Comparative Evaluation Of Sealing Ability Of Zinc Oxide Eugenol Based, Nanoparticle Based And Bioceramic Based Sealers Used Along With Thermoplastisized Obturation Technique - In Vitro Study

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ABSTRACT

Aim: To compare and evaluate sealing ability zinc oxide eugenol based, nanoparticle based and bioceramic based sealer used along with thermoplastisized gutta percha, using a dye penetration method.

Materials and Methods: Eighty extracted human mandibular permanent premolars were selected and decoronated to standardize the root length at 12±1 mm. working length were determined with the help of RVG with 10k file. All the root canals were shaped using NiTi rotary system to size 30, 04 taper. Before obturation, the teeth were divided into 4 groups according to the sealer used, Group I (n=20) No sealer, Group II (n=20) ZOE based, Group III (n=20) Nanoparticle based and Group IV (n=20) Bioceramic based sealers used. Obturation were done using thermoplastisized obturation technique. Access cavities were sealed using Nanohybrid composite. After 7days, teeth air dried and 2 layers of nail varnish applied and immersed in 2% methylene blue dye for 10 min, removed, washed and dried. After sectioning with Tooth sectioning disc, observed with light microscope using 10X, 45X magnification.

Statistical Analysis: The data obtained was statistically analysed using ANOVA test. Results: Nanoparticle based sealer had sealing ability, statistically significant from both sealer. Nanoparticle based sealer had highest sealing ability followed by bioceramic based and zinc oxide eugenol based sealer.

Conclusion: The nanoparticle based sealer sealed the root canals more completely, than with the bioceramic based sealer and zinc oxide eugenol based sealer respectively.

Clinical Implications: The primary goal of successful endodontic therapy is complete obliteration of the root canal space which can be achieved by three dimensional obturation of the canal space. The study enabled us to know the comparative sealing efficacy of different sealing materials employed and hence guide us towards better treatment results.

Introduction

The ultimate objective of the root canal treatment is to achieve maximum eradication of microorganisms from root canal space followed by obturation, in order to form an impervious apical, lateral and coronal seal to prevent re-colonization of the microorganisms.¹ Incomplete sealing and presence of spaces between the root canal wall and the obturating material can lead to failure in treatment.² Different types of sealers have been used in conjunction with gutta-percha for root canal obturation with varied success.³ The main functions of root canal sealer are sealing of the voids, sealing of the patent accessory canals & multiple foramina, forming a bond between the core of filling material & root canal wall, acting as a lubricant while facilitating the placement of filling core.⁴ The most commonly used sealer in root canal treatment is zinc oxide eugenol based sealer.² Zinc oxide eugenol sealer enter into the dentinal wall irregularities and it causes irritation to the periapex, cytotoxicity, staining to the dentinal tubules and have solubility in oral fluids.⁵ To overcome this, various new sealers like Nanoparticle based sealers, Bioceramic based sealers have been introduced.
Nanotechnology is the science of evaluating and producing materials in Nano-dimension by re-location and re-arrangement of atoms to prepare materials with better properties.\(^2\) Nano particles (Np) are ultrafine particles of insoluble constituents with a diameter of less than 100nm. The nanoparticles in sealer causes change in flowability of sealers, results in the penetration of the sealer into minute dentinal tubules, improving the mechanical retention of material by interlocking inside the tubules. They reduce cytotoxicity and improve the sealability. Presence of very small particles leads to superior properties.\(^6\)

Bioceramic sealers are inorganic, non-metallic, non-corrosive materials having mechanical properties similar to dental hard tissues.\(^3\) They include alumina, zirconia, bioactive glass, glass ceramics, hydroxyapatite, calcium silicate and calcium phosphate. They form mechanical interlocking bonds with the dentinal tubules. Their biocompatibility prevents rejection by the surrounding tissues, calcium phosphate in bioceramic which enhances the setting properties of bioceramic and results in a chemical composition and crystalline structure similar to tooth and bone apatite materials, thereby improving sealer-to-root dentin bonding.\(^4\)

The aim of this study is to compare and evaluate the sealing ability of commercial zinc oxide eugenol based sealer, nanoparticle based sealer and bioceramic based sealer used along with thermoplastisized gutta percha, using a dye penetration method.

**MATERIAL AND METHODS:**

- **Root Canal Preparation:**

Eighty extracted human mandibular permanent premolars with single root, single canal and fully formed apices were selected from the Department of Oral and Maxillofacial surgery of our institute. All teeth were cleaned of any debris with ultrasonic Scaler, disinfected by immersing in 5% sodium hypochlorite and then placed in normal saline at 37°C until the experiment. All samples were decoronated using tooth sectioning disc along with straight handpiece under water spray to standardize the root length at 12±1 mm. After insertion of size 10k file, working length was determined with the help of RVG.

Cleaning and shaping of root canals were carried out by rotary system using a torque control motor. All the root canals were shaped using NiTi rotary system to size 30, 04 taper. Between each instrument changeover, canals were irrigated with 1ml of 5% sodium hypochlorite for 1minute using a syringe with 30 gauge side vented needle placed before the binding point but no closer than 2mm to working length. Then irrigation with 1 ml of 17% EDTA Liquid were performed using 30 gauge side vented needle inserted 1 mm from working length. Distilled water was used between each irrigating solution in order to prevent an acid/base reaction between sodium hypochlorite and EDTA. To prevent the action of chemicals with the tissues, the root canals were finally irrigated with 5ml of distilled water to remove chemical residues and then dried internally with paper points.

Before obturation, the teeth were divided into 4 groups according to the sealer used, Group I (n=20) No sealer, Group II (n=20) Zinc oxide eugenol based sealer, Group III (n=20) Nanoparticle based sealer, Group IV (n=20) Bioceramic based sealer used. Then Canal walls were coated with freshly mixed sealer using paste carrier. Obturation were done using thermoplastisized obturation technique, as a rubber stopper was placed 4-5 mm short of working length on a 23-gauge obturation gun needle. At the initiation of every obturation, a new gutta-percha pellet was placed into the obturation gun. When the temperature of the unit touched 200°C, the premeasured needle was positioned in the canal before expressing 3-4 mm of gutta-percha passively into the canal. Vertical pressure with an endodontic plugger was applied for compaction in the apical area. Increments of 3-4 mm were placed in a similar manner, filling the canal to the orifice. Access cavities were then sealed using Nanohybrid composite. For sealer to set completely, teeth were then placed at room temperature for one week.

- **Specimen Staining:**

After 7days, teeth were air dried and 2 layers of nail varnish was applied to the root except 1-2mm around the apex. Specimens were then immersed in 2% methylene blue dye at room temperature for 10 min, removed, washed and dried. Tooth sectioning disc was used to obtain longitudinal section of roots.

After sectioning, split root segments were observed with light microscope using 10X, 45X magnification. Outcome
variables were recorded by measuring amount of dye penetration from coronal side to apex. (Figure No.1)

RESULTS:

Mean dye penetration, standard deviation, Minimum and maximum values were determined for all groups. (Table No.1, Graph No. 1)

To study the variation between and within the groups ‘ANOVA Test’ was used. One way ANOVA showed that there was a statistically significant difference between and within the groups and the null hypothesis was rejected. (Table No. 2)

<table>
<thead>
<tr>
<th></th>
<th>N</th>
<th>Mean</th>
<th>Std. Deviation</th>
<th>Minimum</th>
<th>Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group I</td>
<td>20</td>
<td>2.49</td>
<td>0.42</td>
<td>1.80</td>
<td>3.34</td>
</tr>
<tr>
<td>Group II</td>
<td>20</td>
<td>1.83</td>
<td>0.22</td>
<td>1.50</td>
<td>2.40</td>
</tr>
<tr>
<td>Group III</td>
<td>20</td>
<td>0.77</td>
<td>0.37</td>
<td>0.36</td>
<td>1.80</td>
</tr>
<tr>
<td>Group IV</td>
<td>20</td>
<td>1.21</td>
<td>0.38</td>
<td>0.80</td>
<td>1.90</td>
</tr>
</tbody>
</table>

*std- standard

Table No. 1: The mean, standard deviation, minimum and maximum values of all groups.

DISCUSSION:

The major aim and objectives of the obturation is to achieve three-dimensional sealing of the pulp space to create fluid-tight seal, prevent ingress of microorganisms, their toxins and their flow into the periapical tissues which is achieved with sealers and obturating materials.\(^6\) Sealing ability is a crucial characteristic for endodontic materials.\(^3\) Poor apical sealing of the root canal space accounted for 60% of endodontic treatment failures.\(^1\)

Sealers can be a cause of root canal failure due to microleakage at sealer-dentin or sealer-core material interface. Bonding of the sealer to the root canal dentin wall and formation of a monoblock can eliminate this drawback. Various authors have shown that teeth obturated with gutta percha points and sealer display less leakage than those without sealer. Ahmed G. Subhy et al (2008) concluded that
Table No. 2: Analysis of Variance for depth of Dye Penetration for all groups.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Sum of Squares (SS)</th>
<th>Degrees of Freedom (df)</th>
<th>Mean Square(MS)</th>
<th>F ratio</th>
<th>Level of Significance(p value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Between Groups</td>
<td>33.698</td>
<td>3</td>
<td>11.233</td>
<td>88.200</td>
<td>0.000</td>
</tr>
<tr>
<td>Within Groups</td>
<td>9.679</td>
<td>76</td>
<td>0.127</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>43.376</td>
<td>79</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* The mean difference is significant at the ≤0.05 level. (* P Value ≤0.05 =significant)
<table>
<thead>
<tr>
<th>Group (I)</th>
<th>Mean Difference (I-J)</th>
<th>Std. Error</th>
<th>Level of Significance (p-value)</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group I</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group II</td>
<td>0.66450</td>
<td>0.11285</td>
<td>0.000</td>
<td>Significant</td>
</tr>
<tr>
<td>Group III</td>
<td>1.72250</td>
<td>0.11285</td>
<td>0.000</td>
<td>Significant</td>
</tr>
<tr>
<td>Group IV</td>
<td>1.27950</td>
<td>0.11285</td>
<td>0.000</td>
<td>Significant</td>
</tr>
<tr>
<td>Group II</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group III</td>
<td>1.05800</td>
<td>0.11285</td>
<td>0.000</td>
<td>Significant</td>
</tr>
<tr>
<td>Group IV</td>
<td>0.61500</td>
<td>0.11285</td>
<td>0.000</td>
<td>Significant</td>
</tr>
<tr>
<td>Group III</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group IV</td>
<td>-0.44300</td>
<td>0.11285</td>
<td>0.000</td>
<td>Significant</td>
</tr>
</tbody>
</table>

Table No. 3: Comparison of dye penetration depth between the groups.

*std- standard  * The mean difference is significant at the ≤0.05 level.

(* P Value ≤0.05 =significant)

According to Wu et al over 80% of leakage studies related to endodontics have employed radioisotope penetration or dye penetration. Methylene blue dye was used in this study as its molecular size is similar to bacterial by-products such as butyric acid which can leak out of infected root canals to irritate periapical tissues, also it is easy to use, pH manipulation and availability add to it advantages. Souza EM et al (2007) shown that alkaline materials cause methylene blue discoloration, which may lead to unreliable conclusions in dye penetration tests. Methylene blue discoloration occurs because it is unstable when in contact with alkaline materials. Such materials cause hydrolysis of methylene blue, resulting in formation of a clear compound named thionine.

The most commonly used sealers in root canal treatment are ZOE based sealer. The advantages of zinc oxide eugenol sealer are, its antimicrobial activity, biocompatibility, gets resorbed even if extruded into the periapical tissue and popularity among clinicians. Disadvantages of zinc oxide eugenol sealer are it has poor dimensional stability, it contracts when temperature changes, less sealing ability, it causes irritation to the periapex, cytotoxicity, staining to the dentinal tubules and tooth structure, prolonged setting time, shrinks on setting and have solubility in oral fluids. To overcome these disadvantages, various new sealers like Nanoparticle based sealers, Bioceramic based sealers have been introduced.

Nanoparticle based sealers have been synthesized using the scientific branch of nanotechnology. In the present study, we have used the Nanoseal- S nanoparticle based sealer.
Graph No.1: Mean Plot of depth of dye penetration of the groups.

Graph No.2: Comparison of Mean dye Penetration depth between the Groups.
Nanoparticles (Np) are ultrafine particles of insoluble constituents with a diameter of less than 100 nm. The nanoparticles in sealer causes change in flowability of sealers, results in the penetration of the sealer into minute dentinal tubules, improving the mechanical retention of material by interlocking inside the tubules.\(^6\)

Bioceramic based sealers have been available for use in endodontics for the past thirty years. Bioceramics are inorganic, non-metallic, biocompatible materials that have mechanical properties similar to dental hard tissues.\(^3\) In this study, Ceraseal- B Bioceramic root canal sealer have been used. Bioceramic root canal sealers exhibit chemical bonding to root canal dentin walls as well as its corresponding bioceramic particle impregnated gutta-percha. Bioceramic materials contain calcium phosphate which enhances the setting properties of Bioceramics and results in a chemical composition and crystalline structure similar to tooth and bone apatite materials, thereby improving sealer to root dentin bonding.\(^4\)

When compares the result, Group III showed highest sealing ability , the reason for this difference in the sealability between nanoparticles and other sealers was related to the size of the nanoparticles, which provided them with a greater contact surface area and charge density than bulky powders. Nanoparticle based sealer have size of the nanoparticles with a diameter of 100 nm or less which increases its sealing ability.\(^6\) Group IV showed better sealing ability, as diffusion of the sealer particles into the dentinal tubules (tubular diffusion) to produce mechanical interlocking bonds. Bioceramic sealers seem to positively interact with dentin fluid, potentially inducing biominerlization, with the formation of mineral tags within the dentinal tubules, thus enhancing the biological activity within the root canal.\(^4\) And in bioceramic sealer, methylene blue suffers discoloration when in contact with some alkaline filling materials, which may cause unrealistic results of such materials in leakage studies.\(^9\) The sealing ability of a sealer is linked to its solubility and to its bonding to the gutta-percha cone to the dentin. Several studies have evaluated the sealing abilities of various bioceramic-based sealers in vitro. (10-12) Renu Sroa et al (2017) gave in accordance to our study, EndoSequence sealer, based on aluminium-free calcium silicate nanoparticulate composition. It has better sealing ability than ProRoot MTA bioceramic based sealer.\(^13\) In contrary to our study, Dr. Shafia Rashid (2021) showed that bioceramic based sealer have more sealing ability than nanoparticle based sealer.\(^14\)

Group II showed lowest sealing ability than Group III and Group IV. The reason behind is, Zinc oxide eugenol is

**Figure No. 1:** Dye penetration in **Group I** – No sealer, **Group II** – Zinc oxide eugenol based sealer, **Group III** – Nano particle based sealer, and **Group IV** – Bioceramic based sealer.
water-soluble and has the disadvantage of being decomposed by water through a continuous loss of eugenol. This makes zinc oxide eugenol a weak unstable material. And it shows least penetration ability and highest leakage value.\textsuperscript{15} Also they show certain degree of shrinkage after setting, which might lead to the generation of voids.\textsuperscript{16} also has cytotoxic potential and interference with adhesive procedures due to the free eugenol release in tissue fluids and dental tissues.\textsuperscript{17-19} Chandrika Ramanathan Palanivelu et al (2019) gave study in accordance to our study, concluded that AH plus and NanoSeal- S have excellent apical seal, statistically way much better than zinc oxide eugenol.\textsuperscript{7} Srinidhi V. Ballulayya et al (2017) gave study in accordance to our study that concluded that Traditional, zinc oxide eugenol based sealers have been surpassed by the Bioceramic based sealers which have better sealing ability.\textsuperscript{20}

In vitro leakage studies comprise a major portion of contemporary endodontic research though it is difficult to draw in vivo correlation. Further in vivo studies are required to evaluate whether this seal can be maintained for long periods of time.

CONCLUSION:

Within the limitation of the study, based on the recorded data and statistical analysis the following conclusions can be drawn:

- A statistically significant difference between sealing ability of zinc oxide eugenol based, nanoparticle based and bioceramic based sealer.
- Group III (Nanoparticle based sealer) had highest sealing ability followed by Group IV (bioceramic based sealer), Group II (zinc oxide eugenol based sealer) and Group I (Without any sealer).
- Thereby, the nanoparticle based sealer sealed the root canals more completely than the bioceramic based sealer and zinc oxide eugenol based sealer.

REFERENCES:


