

## Comparative Evaluation of coronal and apical sealing ability of three different types of sealers with single cone obturation technique (In-vitro study)

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### Abstract

**Introduction:** The single cone obturation technique is one of the widely used root canal filling techniques due to its simplicity and time efficiency. However, this technique requires a larger amount of sealer compared to other filling techniques, making it necessary to have a sealer with suitable physical and chemical properties to achieve an effective seal when filling the root canal using the single cone technique.

**Objective:** This experimental study aims to compare three different types of sealers (Endosequence BC, GuttaFlow Bioseal, and Adseal) in terms of their ability to achieve coronal and apical seals when filling the root canal using the single cone obturation technique. The study used the dye penetration test to assess the sealing ability.

**Methodology:** The study included 75 extracted human mandibular premolars: 30 for coronal and 30 for apical leakage assessment, each divided into three sealer subgroups (n = 10), plus a positive control (n = 10; 5 per leakage type) and a negative control (n = 5). Teeth were decoronated, standardized, prepared to size 30/4% with rotary files, and filled using the single-cone technique with matching gutta-percha. After 48 h of sealer setting, specimens were immersed in 1% methylene blue for 72 h, rinsed, sectioned bucco-lingually, and dye penetration was quantified using ImageJ.

**Results:** In the coronal leakage study, Adseal sealer demonstrated the lowest coronal leakage, followed by GuttaFlow Bioseal, with no significant statistical difference between them. Endosequence BC sealer showed the highest coronal leakage. In the apical leakage study, Endosequence BC sealer showed the least apical leakage, followed by GuttaFlow Bioseal, with no significant statistical difference between them, and Adseal demonstrated the highest apical leakage.

**Conclusions:** Coronal sealing ability was superior with the epoxy resin-based sealer (Adseal), followed by GuttaFlow Bioseal, with EndoSequence BC exhibiting the highest coronal leakage. Apical sealing ability was superior with the bioceramic sealer (EndoSequence BC), followed by GuttaFlow Bioseal, whereas Adseal demonstrated the highest apical leakage. GuttaFlow Bioseal demonstrated consistent performance in both coronal and apical regions, highlighting its reliability as an alternative material.

**Keywords:** Root canal filling, single cone obturation technique, sealers, coronal seal, apical seal, dye penetration, lower premolars.



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## Introduction

Root canal therapy (RCT) is a fundamental procedure in the management of pulpal diseases, aiming to eliminate microorganisms residing within the root canal system and to prevent their recolonization through comprehensive chemo-mechanical debridement followed by three-dimensional obturation [1]. Achieving long-term success in endodontic treatment depends on several factors, among which the quality of root canal filling plays a pivotal role as a prognostic indicator [2]. Complete eradication of microorganisms is often unattainable due to the complex anatomy of the root canal system. Therefore, achieving a hermetic seal is critical to prevent bacterial ingress from the oral cavity and to entomb residual microorganisms within the canal [3]. Adequate cleaning, shaping, irrigation, and application of intracanal medicaments are essential prerequisites for disinfection, while three-dimensional obturation remains indispensable for preventing reinfection by sealing the system and promoting periapical tissue healing [4].

Successful endodontic outcomes rely heavily on establishing a tight apical and coronal seal following chemo-mechanical preparation. This prevents recontamination and facilitates periapical healing [5]. The objectives of root canal filling are to completely obturate the canal system, entomb remaining microorganisms, reinforce the tooth structure, and provide a coronal seal against microbial leakage, thereby ensuring long-term treatment success [6].

Over the years, no filling material has shown ideal performance in producing a complete seal. The universally accepted gold standard for obturation remains gutta-percha, owing to its biocompatibility, non-toxicity, thermal plasticity, retrievability, and inertness after placement. However, gutta-percha lacks adhesiveness to canal dentin, leading to microgaps. This limitation underscores the importance of incorporating a root canal sealer, which fills the voids between the dentinal wall and the core material, as well as irregularities and accessory canals [7]. Endodontic sealers are self-curing cements used in conjunction with solid or semi-solid core materials. In some cases, they may be used alone to fill the canal; however, due to their variable resorbability, they are not recommended as standalone obturation materials. Instead, sealers play a critical role in filling voids, adapting the core material to the canal walls, and sealing lateral canals [8,9]. Leakage is considered the primary cause of root canal treatment failure, as fluids

carrying microorganisms infiltrate between the sealer and the dentinal walls coronally or apically [10]. Coronal leakage, in particular, can result in long-term failure and the development of periapical disease due to reinfection after loss of coronal integrity [11]. Therefore, sealers must possess sufficient sealing ability, chemical stability, antibacterial properties, and resistance to dissolution [12]. From a histological perspective, the three-dimensional seal strongly depends on the sealer's ability to penetrate dentinal tubules, increasing the contact surface with dentin, enhancing mechanical retention, and entombing microorganisms [13]. Thus, deeper and more consistent tubular penetration is considered a critical factor in the clinical success of sealers [14].

Currently, resin-based sealers are the most widely used materials due to their favorable physical properties, dimensional stability, and adequate apical sealing. However, they lack chemical bonding to dentin, which has driven the search for alternative sealers capable of bonding to root canal walls [15]. Calcium silicate-based sealers were introduced into endodontics with the advent of Mineral Trioxide Aggregate (MTA) by Torabinejad, initially developed for perforation repair, root-end filling, and pulp capping [16,17]. Recent formulations based on tricalcium or dicalcium silicates have gained attention due to their biocompatibility, antimicrobial activity, alkalinity, and bioactivity, including hydroxyapatite formation and chemical bonding to dentin [18].

In addition, silicone-based sealers have demonstrated favorable physicochemical and biological properties [19]. Various obturation techniques have been proposed, such as cold lateral condensation, thermoplastic techniques, and more recently, the single-cone technique (A logical and reproducible volume of 0.05 mL of sealer was applied to each canal, ensuring complete coating of the canal walls without excessive extrusion), which employs a gutta-percha cone matching the taper of the final rotary file used in instrumentation [20]. This technique is widely used due to its simplicity and efficiency, especially with modern rotary systems, though it carries the limitation of potential void formation in irregular canal spaces [9]. As the single-cone technique relies more heavily on the sealer, the physicochemical and biological properties of the sealer play a decisive role in treatment outcomes [21].

## Research Methodology

### Sample Size Calculation

*Sample size was calculated a priori using GPower software (v3.1.9.7, Heinrich-Heine-Universität Düsseldorf, Germany) for one-way ANOVA ( $\alpha=0.05$ , power=0.85, medium effect size  $f=0.30$ ), yielding a minimum of 9 specimens per group, which was increased to 10 to account for anticipated technical loss during sectioning. The study sample consisted of 75 extracted human mandibular premolars with a single root canal.*

### Inclusion criteria:

- Mandibular premolars with a single canal and straight root.
- Intact teeth with fully developed apices, free from previous endodontic treatment.
- Teeth free from caries, fractures, or root cracks.
- Absence of internal or external resorption.
- Comparable root lengths.

### Exclusion criteria:

- Mandibular premolars with multiple canals or roots.
- Teeth with incompletely developed apices.
- Teeth previously treated endodontically.
- Mandibular premolars with curved canals ( $> 5^\circ$  curvature using the Schneider method with J Image software).
- Teeth presenting caries, fractures, or root cracks.
- Teeth exhibiting internal or external resorption.

All inclusion and exclusion criteria were verified both clinically and radiographically. Radiographic evaluation was performed using a Nanopix 1 sensor (Eighteeth, China) in conjunction with a

Hyper Light DC portable X-ray unit (Eighteeth, China) to confirm compliance with the required sample criteria.

After removing external debris from the surface of the extracted teeth, the specimens were immersed in 2.5% sodium hypochlorite (NaOCl) solution for two hours, followed by storage in 0.9% physiological saline (PBS) to neutralize potential effects on dentin properties.

### **Crown Sectioning**

The coronal portion of each tooth was sectioned to standardize root length. Sectioning was performed using thin zirconia discs with a diamond base mounted on a contra-angle handpiece (Being, China). The desired root length was determined and standardized using a digital caliper.

### **Root Canal Preparation**

Canal patency was confirmed by inserting a stainless steel K-file #10 (UDG, China) until it extended 1 mm beyond the anatomical apex. The working length was determined by inserting the hand file until it was visible at the apical foramen, and then subtracting 0.5 mm from this measurement.

Following determination of the working length, the root canals were prepared and cleaned using the M3 Pro V3 rotary system up to size 30 (blue) with a 4% taper. During instrumentation, irrigation was performed with 2 ml of 5% sodium hypochlorite (NaOCl) after each file change.

Final irrigation was performed with 5 ml of 17% EDTA for 1 minute, followed by 5 ml of 5% NaOCl, with physiological saline solution used in between to avoid interactions. A final flush was performed with 5 ml of 0.9% physiological saline solution to remove any remnants of irrigants. All irrigation procedures were carried out using a side-vented irrigation needle inserted up to 1 mm short of the apical foramen. The canals were then dried using paper points size 30, 4% taper (Gapadent, China).

### Root Canal Obturation Using the Single-Cone Technique

Obturation was performed using a gutta-percha cone size 30, 4% taper (HTM, China), corresponding to the final rotary file (MAF). Each cone was inserted to the full working length, and tug-back was confirmed in all specimens.

### Sample Grouping

The specimens were randomly divided into nine groups:

- **Three groups for coronal microleakage assessment (30 teeth):**
  - Group 1 (10 teeth): Single-cone obturation using a bioceramic sealer based on tricalcium silicate (EndoSequence BC Sealer).
  - Group 2 (10 teeth): Single-cone obturation using a silicone-based sealer (GuttaFlow Bioseal).
  - Group 3 (10 teeth): Single-cone obturation using an epoxy resin-based sealer (Adseal).
- **Three groups for apical microleakage assessment (30 teeth):**
  - Group 1 (10 teeth): Single-cone obturation with EndoSequence BC Sealer.
  - Group 2 (10 teeth): Single-cone obturation with GuttaFlow Bioseal.
  - Group 3 (10 teeth): Single-cone obturation with Adseal.
- **Three control groups (15 teeth):**
  - Positive control for coronal leakage (5 teeth): Single-cone obturation performed without sealer.
  - Positive control for apical leakage (5 teeth): Single-cone obturation performed without sealer.
  - Negative control (5 teeth): Root canals prepared but not obturated.

**Obturation with EndoSequence BC Sealer:**

EndoSequence BC Sealer is a premixed injectable bioceramic sealer that does not require manual mixing. The syringe tip was inserted into the canal up to the junction of the middle and apical third, and the sealer was slowly and gently injected. Subsequently, a gutta-percha cone was carefully placed to the full working length.

**Obturation with GuttaFlow Bioseal Sealer:**

The sealer was extruded through its self-mixing tip onto a mixing pad. The pre-selected gutta-percha cone was then coated with the sealer to ensure uniform distribution along the root canal walls. A small amount of sealer was applied to the apical tip of the cone, which was subsequently inserted gently into the canal to the full working length.

**Obturation with Adseal Sealer:**

The sealer was mixed on a mixing pad according to the manufacturer's instructions. The pre-selected gutta-percha cone was then used to coat the root canal walls with the sealer. A small amount of sealer was placed on the apical tip of the cone, which was subsequently inserted gently into the canal to the full working length.

Finally, the gutta-percha cone was sectioned at the canal orifice using a heated instrument and vertically condensed with a plugger.

Upon completion of obturation, all specimens were stored in an incubator at 37 °C and 100% humidity for 48 hours to allow complete setting of the tested sealers.

After incubation, the root surfaces of all teeth were thoroughly dried and coated with two layers of nail varnish, with the following exceptions:

- In the apical microleakage groups and the positive apical control group, the apical 2 mm of the root was left uncoated.
- In the coronal microleakage groups and the positive coronal control group, the coronal 2 mm of the root was left uncoated.

- In the negative control group, the entire root surface was completely coated with nail varnish.

The first layer of nail varnish was allowed to dry completely before the application of the second layer to ensure proper coverage and sealing of the root surface.

Following this stage, the specimens were immersed in 1% methylene blue solution for 72 hours in an incubator maintained at 37 °C and 100% humidity.

The use of 1% methylene blue has been shown to provide superior outcomes compared with other dyes commonly used in dentistry [12]. The specimens were suspended vertically in the dye to allow penetration from the apical portion via capillary action.

After removal from the dye, the teeth were rinsed thoroughly under running water for 15 minutes. Subsequently, the nail varnish coating was carefully removed using a 11 surgical blade.

After completion of the dye immersion stage, the specimens were sectioned longitudinally, parallel to the long axis of the tooth, in a bucco-lingual direction. Sectioning was performed using thin zirconia separation discs with a diamond base, operated at low speed and mounted on a Being contra-angle handpiece, (Figure 1 ,2 ,3).

#### **Image Acquisition and Microleakage Measurement**

The longitudinal sections of the teeth were photographed using an iPhone 11 Pro Max camera. The images were subsequently imported into ImageJ software for analysis of dye penetration values.

Within the software, the unit of measurement was set to millimeters (mm), and the scale was calibrated individually for each specimen using the millimeter ruler included in the photographs of the tooth sections.

Finally, the microleakage values obtained for each type of sealer, in both coronal and apical leakage assessments, were archived for subsequent statistical analysis and evaluation of the results.

## **Statistical study and results**

Statistical analyses were performed using R version 4.3.3 (R Foundation for Statistical Computing, Vienna, Austria), an open-source statistical computing environment available at <https://www.r-project.org/>.

### **Control Groups**

#### **Positive Control Group**

This group was divided into a positive coronal leakage control group (5 teeth) and a positive apical leakage control group (5 teeth).

Mean leakage values:

- Coronal: 8.96 mm
- Apical: 5.64 mm

These high values confirm the accuracy of the testing method, demonstrating detectable leakage in the absence of an adequate seal, which emphasizes the importance of using sealers during root canal obturation.

#### **Negative Control Group**

In this group, all specimens were completely isolated from all surfaces using a leakage-resistant coating (nail varnish).

Mean leakage value: 0 mm. All samples showed zero leakage, confirming the effectiveness of complete sealing and the capacity of the coating material to prevent dye penetration.

The distinct contrast between the positive control (highest leakage) and the negative control (no leakage) validates the reliability of the model employed in this study to accurately measure dye penetration.

### Statistical Analysis

A statistical analysis was conducted to compare the sealing effectiveness of three different sealers: Adseal, EndoSequence BC, and GuttaFlow Bioseal, in achieving coronal and apical seals. A significance level of 5% ( $\alpha = 0.05$ ), corresponding to a 95% confidence level, was adopted for the study. All data were tested for normality using the Kolmogorov–Smirnov test ( $p > 0.05$ ) and for homogeneity of variance using Levene's test ( $p > 0.05$ ), justifying the use of parametric statistical tests.

### Statistical Tests Applied

- One-way ANOVA was employed to detect overall differences among the groups.
- Tukey's Honestly Significant Difference (HSD) test was applied for post-hoc pairwise comparisons to identify the specific groups exhibiting statistically significant differences.

All analyses were conducted using the actual raw values obtained for each specimen.

### One-way ANOVA Results for Coronal Leakage

Descriptive statistics were found for the Coronal Leakage sample (Table 1), and an analysis of variance test was conducted, where we found that:

- F-value  $\approx 45.21$
- p-value  $\approx 0.0000$  ( $< 0.05$ )

These results demonstrate the presence of highly significant statistical differences among the three sealers with respect to coronal microleakage.

According to the Tukey HSD test for coronal leakage (table 2):

- The difference between Adseal and EndoSequence BC was statistically significant ( $p < 0.05$ ).
- The difference between EndoSequence BC and GuttaFlow Bioseal was also statistically significant ( $p < 0.05$ ).

- The difference between Adseal and GuttaFlow Bioseal was not statistically significant ( $p > 0.05$ ).

### **Coronal Leakage Results**

The findings indicated that Adseal demonstrated the lowest coronal leakage, followed by GuttaFlow Bioseal, with no significant difference between the two. EndoSequence BC Sealer exhibited the highest coronal leakage, with statistically significant differences compared with the other two sealers

### **One-way ANOVA Results for Apical Leakage**

Descriptive statistics were found for the Apical Leakage sample (Table 3), and an analysis of variance test was conducted, where we found that:

- F-value  $\approx 15.25$
- p-value  $\approx 0.00004$  ( $< 0.05$ )

These results confirm the presence of statistically significant differences among the three sealers with respect to apical microleakage.

According to the Tukey HSD test for apical leakage (Table 4):

- The difference between Adseal and EndoSequence BC was statistically significant ( $p < 0.05$ ).
- The difference between EndoSequence BC and GuttaFlow Bioseal was not statistically significant ( $p > 0.05$ ).
- The difference between Adseal and GuttaFlow Bioseal was statistically significant ( $p < 0.05$ ).

### Apical Leakage Results

The results demonstrated that EndoSequence BC Sealer provided the best apical seal, followed by GuttaFlow Bioseal, with no significant difference between the two. Adseal exhibited the highest apical leakage, with statistically significant differences compared to both of the other sealers.

### Graphical Representation

The figure 4 illustrates the dye penetration values (in millimeters) for the three tested sealers (Adseal, GuttaFlow Bioseal, and EndoSequence BC) at both the coronal and apical levels.

### Discussion

The results of this study revealed the presence of both coronal and apical microleakage in all tested specimens, confirming that none of the materials or obturation techniques used are capable of achieving an ideal, completely hermetic seal of the root canal system. Moreover, the mean coronal leakage values were consistently higher than the apical leakage values. This finding may be attributed to the structural differences in dentin between the two regions; the coronal portion of root dentin exhibits a greater number of dentinal tubules with wider diameters, thereby providing a more favorable environment for leakage. Although performance was described as 'consistent,' results showed variability between apical and coronal regions. Therefore, the term 'consistent' refers to relatively stable performance compared with other tested materials rather than absolute uniformity.

### Coronal Leakage

The results demonstrated that the epoxy resin-based sealer (Adseal) exhibited the lowest coronal leakage, followed by the silicone-based GuttaFlow Bioseal, with no significant difference between the two. The bioceramic sealer (EndoSequence BC) showed the highest coronal leakage, with statistically significant differences compared to the other sealers.

This superiority of Adseal in coronal sealing may be explained by its favorable setting properties and adhesive characteristics. Epoxy resin sealers are capable of penetrating dentinal tubules and

establishing a mechanical interlocking with both dentinal walls and the gutta-percha cone [22]. These findings are in agreement with Vo et al. (2022) [10], who also reported the superior sealing ability of AH Plus (chemically similar to Adseal) compared with EndoSequence BC. Conversely, Barham et al. (2023) [23], reported different outcomes, where One-Fil (bioceramic) showed superior coronal sealing compared with Adseal.

The weaker performance of EndoSequence BC at the coronal level may be linked to its dependence on moisture for setting. In the coronal region, where a larger volume of material may remain and microvoids may hinder adequate hydration, incomplete setting could compromise the seal [24].

### **Apical Leakage**

In contrast, EndoSequence BC exhibited the lowest apical leakage, followed closely by GuttaFlow Bioseal, with no significant difference between them. Adseal presented the highest apical leakage, with statistically significant differences compared to the other two sealers. These results highlight the superior performance of bioceramic and silicone-based sealers in the apical third.

This can be explained by the moisture-rich apical environment, which favors the hydration reaction and setting of EndoSequence BC. The release of calcium and hydroxyl ions during its setting fosters hydroxyapatite crystal formation at the dentin–sealer interface, generating a stable chemical bond with dentin [25]. Furthermore, bioceramic sealers demonstrate deeper penetration into dentinal tubules compared with hydrophobic resin sealers [26].

GuttaFlow Bioseal also performed well apically due to its thixotropic properties and slight expansion upon water absorption, ensuring improved sealing [22, 27]. Additionally, Hseen et al. (2024) [28], demonstrated that GuttaFlow Bioseal achieved leakage rates comparable to MTA when used as a retrofilling material, reinforcing its reliability.

The inferior apical sealing of Adseal may be explained by its hydrophobic resin composition and lack of expansion upon setting, which, combined with potential dimensional changes in moist environments, could lead to microgaps over time [22],

The present findings align with Kaul et al. (2021) [12], who found that EndoSequence BC had the lowest apical leakage, followed by GuttaFlow Bioseal, whereas AH Plus exhibited the highest leakage. Similarly, Lee et al. (2020) [22] and [25], confirmed the superior sealing ability of EndoSequence BC and GuttaFlow Bioseal compared with epoxy resin sealers.

However, other studies have reported conflicting results. Huang et al. (2018) [29], found no significant difference between EndoSequence BC and AH Plus in apical sealing ability using SEM and micro-CT, while Vo et al. (2022) [10], reported AH Plus had significantly lower apical leakage compared with EndoSequence BC. These inconsistencies may be attributed to differences in methodologies (dye penetration, fluid filtration, bacterial leakage), tooth types (molars vs. premolars), canal morphology, irrigation protocols, and storage conditions.

### Conclusions

Within the limitations of this in vitro study, the following conclusions can be drawn:

1. No material or obturation technique provided a completely hermetic seal of the root canal system, as both coronal and apical leakage were observed across all groups.
2. Coronal sealing ability was superior with the epoxy resin-based sealer (Adseal), followed by GuttaFlow Bioseal, with EndoSequence BC exhibiting the highest coronal leakage.
3. Apical sealing ability was superior with the bioceramic sealer (EndoSequence BC), followed by GuttaFlow Bioseal, whereas Adseal demonstrated the highest apical leakage.
4. GuttaFlow Bioseal demonstrated consistent performance in both coronal and apical regions, highlighting its reliability as an alternative material.

These findings emphasize that the selection of a root canal sealer should be guided by its chemical composition, setting reaction, and interaction with the dentinal substrate, to achieve optimal long-term sealing and clinical success.



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M.A. conceived the study, conducted the experimental work, and drafted the manuscript. M.H. contributed to the theoretical review and provided scientific supervision. Both authors reviewed and approved the final manuscript.

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## Tables and Legends

Table 1. Descriptive statistics of coronal leakage

| Type of sealer    | Mean  | SD    | SE    | N  |
|-------------------|-------|-------|-------|----|
| ENDOSEQUENCE BC   | 5.804 | 2.018 | 0.638 | 10 |
| GUTTAFLOW BIOSEAL | 1.336 | 0.469 | 0.148 | 10 |
| ADSEAL            | 1.185 | 0.526 | 0.166 | 10 |

Table 2. Results of the Tukey HSD test for coronal leakage

| Comparison                           | Mean Difference | p-value |
|--------------------------------------|-----------------|---------|
| ADSEAL VS ENDOSEQUENCE BC            | 4.619           | <0.001  |
| GUTTAFLOW BIOSEAL VS ENDOSEQUENCE BC | 4.468           | <0.001  |
| ADSEAL VS GUTTAFLOW BIOSEAL          | 0.151           | 0.9600  |

Table 3. Descriptive statistics of apical leakage

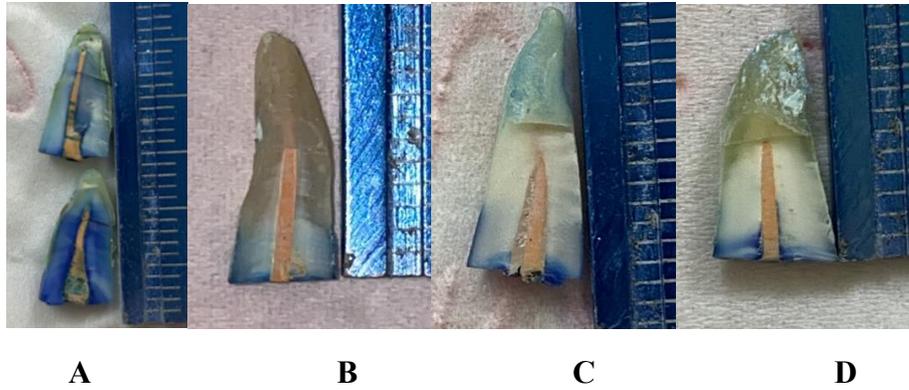
| Type of sealer    | Mean  | SD    | SE    | N  |
|-------------------|-------|-------|-------|----|
| ENDOSEQUENCE BC   | 0.500 | 0.340 | 0.107 | 10 |
| GUTTAFLOW BIOSEAL | 0.670 | 0.385 | 0.121 | 10 |
| ADSEAL            | 1.369 | 0.390 | 0.123 | 10 |

Table 4. Results of the Tukey HSD test for apical leakage

| Comparison                           | Mean Difference | p-value |
|--------------------------------------|-----------------|---------|
| ADSEAL VS ENDOSEQUENCE BC            | 0.869           | <0.001  |
| GUTTAFLOW BIOSEAL VS ENDOSEQUENCE BC | 0.17            | 0.5714  |
| ADSEAL VS GUTTAFLOW BIOSEAL          | 0.70            | <0.001  |

**Figures and Legends**

**Figure 1. Coronal leakage (A): Endosequence BC , (B): GuttaFlow Bioseal , (C): Adseal , (D): Positive control specimen**



**Figure 2. Apical leakage (A): Endosequence BC , (B): GuttaFlow Bioseal , (C): Adseal , (D): Positive control specimen**

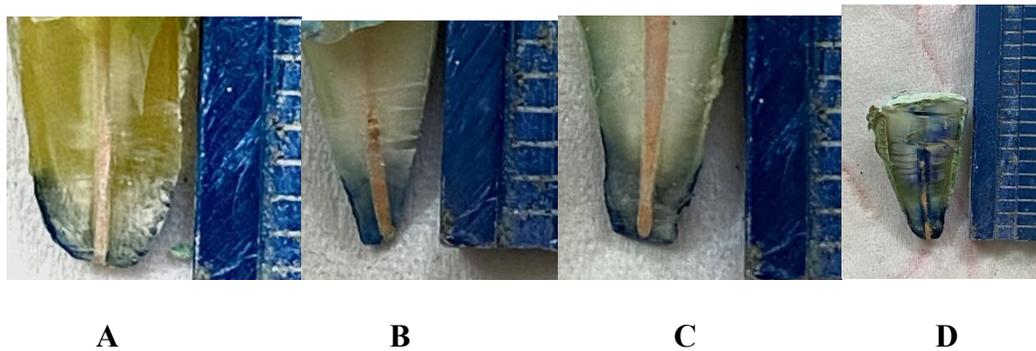


Figure 3. Negative control specimen



Figure 4. Dye leakage results at both coronal and apical levels for the three sealers tested in this study

