFlow 2), Group C – Hydrophilic sealer (Bio-C), Group D.- Hydrophilic sealer (BioRoot RCS) . Superficial longitudinal grooves were created on the mesial and distal wall of the root with diamond disc and each specimen was evaluated at the level of 3 mm from the apex and evaluation of microgap was done by examining under scanning electron microscope in micrometer(µm)

RESULTS - The result revealed that hydrophilic sealers, BioRoot RCS (Group D) and BIO-C (Group C) showed less microgap when compared with hydrophobic sealers, Group A (AH Plus) and Group B (GuttaFlow 2).

CONCLUSION-To achieve hermetic seal various sealers have been used but the recently introduced hydrophilic sealers provide a better seal as compared to traditionally present hydrophobic sealers as proved under the experimental conditions and within the limitations of this study.

1. Introduction

The success of root canal treatment depends on various factors such as proper diagnosis, adequate biomechanical preparation, complete 3-D obturation of the root canal and post endodontic restoration.[1,2] Ingle et al reported about 58 % of root canal failures due to incomplete obturation of the root canals.[3]

According to study conducted by **Mozami F et al** presence of voids in obturated canal can lead to endodontic failures as it acts dead space which lead to growth of microorganism.[4]

Therefore, thorough debridement of the root canal system should be done to eliminate pathogenic organisms. The objective of obturation is to provide fluid tight hermetic seal of the root canal system. The success of endodontic therapy depends on the effectiveness of apical seal.[5]

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"Root canal sealers are used with biologically acceptable semisolid or solid obturating materials to establish an adequate seal of the root canal system."[7] The effectiveness of root sealers is improved by reducing the amount of sealer to be used and which has good adaptation and sealer's penetration into root dentin.[8]

Microleakage is main cause for root canal failure which is due to bad contact in between dentin and sealer, sealer and gutta percha and it can be due to presence of voids in the sealer. Therefore, adaptation of sealer with dentin in an important factor which influences reinfection of canal and microleakage.[9]

There are various functions of sealers which are it fill the voids and irregularities in the root canal, lateral and accessory canals, it forms a bond between core material and root canal wall and to fill spaces between gutta-percha points used in lateral condensation and it also act as lubricant and it facilitates placement of core material.

Traditionally sealers available in the market were hydrophobic in nature resulting in improper bonding with the dentinal wall. Hydrophobic sealers available in the market are epoxy resin based sealers and silicone based sealers. Example of the epoxy resin based sealer is AH Plus and silicone based is guttaflow 2

Drawbacks of hydrophobic sealers are: - 1) Hydrophobic sealers which were used earlier did not bond to tooth, therefore, there were chances of microleakage leading to failure of root canal treatment.2) There is less penetration of root canal sealer when compared with hydrophilic sealer. Therefore, recently new sealers are developed which are hydrophilic. Hydrophilic sealer utilizes moisture within canal to complete the setting reaction and it expands laterally thus providing a better adaptation and seal.

The advantages of hydrophilic sealers are they provide better resistance to bacterial leakage, better adaptation and seal in the root canal, biocompatible and improved sealer to root dentin bonding. Disadvantages of hydrophilic sealers are they are difficult to remove when they are set from root canals specially in cases of post and core and retreatment cases and these sealers are comparatively expensive.[6]

Hydrophobic sealers used in the study were as follows:

AH Plus which is resin-based sealer with good dimensional stability and have property of expansion. It sets by polyaddition reaction of diamines present in the composition.[8] It is considered the 'gold standard' of a root canal sealer because: 1) it has high bond strength to dentin, 2) radiopaque, 3) dimensional stability 4) flow 5) Low solubility. [10]

Guttaflow 2 is an improved adaptation of RoekoSeal, as it is a self-curing material, also having cold-flowable property. It is composed of gutta-percha powder having less than 30 μm particle size, nanosilver particles and polydimethylsiloxane. The nano-silver in the material which impart bacteriostatic property in preventing further spread of bacteria and is biocompatible.

The two hydrophilic sealers used in the study are as follows:

Bioceramic based root sealer interact with dentin as there is chemical uptake of calcium and silicon which occurs in presence of phosphate buffer solution. This interaction is known as "**mineral infiltration zone**" which is caused by the caustic effects of the sealer's by-products which are alkaline in nature and the penetration of minerals consisting of calcium, carbonate and silica into intertubular dentin as a result of denaturation of the collagen fibers.[17]

Bioceramic sealers have advantages such as 1) cytotoxicity is lower, 2) antimicrobial activity is excellent because it has high pH value, also hard tissue formation is promoted and hydroxyapatite layer is formed. [18]

BioRoot RCS a recently introduced bioceramic material containing powder/liquid and is a tricalcium silicate-based sealer. It is available in market since 2015. BioRoot RCS is stated to produce angiogenic and osteogenic growth factors in vitro samples by human periodontal ligament cells. It also has lower cytotoxicity when compared with other conventional root canal sealers as conducted in the study by **Lin et al**.[19] is a powder/liquid hydraulic tricalcium silicate-based cement (Gilles & Oliver 201 2) marketed since February 2015 and recommended for single-cone technique or co ld lateral condensation root filling. The powder contains tricalcium silic ate,povidone and zirconium oxide; the liquid is an aqueous solution of calcium chloride and polycarboxylate. BioRoot RCS has been reported to induce BioRoot RCS has been reported to induce in vitro the production of angiogenic and osteog enic growth

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factors by human periodontal ligament cells (Camps et al. 2015); moreover, it has a lower cytotoxicity than other conventional root canal sealers, may induce hard tissue deposition (Dimitrova Nakov et al. 2015, Pr€ullage et al. 2016) and has antimicrobial activity.

Bio-C Sealer is a recently introduced bioceramic sealer which is premixed and it can be used for root end filling, perforation repair and also as sealer in the root canal treatment. It is available in single syringe, and is composed of calcium silicates, calcium oxide, calcium aluminate, zirconium oxide, silicon dioxide, iron oxide and dispersing agents. Its bioactivity is because of the release of calcium ions which stimulate the formation of hard tissue.[18]

Various methods which can be used for evaluating the apical sealing of root canal sealers are as follows**;** dye penetration, fluid filtration, dye extraction or dissolution method, bacteria and toxin infiltration method, air pressure method, electrochemical method, neuron activation method, radioisotope method, metal solution tracers, reverse diffusion method, artificial caries and three-dimensional method. In addition, other methods can also be used such as: Scanning electron microscopy, Transmission electron microscope and Micro-computed tomography.[24]

A Scanning Electron Microscope (SEM) is a powerful magnification device that utilizes focused beams of electrons to obtain information. SEM gives the detailed three-dimensional and topographical imaging and the information gathered from different detectors
Thus, this in-vitro study was designed to evaluate the sealing ability of hydrophilic and hydrophobic obturation materials in the apical one third of the root canal which is the most vulnerable area to microleakage using scanning electron microscope.

Clinical Significance

Root canal sealers should be used with biocompatible obturating materials for hermetic seal of the root canal. They are important as they block the dentinal tubules of the radicular dentin and decreases the entrance of microorganisms in the root and thus aids in successful root canal therapy.

Methodology

This in-vitro study was conducted in the Department of Conservative Dentistry and Endodontics, Maharishi Markandeshwar College of Dental Sciences and Research, Mullana, Ambala.

Procedure

A total number of 60 undamaged human mandibular premolar teeth with single root canal and apical foramen were included in this study. Undamaged mandibular premolar teeth with single root canal, fully developed root apices, teeth should be free from caries, resorption or root fracture, teeth free from restoration and teeth with straight canals.

Teeth with any calcification, resorption, extra canals and curvature of the root canal, internal and external resorptions, root caries, root fracture, cracks, craze lines, cervical abrasion, old restoration and open apices.

Sterilization of Samples

Extracted teeth were collected, stored, sterilized and handled following the guidelines of OSHA) and CDC recommendations and guidelines.

Preparation of samples- Single rooted teeth with single patent canals were used and decoronated with diamond disc in order to establish same standard root length of 14mm (Fig. 1) .

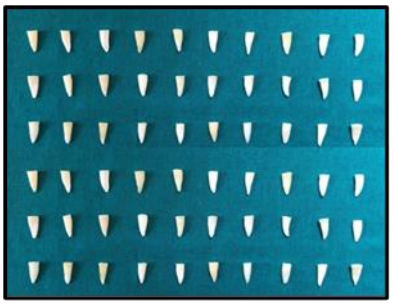

Fig. 1 Preoperative radiographs (RVG) were taken and working length was taken with #10 K- FILE. Glide path was prepared using ProGlider which is of size 16 and 2% taper. (Dentsply Mailifer, Switzerland). Root canal cleaning and shaping was done using WaveOne Gold (Dentsply, Maillifer). The root canals were prepared till WaveOne Gold medium medium file i.e 35 size which is of 6% and irrigated with 5 mL of 5.25% sodium hypochlorite solution in between the use of each file.

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Before placement of sealer in the sealer, gutta-percha cone which was fitting properly and also gave a tug-back at working length was selected. **(Fig. 2)**

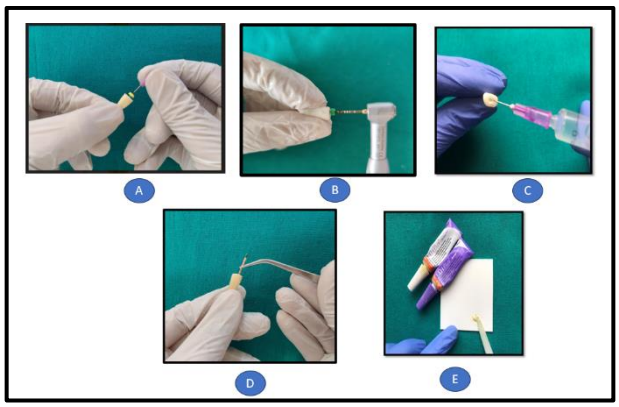


Fig. 2- Preparation of samples A to E.

Measurement of microgap

- Superficial longitudinal grooves were created on the mesial and distal wall of the root with diamond disc using a slow handpiece
- Using a chisel and mallet, teeth was split into two halves and the buccal and lingual halves were separated.
- Each specimen was evaluated at the level of 3 mm from the apex and evaluation of microgap was done by examining under scanning electron microscope
- Maximum gap was recorded in micrometer(µm).
- All specimens were kept in ethanol for dehydration and percentage was increased incrementally from 70%, 80%, 90%, 95%, 100%) for enhancing the image performance in the Scanning electron microscope. High vacuum conditions were used for drying of wet samples quickly.
- Sputtering was done in gold in vacuum coater to make electrically conductive surface and to prevent from charge buildup on specimen surface which is essential for SEM in thickness of 10 nm and it was done for 1 minute. 15 kV high accelerating voltage was used and magnification used in the study was 500x. **Fig. - 3**

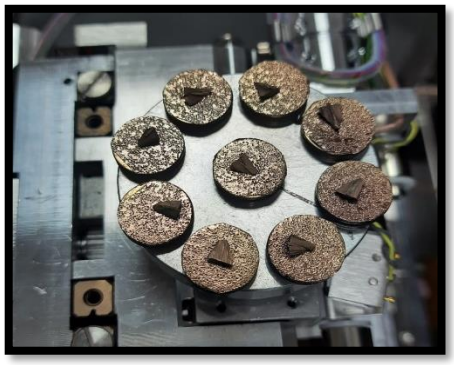
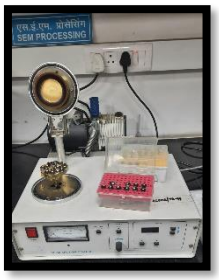


Fig 3- Gold sputtering of the samples

- SmartSEM software was used to analyze the images. In this Smartbrowse software was used which is patented by Zeiss for post imaging acquisition. They both facilitated complete understanding of multiple images in terms of imaging and space parameters.
- Smartstitch software is used for acquisition of image and stitching the software and used for large area mapping as it takes multiple images in series and create seamless montage. **(Fig. 4 to 8)**

Fig. - 4

4.



Statistical analysis-The data was collected and subjected to further statistical analysis and the results were concluded based on the statistics. Statistical analysis was performed by means of One-Way ANOVA test and post-hoc Tukey's test.A p-value of 0.0001 was set for statistical significance.

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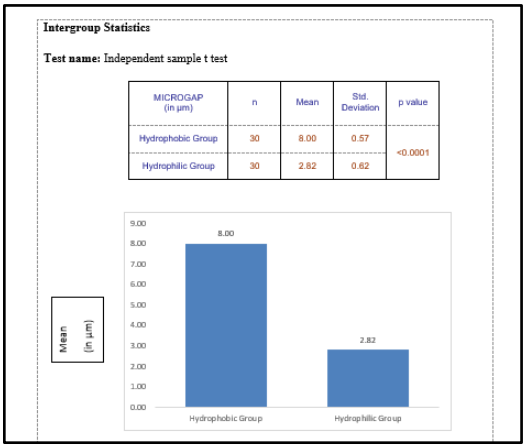
Results and Observations

- The present in vitro study was undertaken to study the apical microgap present after obturation hydrophilic and hydrophobic obturation system using scanning electron microscope.
- A total of 60 extracted human mandibular premolar teeth with length 14 mm were taken to conduct this in vitro study.
- Samples were divided into 4 groups (n=15) according to type of sealers used.
- Group A: AH Plus
- Group B: GuttaFlow 2
- Group C: Bio-C
- Group D: BioRoot RCS

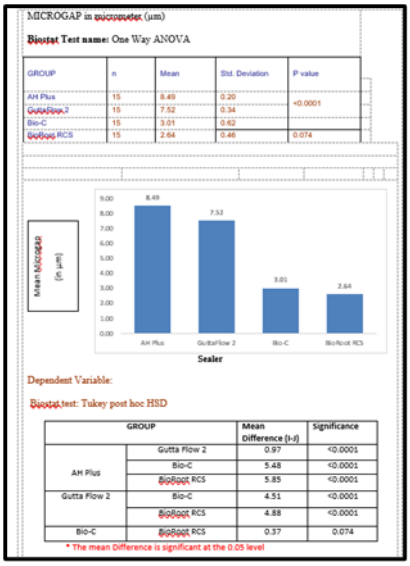
Master chart (in µm) (Adaptation of sealer with Gutta percha) – **TABLE 1**

S.No.	AH Plus	GuttaFlow 2	Bio-C	BioRoot RCS
1	8.63	8.26	2.75	3.75
2	8.25	7.4	3.2	2.06
3	8.75	7.22	2.8	3.01
4	8.25	7.61	2.96	2.63
5	8.65	7.82	3.12	3.19
6	8.62	8.1	3.18	2.65
7	8.4	7.05	3.03	3.15
8	8.18	7.63	2.69	2.2
9	8.34	7.36	2.86	2.32
10	8.32	7.5	3.22	2.56
11	8.81	7.42	3.3	2.61
12	8.38	7.28	2.78	2.4
13	8.46	7.26	2.81	2.36
14	8.58	7.16	3.26	2.46
15	8.71	7.68	3.19	2.18



Graph 1 showing mean value intergroup comparison between hydrophilic and hydrophobic groups

When intergroup comparison was done between hydrophilic and hydrophobic group, the mean value was 8µm for the hydrophobic group and 2.82µm for the hydrophilic group and the result was statistically significant with p value of less than 0.0001.



Graph-2 Showing mean difference values between different groups

Group A: AH Plus

By comparing Group A and Group B the mean difference is 0.97 which is statistically significant ($p<0.0001$) with Group A (8.49) having higher microgap value between sealer and dentin wall than Group B (7.52).

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By comparing Group A and Group C the mean difference is 5.48 which was statistically significant ($p<0.0001$) with Group A (8.49) having higher microgap value between sealer and dentin wall than Group C (3.01).

By comparing Group A and Group D the mean difference is 5.85 which was statistically significant ($p<0.0001$) with Group A (8.49) having higher microgap value between sealer and dentin wall than Group D (2.64).

Group 2: GuttaFlow 2

By comparing Group B and Group C the mean difference is 4.51 which was statistically significant ($p<0.0001$) with Group B (7.52) having higher microgap value between sealer and dentin wall than Group C (3.01).

By comparing Group B and Group D the mean difference is 4.88 which was statistically significant ($p<0.0001$) with Group B (7.52) having higher microgap value between sealer and dentin wall than Group D (2.64)

Group 3: Bio-C

By comparing Group C and group D the mean difference is 0.37 which was statistically non-significant ($p=0.074$) with Group C (3.01) having higher microgap value between sealer and dentin wall than Group D (2.64).

Inference of Results

The result revealed that hydrophilic sealers, BioRoot RCS (Group D) and BIO-C (Group C) showed less microgap when compared with hydrophobic sealers, Group A (AH Plus) and Group B (GuttaFlow 2). The intergroup comparison was done with mean value of 2.82 µm of hydrophilic group which was lesser when compared with mean value 8µm of hydrophobic group.

Amongst all the groups, AH Plus (Group A) showed maximum microgap when compared with other groups and BioRoot RCS (Group D) showed minimum microgap and the difference in between all the groups was statistically significant except between Group C and Group D which was not significant.

According to results obtained the order of mean values of microgap (in ascending order) observed were as follows:

GROUP D < GROUP C < GROUP B < GROUP A

(2.64 µm) (3.01 µm) (7.52 µm) (8.49µm)

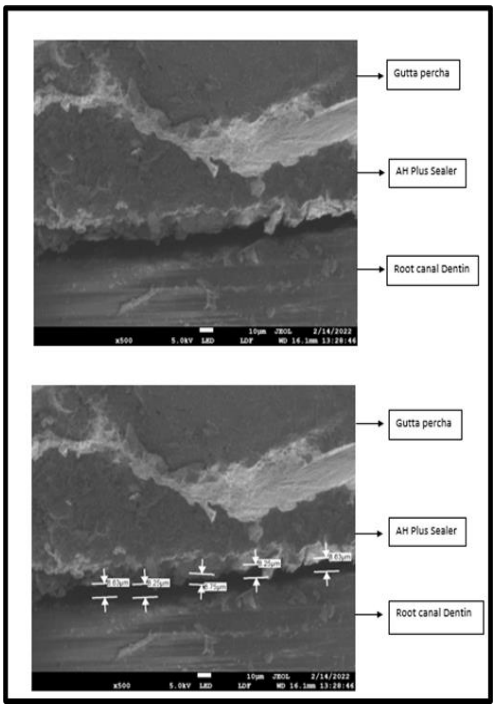

Fig.5 SEM image showing microgap in AH Plus group

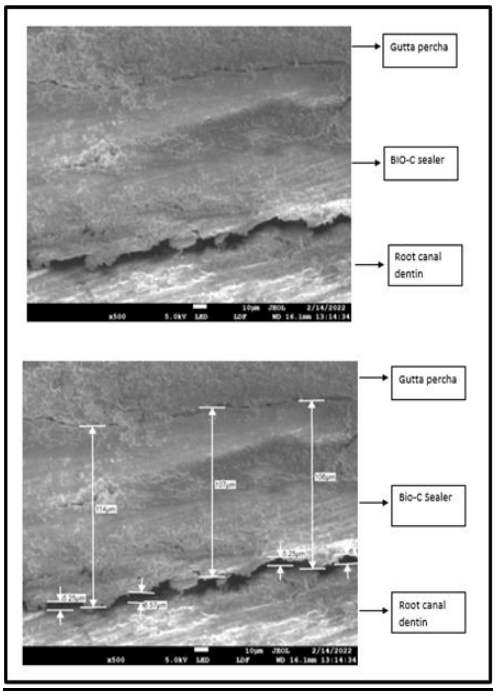

Fig.6 SEM image showing microgap in GuttaFlow 2

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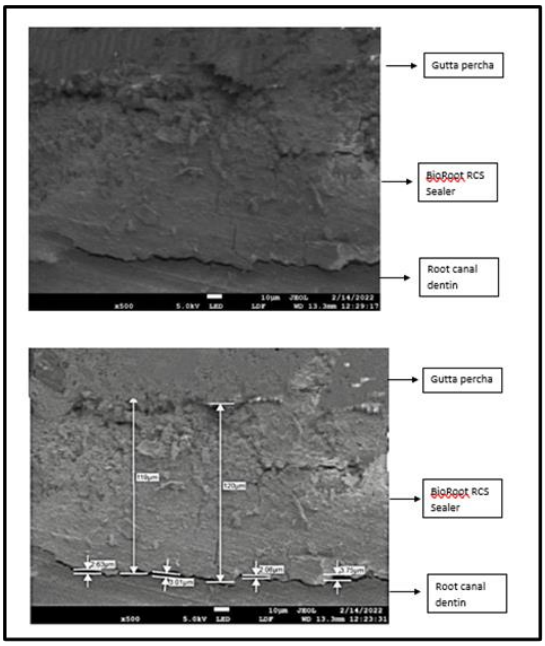

Fig.7- SEM image showing microgap in BIO-C sealer

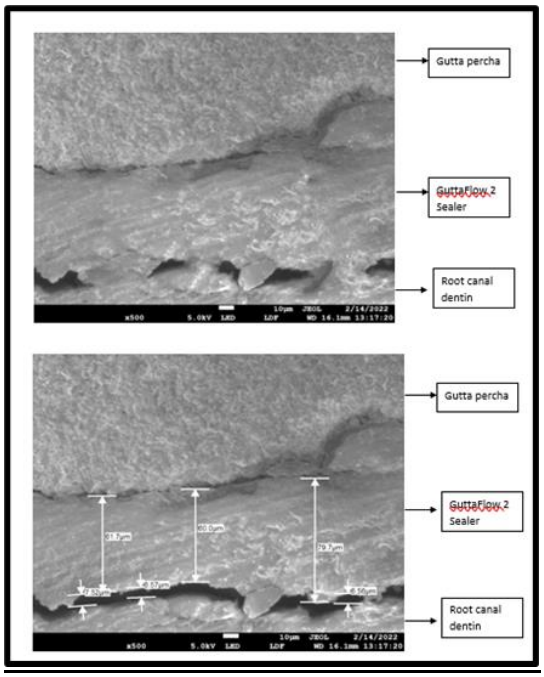

Fig.8- SEM image showing microgap in BIOROOT RCS

Discussion

Microorganisms are the main reason for causing pulpal and periradicular pathology. Once the bacteria have invaded the pulp which is the heart of the tooth, the damage is almost irreparable. W.D Miller in 1890 confirmed the presence of bacteria in pulpal and periapical infection.

The main objective of root canal treatment is to stop further infection or reinfection by eradicating microbes within the root canal system. For a root canal treatment to be successful thorough knowledge of both tooth anatomy and root canal morphology are required.[55] Obturation of the root canal is the key factor in achieving success in endodontic treatment. It refers to three-dimensional sealing of canal involving gutta percha points and root canal sealers. The goal of obturation is to stop the recontamination of the canal either from the residual microbes or from new microbes entering from lateral accessory canals.

Root canal sealers are needed to fill in the voids between the gutta-percha cones as well as to seal irregularities between the gutta-percha cones and root canal dentinal walls. Sealers form a physical barrier by penetrating inside dentinal tubules thereby increasing retention of the filling material. Sealers play many important role in obturation as it has antibacterial property, acts as lubricating agent, has good sealing capability, biocompatibility, bond strength and radio-opacity.

Depth of penetration of sealer depends upon several factors such as removal of smear layer, permeability of dentinal tubules (as it depends on diameter and number of tubules), chemical and physical properties of the sealer.[60]

Size of particle is an important factor in flow characteristics of a sealer and it is inversely proportional to flow of the sealer. If flow is less, then there are unfilled irregularities within the root canal wall and in between core material and root canal wall, higher flow rates may lead to extrusion of sealer from the apical foramina which is undesirable for the success of root canal treatment. Hence, moderate flow of sealer is better.[61]

The adaptability of a sealer to the dentin is the primary factor influencing microleakage and reinfection of the root canal.[9] Adhesion of root canal filling material to dentinal walls is important in both static and dynamic situations. In a static situation, it should remove any space which allows the percolation of fluid between the filling material and the wall. In a

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dynamic situation, it is needed to resist dislodgement of the filling during subsequent manipulation.[62]

An optimal root canal sealer should also have reduced surface tension to permit improved penetration into irregularities, increased wettability to provide liquid tight seal and must be biocompatible.[63] The moisture condition of the root canal dentin may influence the adhesion of sealers to dentin by influencing dentin wettability and, thereby, the sealer penetration. Canal wettability of the sealer is hence an important factor.[64] Canal wettability leads to formation of solid-liquid interface and it is formed as there is expulsion of air from the canal. Lower contact angle leads to better flow and greater adhesion properties and as moist environment is present in root canal and hydrophilic properties of root canal sealer are essential for achieving better sealing and adaptation hence this play an important role in achieving fluid tight seal.[65] Insufficient filling of root canal space leads to approximately 60% of endodontic failures. Therefore, novel endodontic substances and obturation techniques were developed.

The currently available commercial root canal sealers can be broadly categorized as ZOE based, glass ionomer-based, calcium hydroxide- based, resin- based, silicone-based and the more recently introduced, calcium silicate based root canal sealers.

Traditional sealers such as zinc oxide eugenol and calcium hydroxide based sealers had several favourable characteristics such as antimicrobial property, adequate setting time, fluidity and radiopacity; but lacked resistance to microleakage due to weak bond and poor dimensional stability, shrinkage and dissolution when in contact with periradicular tissue fluids. Various new sealers which were resin based were developed to improve the bonding with the root canal dentin and to reduce microleakage but these sealers lacked biocompatibility. However, at present none of the existing sealers satisfies all the above-mentioned criteria. Recently introduced calcium silicate-based materials have attracted considerable attentions because of their good biocompatibility and bioactivity.[67]

Traditionally, hydrophobic sealers have been used which have poor wetting of the canal. Therefore, recently hydrophilic sealers are introduced which have low contact angle that allows the sealer to easily spread in the canal and also helps in filling of lateral canals, hence providing better adaptation as compared to hydrophobic sealers.[46] The other advantages of using hydrophilic sealers are these sealers expand instead of shrinking and are dimensionally stable and they form a chemical bond which leads to less space between the sealer and dentin walls. In hydrophilic sealers calcium phosphate formation occurs which increases strength of root canal system and these are also biocompatible. The sealing ability achieved with hydrophilic obturation system is significantly more when compared with the conventional hydrophobic obturation. **Hegde and Arora and Pawar et al.** found that the hydrophilic sealers demonstrate increased marginal adaptation and lesser apical micro leakage values.[6,34] The hydrophilic nature of the sealer should be taken into account as an essential factor when deciding an obturating system.

Most critical area of prepared root canal is the apical 2–3 mm as there is less density and also less diameter of dentinal tubules in this region. Hence, sealing ability of the commonly used sealers were evaluated in the apical third region in the present study. **The apical third area of the root is quite complex as it is difficult to clean, has many variations in anatomy such as ramifications, accessory canals and lateral canal which makes it susceptible to leaking.[68]** Apical leakage is the entry of micro-organisms or leakage of tissue fluids into the canal space. Leakage will however occur within the spaces of sealer, between the obturating material and sealer, sealer and dentin. Sealer penetration also depends on tubule patency and is usually less in the apical region due to sclerotic dentine. This was observed in study conducted by **Aritkala SK et al[8]** and **Jain S et al**.[48]

The present study focuses on the comparative evaluation of apical sealing ability of hydrophilic and hydrophobic sealers. In study conducted by **Hachem et al.[69]** they evaluated influence of endoactivator on tubule penetration of tricalcium silicate sealer and they concluded that endoactivator improved interfacial adaptation of the sealer and leading to reduction inn gap region and thus promoting better root canal treatment.

The prepared samples were randomly divided into four groups depending on type of sealers used.

Group A: AH Plus (Dentsply DeTrey, Germany)
Group B: GuttaFlow 2 (Coltene, India)
Group C: Bio-C (Angelus, Brazil)

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Group D: BioRoot RCS (Septodont, UK)
Out of four root canal sealers, i.e, AH Plus, GuttaFlow 2 are hydrophobic and Bio-C and BioRoot RCS are hydrophilic. All the sealers were manipulated according to manufacturer's instructions and after mixing root canal sealer was applied using a lentulo spiral number 25. Applying the cement sealer with a paste carrier obtains better depth and percentage of penetration, given that it pushes the cement in a centrifugal manner into the root canal walls. A study conducted by **Dash AK et al** evaluated three techniques for sealer placement and found that using lentulospiral showed better penetration when compared to ultrasonics and bidirectional file. [60]

After selecting gutta percha, apical portion was coated with sealer and it was inserted slowly into root canal till the working length. Then SuperEndo Alpha II (B&L Biotech, USA) was used to cut excess gutta percha at the level of orifice. After this, Resin modified Glass ionomer cement was used to seal the root canal openings.

Single cone technique is a filling technique that uses a single gutta-percha cone which was properly matched and sealer was used with these cones. It is simple, saves time for clinicians and easy to master. The risk of root fracture is decreased and no thermal damage to periodontal membrane in teeth filled by using the single cone technique. [65]

Single-cone obturation was used in the present study to simulate most frequent method employed in clinical conditions and to maintain similarity among groups. The concept of the single-cone technique has been recently re-visited by **Wu et al.**, and the volume of the sealer used in the present study was minimized because of use of calibrated gutta-percha.[16]

Samples were kept in incubator for one week at temperature of 37° C and 100% humidity and then samples were subjected to scanning electron microscope (SEM) for microgap evaluation.

Similar methodology to this study was followed in studies conducted by **Hegde V** and **Murkey LS** and **Dsouza AP et al.**[53]

4 sealers were compared in the present study namely AH Plus, GuttaFlow 2, Bio-C and BioRoot RCS. **AH Plus** is an epoxy resin based sealer and is considered gold standard as it is easy to use, readily available and due to its radiopacity. It bonds to root canal as it contains adamantine.[34] It is a two paste root canal sealer and contains resin with faster setting time causing shrinkage and early debonding. Its solubility is 0.3%. Colour change is not observed with this sealer. It has calcium tungstate particles which are larger in size with average size of 1.5µm and as the size of dentinal tubules is less the sealer doesn't enter easily. It shows polymerization due to the presence of polyamine monomers (1-adamantine amine, N,N'-dibenzyl-oxanonandiamine 1,9,TCD- Diamine). When polyamine paste and diepoxide paste are mixed formation of covalent bonds occurs and aliphatic cycle of amine groups leads to modification in curing rate cross linked density and modification in morphology of covalent bonds. Heavily cross-linked polymer results in high rigidity and strength as epoxide group reacts with each amine group.[33]

GuttaFlow 2 is an improved version of RoekoSeal. GuttaFlow 2 is developed in 2014 by Coltene Roekoseal. It has similar composition to GuttaFlow but they are present in different proportion.[67] Being a novel root canal filling material, it combines two products in one. Gutta percha particles have a size of less than 30 µm in powder form and has a homogenous structure which has better adaptation to root canal wall with no shrinkage after placement. It is a self-curing and cold-flowable sealer and has better adaptability and provides good seal.[14] Flow properties of this sealer are outstanding as it has thixotrophic properties which means its viscosity reduces when shear stress is applied.[13]Advantages include 0.2% expansion of the sealer on setting, which helps in better adaptation to both the gutta percha as well as dentin which enhanced sealer flow and adhesion to dentinal tubules. It has almost zero solubility and biocompatiblity.

Developed in 2015 by Septodont, **BioRoot RCS** is a hydraulic cement, that is available as a powder composed of tricalcium silicate, zirconium oxide, povidone and a water- based liquid, with additions of calcium chloride and polycarboxylate. Alkaline nature in bioceramic sealers is due to the by-products formed and they help in better penetration of sealers inside the dentinal tubules as it helps in denaturation of collagen fibres. It consists of particles which have diameter of 5-30µm. It has bioactive properties as when it comes in contact with periapical tissue there is release of BMP-2, FGF-2 and VEGF.[54]

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Bio-C was developed in the year in 2019 by Angelus. It is ready to use as it is available in single syringe and it has ingredients which are biologically active that heals in sealing and healing of the root canal.[70] It consists of tricalcium silicate, dicalcium silicate, calcium oxide, zirconium oxide, silicon oxide, polyethylene glycol and iron oxide. Bioactive property of this sealer is due to release of calcium ions. It interacts with dentin thereby leading to prevention of bacterial infiltration. It has a particle size less than 2µm.[71]

There are several methods for evaluating the apical sealing of root canal sealers, such as bacterial penetration, fluid transport, clarification, penetration of radioisotopes, electrochemical methods and gas chromatography.[74] The adaptation of a sealant to the dentin has generally been evaluated using stereo-microscopy, confocal laser microscopy, scanning electron microscopy (SEM), leakage tests, and digital imaging. In this study, a scanning electron microscope was utilized for the assessment of marginal gap at 3mm level of apex at magnification of 500x between the root canal dentin and the sealer interface and between sealer and gutta percha.[65]

Intergroup Comparison (Hydrophilic versus Hydrophobic groups)

In this study, according to the results obtained hydrophilic group had value 2.82 µm of microgap in the apical third which was better than hydrophobic group which had 8.00 µm and when values were compared statistically, the result was statistically significant. It significance being hydrophilic group having better apical seal as compared to hydrophobic group.

The result are in accordance with studies performed by Hegde V et. al. (2015)[6] who conducted a study comparing hydrophilic with hydrophobic system of obturation using different sealers The result showed that hydrophilic sealers were better when compared to hydrophobic sealers. They concluded that hydrophilic obturating system was better when compared to hydrophobic systems.

Vats A et. al. (2019)[2] conducted a study evaluating leakage in apical area of hydrophic and hydrophobic systems of obturation and dye penetration method and proved that AH Plus showed the highest leakage of dye amongst all groups.

Few studies which are **not in agreement** with our results were those conducted by **Kem Y et al (2019)[78]** performed a study evaluating penetration of BioRoot RCS sealer with AH plus by using confocal laser scanning microscope. The reason stated could be different methodology used for obturation and different parameter used for evaluation.

Possible reasons for our result could be:

1) Hydrophobic nature of Gutta percha cones and sealer has the tendency to pull away the sealer from gutta percha after it sets and thus prevents good adaptation in incompletely dried canal thereby leading to microgap.[23]
2) The hydrophilic nature of the biocramic sealers allows a deeper penetration and in more homogeneous way. The hydrophilic property leads to higher water absorption, low contact angle and slight lateral expansion and thus enabling the sealer to spread easily over the dentinal wall and penetrate into the dentinal tubules.[6]
3) Particle size of hydrophilic sealers is smaller when compared with hydrophobic sealers and is a major factor affecting the penetration of sealer into dentinal tubules in the apical root area.[76]

Limitation of Present Study

These are the following limitations of this study:

- It is an in-vitro study, not an in-vivo study so simulation of the oral environment couldn't be done.
- There is high risk in sectioning of root canals which are filled as it can result in dislodging or ripping of gutta percha.
- Fully hydrated specimens are evaulated by using environmental SEM as it helps in differentiating gaps between dentin and root filling from artifactual gaps which may be created in conventional SEM after the procedure of vacuum dessication.
- Micro-CT could be used which also helps in reconstruction of filled teeth three-dimensionally for evaluating sealant's adaptation.

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Hence, further investigations are needed to evaluate the microgap in the obturation done with hydrophilic and hydrophobic obturation systems.

Conclusion

The objective of endodontic treatment is to establish a hermetic seal in the root canal space which prevents penetration of bacteria or their by-products and tissue fluids into the root canal system. Therefore, to achieve this hermetic seal various sealers have been used but the recently introduced hydrophilic sealers provided a better seal as compared to traditionally present hydrophobic sealers. Under the experimental conditions and within the limitations of this study, the following result can be concluded: Microgap between the sealer and gutta percha noted in the apical third region after obturation in different groups. The microgap evaluated using SEM in the ascending order as:

Hydrophilic group < Hydrophobic group

(2.82 µm) (8.00 µm)

Group D (2.64 µm) < Group C (3.01 µm) < Group B (7.52 µm) < Group A(8.49 µm)

BioRoot RCS < Bio-C < GuttaFlow 2 < AH Plus

Based on the intergroup results obtained, it was concluded that minimum gap formation was seen in hydrophilic sealers and were better as compared to hydrophobic sealers as mean value of hydrophilic group was 2.82µm when compared with hydrophobic group which had 8µm. Among hydrophilic sealers. BioRoot RCS was better when compared to Bio-C sealer. Although, statistically there was no significant difference between group C and group D. Therefore, sealing ability of hydrophilic sealers is more as compared to hydrophobic sealers.

It is important to emphasize that the results of this in vitro study cannot be extrapolated to all clinical conditions hence further investigations are required.

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