



RESEARCH ARTICLE

In vitro apical microleakage evaluation for different endodontic sealers by spectrophotometric analysis: an observational study [version 1; peer review: 1 not approved]

Zahraa Sahib Mezaal¹, Shatha Abdulkareem ¹, Laith G. Shareef ²

¹Department of Restorative and Esthetic Dentistry, College of Dentistry, University of Baghdad, Baghdad, 10011, Iraq

²Department of Pharmacy, Al-Rasheed University College, Baghdad, 10011, Iraq

V1 First published: 28 Nov 2022, 11:1396
<https://doi.org/10.12688/f1000research.127911.1>

Latest published: 28 Nov 2022, 11:1396
<https://doi.org/10.12688/f1000research.127911.1>

Abstract

Background: It has been established that several filling techniques can affect apical leakage, which is responsible for 59% of endodontic failures. The primary goal of endodontic therapy is to create a tight seal that aids in repairing the periapical tissues, prevents apical periodontitis, and shields against root canal infection. The study aims to compare the apical sealing ability of epoxy resin based sealer (AH plus), which is an epoxy-resin-based root canal sealer, GuttaFlow 2, which is a silicone-based root canal sealer, GuttaFlow bioseal is a bioactive glass-based root canal sealer, TotalFill HiFlow bioceramic (BC) sealer is a silicate-based root canal sealer (bioceramic sealer) using a single cone technique by spectrophotometric analysis.

Methods: This study chose 64 maxillary first molars with a straight palatal root. Following palatal roots sectioning to 11 mm from the root apex, the canals were instrumented up to size X4 using Protaper Next. They were separated into four experimental groups (n = 15) per each group based on the kind of sealer utilized, Group I: AH plus, Group II: GuttaFlow 2, Group III: GuttaFlow bioseal, Group IV: TotalFill BC HiFlow, and two control groups (n = 2 for each). After completing obturation, the dye extraction technique was used to evaluate apical leakage. A spectrophotometer was used to measure the optical density of the solution after all the dyes were liberated from the interfacial regions.

Result: A statistically non-significant difference between the investigated root canal sealers in apical microleakage $P > 0.05$ was noted. The highest mean values were observed in group IV (0.08046), group I (0.07731), and group II (0.06209), while the lowest mean value in group III (0.05719)

Conclusions: All types of sealers used in this investigation showed various amounts of microleakage. None of the sealants could completely close the apical foramen to provide a fluid-tight seal.

Open Peer Review

Approval Status 


1

version 1

28 Nov 2022



[view](#)

1. **Dr. Ajinkya Pawar** , Nair Hospital Dental College, Mumbai, India

Any reports and responses or comments on the article can be found at the end of the article.

Keywords

Dye extraction, PTN file, BC HI Flow, Gutta Flow Bioseal, AH Plus.

Corresponding author: Laith G. Shareef (Laithalkunani@yahoo.com)

Author roles: **Sahib Mezaal Z:** Conceptualization, Data Curation, Methodology, Project Administration, Writing – Original Draft Preparation; **Abdulkareem S:** Conceptualization, Data Curation, Methodology, Supervision, Validation, Writing – Review & Editing; **G. Shareef L:** Formal Analysis, Resources, Software, Validation, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

Grant information: The author(s) declared that no grants were involved in supporting this work.

Copyright: © 2022 Sahib Mezaal Z *et al.* This is an open access article distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

How to cite this article: Sahib Mezaal Z, Abdulkareem S and G. Shareef L. ***In vitro* apical microleakage evaluation for different endodontic sealers by spectrophotometric analysis: an observational study [version 1; peer review: 1 not approved]** F1000Research 2022, **11**:1396 <https://doi.org/10.12688/f1000research.127911.1>

First published: 28 Nov 2022, **11**:1396 <https://doi.org/10.12688/f1000research.127911.1>

Introduction

The primary goal of a root canal filling is to prevent germs and byproducts from leaking into and infecting the root canal. Apical leakage, which Ingle stated to be responsible for 59% of endodontic failures, is known to be impacted by various filling procedures, the physical and chemical properties of root canal filling materials, and the presence or absence of a smear layer.^{1,2} This highlights the importance of using materials capable of sealing off the root canal system from the periapical tissues hermetically. Microleakage studies on the sealing abilities of endodontic materials have thus become essential. Numerous root-filling methods and materials are available today, and endodontic therapy frequently employs a combination of gutta-percha points and root canal sealers.³ There are currently seven main types of root canal sealers, which can be classified based on their chemical composition. These include glass ionomer, epoxy, silicone, bioceramics, calcium hydroxide, and zinc oxide eugenol-based sealers and sealers based on mineral trioxide aggregate (MTA). All new sealants and bondable root canal filling products have to compare well to superior sealing properties of AH-Plus® (Dentsply Sirona), based on the epoxy resin.⁴ Numerous tests comparing the sealing abilities of various sealers have been conducted, and some of them revealed that the resin-based sealer provided a better seal.⁵ However, the bioceramic (BC) sealer poses some concerns since it may be affected by temperature, reducing its flow ability and setting time when heat is applied, which might negatively affect the obturation quality when a heated obturation technique is used. To address this issue, a novel sealer with the trade name TotalFill BC Sealer HiFlow was recently developed by FKG Dentaire company (FKG). The company claims that the physicochemical qualities remain constant at temperatures similar to those reached inside the root canal during heated operations around 200 C. In addition, it is more radiopaque than its predecessor; it has a lower viscosity, thinner films, and better flow properties when heated. HiFlow's biocompatibility profile is identical to the original BC sealer, according to a cell culture cytocompatibility study.⁶ Contrarily, BC sealers have been promoted for use in single-cone techniques as the material's small expansion makes it unnecessary to apply less sealant.⁷ According to other research, there is no visible difference in the depth or percentage of sealer penetration when AH plus sealers were compared with BC sealer and BC sealer HiFlow. Additionally, there was no discernible difference between the heated vertical obturation approach and the single-cone technique for sealer penetration depth or percentage.⁸ GuttaFlow® bioseal (Coltene/Whaledent, Altstätten, Switzerland), commonly known as GuttaFlow 3, is a novel root canal sealer using calcium silicate that was produced in 2015 and introduced to the market in 2016. Gutta-percha, polydimethylsiloxane, zirconium oxide, platinum, and bioactive ceramic glass make up this substance.⁹ When evaluated by a fluid filtration device, a systematic review conducted by Alghamdi and Abduljawad revealed that AH plus had the best overall sealing ability among sealers (iRoot, MTA, SealApex, Epiphany, Hybrid root seal, Endo REZ) with various obturation techniques, and further research should be done in that regard.¹⁰ To the best of our knowledge, despite studies that produced a range of findings, the present study is novel as there are no studies evaluating the sealing ability of TotalFill BC HiFlow compared with the different obturating systems. Therefore, the purpose of this research is to quantitatively analyze and compare when using a single cone approach, the apical sealing abilities of the following sealers: TotalFill BC sealer HiFlow; GuttaFlow bioseal sealer; GuttaFlow 2 sealer; and AH plus sealer. Under null hypothesis, there are no significant differences in the apical sealing ability between experimental groups.

Methods

Ethical consideration

The research protocol received formal clearance from the College of Dentistry, University of Baghdad ethics committee on 19th January, 2022 (ethics board approval code: 498522). After briefly describing the goal of the study, the researcher obtained an informed written consent form from each participant.

Study design

An *in vitro* study examined 64 freshly extracted maxillary first molars teeth with a straight palatal root canal, and mature apices¹¹ gathered from various healthcare facilities in Iraq, including Baghdad dental center, Karbala dental center, and Al-Samawa dental center during the period from 1st February 2022 to 31st May 2022.

Sample size

The size of the sample was calculated using **G*Power (RRID: SCR 013726) version 3.1.9.7**. The smallest total sample size was 59, with 90% power at a 95% confidence interval two-tailed alpha of 0.05 and an effect size of 0.80. (f). The research involved 60 freshly extracted maxillary first molars teeth with a straight palatal root canal and mature apices and 4 as a control.

Specimen preparation

To obtain a flat reference point for measuring, the palatal roots were separated vertically along their long axis at the furcation site. A standard length of 11 mm was achieved by cutting the palatal roots using a straight handpiece with diamond disc bur (Komet, Lemgo, Germany) at a speed of 30 rpm and water cooling.¹² Size 20 K-file (manufactured by

Dentsply- Sirona) was utilized to define the initial size of the canal; Protaper Next system (Dentsply) is the file system that was employed. According to the manufacturer's recommendations, file sizes of X1(0.17/0.04), X2 (0.25/0.06), X3 (0.30/0.07), and X4 (40/06) were all used at a speed of 300rpm and 260n torque.¹³ A size 20 hand K-file (Dentsply) was used to recapitulate after each rotary file to maintain the glide route and assist the lubricant in getting to the canal terminal. To eliminate debris during canal preparation, 1 mL of 5.5% sodium chloride (NaCl) (CERKAMED, Stalowa Wola, Poland) irrigation was utilized between instruments using a 30-gauge needle side vented (China, Sinalident) that was 3 mm shorter than the working length. Following instrumentation, 3 mL of 5.5% Sodium hypochlorite NaOCl was used to irrigate the canals.¹⁴ Finally, ultrasonic endo activate device (Woodpecker) was used with irrigation tip size 25 to activate 1mL of 17% ethylenediaminetetraacetic acid (EDTA) (Produits Dentaires, Vevy, Switzerland) for 1 minute, and then shaken for 1 minute with 3 mL of 5.5% NaCl and 5 mL of saline solution (Iraq) as the last rinse.¹⁵

Experimental groups

The samples were randomly split into two control groups (n = 2 for each): a positive control group (in which teeth were instrumented but not varnish-coated) and a negative control group (in which teeth were covered in varnish),¹⁶ and four experimental groups (n = 15 for each) based on the kind of sealer that was utilized; Group I: AH plus sealer (Dentsply Sirona), Group II: GuttaFlow 2 sealers) Coltène/Whaledent), Group III:

GuttaFlow bioseal sealer) Coltène/Whaledent), Group IV: TotalFill BC sealer HiFlow (FKG). Samples size collection was done according to another study.¹⁷ All the canals were dried using paper points of the appropriate size and taper (40/0.06) until a dry paper point emerged. Then, all groups were obturated using a single cone technique. Radiographs were taken to assess the quality of the obturation.¹⁸

Group I: AH plus sealer using single cone technique

A lentulo was used to introduce AH plus (manufactured by Dentsply Sirona, USA, LOT No.: 2111000958) into the root canals after being blended following the manufacturer's recommendations (Dentsply Maillefer, Ballaigues, Switzerland). Root canals were filled with gutta-percha point, 40/06, at a working length of 10 mm. After the excess material was eliminated using a hot instrument 1 mm below the access orifice, the samples were coronally sealed using glass ionomer cement.¹⁹

Group II: GuttaFlow 2 sealer using a single cone technique

The single cone approach (Gutta-percha points 40/06 + GuttaFlow 2) was utilized for the obturation operation. GuttaFlow 2 (manufactured by Coltène/Whaledent, Langenau, Germany, LOT No.: L22402) was employed in accordance with the manufacturer's instructions utilizing automixing tips and a mixing pad. First, using paper points 40/06, the canals were adequately dried, allowing GuttaFlow 2 sealer to cure fully. Then, to achieve thorough obturation, the sealer was repeatedly inserted into the canals using a 40/06 gutta-percha point until surplus GuttaFlow 2 sealer began to leak out of the canals. The gutta-percha (GP) was then extended to its maximum working length and cut off by hot ash at the level of the canal orifice. Following that, the samples get coronally sealed using glass ionomer cement.²⁰

Group III: GuttaFlow bioseal sealer using a single cone technique

To ensure that the GuttaFlow bioseal sealer (Coltène/Whaledent, Langenau, Germany, LOT No.: L04759) would fully set when the plunger was squeezed, and the component was transferred *via* specific tips onto the mixing pad, the canals were dried correctly using paper points. A single cone approach (GP size 40 taper 0.06 + GuttaFlow bioseal) was used for the obturation operation. According to the manufacturer's recommendations, the sealer was continuously put into the canals using GP until extra GuttaFlow bioseal sealer started to flow out of the channels to achieve full obturation. Finally, the samples were coronally sealed with glass ionomer cement after the GP was introduced to its entire working length and cut off by a hot instrument 1 mm below the access aperture.²¹

Group IV: Bioceramic sealer using single cone technique with bioceramic cone

The intracanal tip provided by the manufacturer was used to inject TotalFill BC sealer HiFlow (manufactured by FKG, La Chaux-des-Fonds, Switzerland, LOT No.08.650.00.008.XX) straight into the canals after being mixed following the manufacturer's recommendations. The single-cone obturation technique's size will be determined using 40/06 BC coated gutta-percha. After being gently coated with TotalFill BC sealer Hi-flow, the tip of the prefitted master cone was pushed into the prepared canal using an up-and-down pumping action until it reached its entire working length. Additional BC cone was first removed using a specialized high-speed bur provided by the manufacturer, and then they were coronally sealed using glass ionomer cement.²²

Sample preparation for microleakage evaluation and spectrophotometric analysis

To allow the sealer to cure, the obturated samples were maintained in an incubator for a week at 37°C and 100% humidity.²³ With the apical 0.5 mm being the exception, the roots were painted with two layers of nail polish (Minisu

Japan) once the obturation was complete. Each coat was allowed to dry completely before the root's surface was painted with subsequent coating.²⁴

The samples were submerged for 24 hours at 37°C and normal air pressure in a buffered solution containing 2% methylene blue (Avonchun UK). The teeth were removed from the dye and washed with tap water for 30 minutes. The varnish was removed using a surgical blade (Demophorius, Cambridge, UK) and polishing disks. Then, samples were kept in a sterile container for 3 days with 4 mL of 65% nitric acid (India). This solution was then centrifuged for 7 minutes at 4000 rpm to separate the debris from the dye that had been eliminated.²⁵

Each sample's supernatant layer was transferred in quantities of 2 mL to plastic cuvettes. Then, using concentrated nitric acid as the blank, samples were read their optical density using an automated spectrophotometer (Shimadzu Europe - UV-1650PC, Germany).²⁶

Statistical Analysis

Statistical analysis was done using IBM SPSS Statistics (RRID: SCR_016479) version 27 for Windows; The Shapiro-Wilk test was used to determine whether the data were regularly distributed. Finally, the information gathered was analyzed utilizing way ANOVA test.

Results

Data representing the spectrophotometric measurements of the dye's optical density that seeped into the samples of each group were tested for normality of distribution. The Shapiro-Wilk test results at $p > 0.05$ showed that microleakage was consistently distributed among groups. Descriptive statistics of the optical density of the dye are shown in Figure 1. The highest mean value (0.08046) was shown in group IV (TotalFill BC Sealer HiFlow), then the group I (0.07731), and group II (0.06209), while the lowest mean value was found in group III (GuttaFlow bioseal) (0.05719).

A one-way ANOVA test was carried out to identify the presence of statistically significant difference for spectrophotometric values among groups. The result of ANOVA revealed no significant difference as $p > 0.05$ among groups, as shown in Table 1.

Discussion

One of the primary goals of the various obturation procedures used in root canal therapy is three-dimensional obturation with complete closure of the whole canal system, especially the apical third. Therefore, a thorough sealing is necessary for

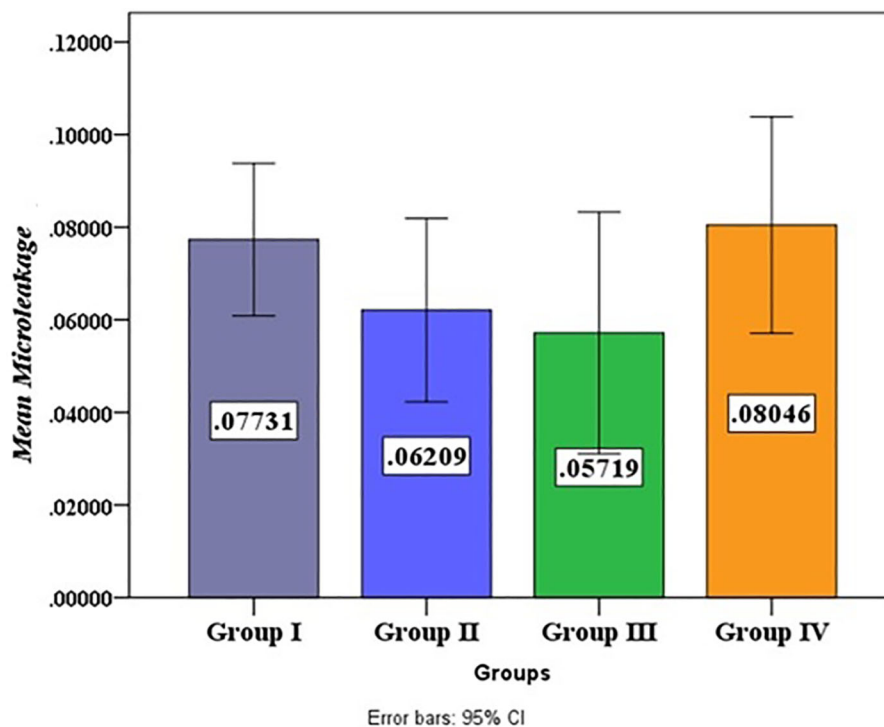


Figure 1. The mean value of the microleakage in different groups.

Table 1. Microleakage among groups using One-Way ANOVA (“analysis of variance”).

	Sum of Squares	df	Mean Square	F	P-value
Between groups	0.006	3	0.002	1.255	0.299
Within groups	0.086	56	0.002		
Total	0.092	59			

the greatest clinical outcomes and to prevent microleakage in teeth that have undergone endodontic therapy. Endodontic treatment’s effectiveness is increasing because of advancements in obturation materials.²⁷ The root canal system’s apical third is intricate, challenging to clean, and has various anatomical variations, creating significant chance of microleakage as a result.²⁸ In this study, when we compare the apical sealing ability of four experimental groups, the null hypothesis was accepted; this might be explained by various variables, including physical characteristics and the capacity to bind with the dentinal tubules to form a monobloc. The findings of this investigation were in accordance with the results of Shi *et al.*, who compared the sealing ability of AH plus sealer, GuttaFlow 2, and GuttaFlow bioseal in the apical part of the root, which shows the length of dye penetration among groups was not significantly different $P > 0.05$.²⁹

This study contradicted the conclusions of other studies, which found a substantial difference between GuttaFlow bioseal and AH plus.

When used with the single-cone obturation technique, GuttaFlow bioseal provided a more fluid-tight seal than AH plus. This could be explained by the various microleakage test techniques, the chosen teeth, or both.³⁰ Maximum microleakage was found in group IV; these results were accurate despite the 0.2 μm particle size of HiFlow. According to Alegre *et al.* study, these characteristics may be responsible for the significantly lower degree of sealer penetration in the apical third compared to the coronal third observed with the SC method.³¹ Of the treatment groups in this investigation, GuttaFlow bioseal had the least microleakage; the bioactive glass particles in the sealer, which create a tag-like structure inside dentinal tubules and at the tubule entrance, may be responsible for these results. It was determined that these crystals were hydroxyapatite, significantly improving adherence to the dentinal wall. Moreover, its limited solubility, alkalinizing activity, minimum calcium release, and ideal Ca/P ratio, which cause an anapaite layer to develop in 3 days, may contribute to its water sorption capacity to seal by volumetric expansion.³² While BC sealer’s solubility is high and doesn’t meet the recommended ISO 6876:2012.³³ Due to its sealing ability, appropriate radiopaque, high binding strength to dentine, dimensional stability, flow, low solubility, and high resistance, AH plus, a kind of epoxy resin-based sealer, was utilized in this investigation as the control. AH plus is routinely used in clinical practice.³⁴ GuttaFlow 2 shows no significant difference from AH plus. Still, lower microleakage may be due to improved handling (automix syringe), in line with another study.^{35,36} Several methods were used to evaluate leakage in endodontics. One of these methods relies on the dye being released from the contact when the tooth substrate is dissolved in acid. This method is known as the dye extraction and dissolution method. The optical density of the dye is then determined using a spectrophotometer. This method has been used in endodontic studies for a long time since it provides quantitative data, doesn’t require complex materials, and accounts for all absorbed dye in the samples, as the systematic review reveals.³⁷ This technique also reduces human measurement error, calculates the amount of leakage rather than measuring the liner, and is repeatable.²⁶

Methylene blue dye has a low molecular weight that is less than bacterial toxins; it penetrates deeply along root canal filling materials, is simple to handle, quick, inexpensive, has a high staining amount, and is not absorbed by dentine hard tissue. This makes it the ideal dye for this study.³⁸ Because it was easy to collect and standardize, and most of these roots are straight and have a single canal without an isthmus, this study employed the palatal root of the maxillary first molar. In addition, a micro computed tomography (CT) study showed that the palatal canal geometry had an unusual incidence of apical deltas.³⁹ The circular palatal root canal was divided in the furcation area to eliminate potential differences in access preparation and establish a level reference point for measurements.⁴⁰ The instrumentation and irrigation technique was the same for all the samples obtained, even though this experiment was done *in vitro* on naturally removed teeth to minimize the number of variables across groups. This experiment used distilled water, 17% EDTA, and 5.25% NaOCl to irrigate the root canal. For greater exposure of the dentinal tubules, to create a dentine substructure, and to remove the smear layer, an ultrasonic endoactivator device was utilized to stir the irrigant.⁴¹ The instrumentation methodology was the same for all the obtained samples in attempts to lessen the number of variables across groups. The Protaper next file (PTN) rotary system was used for instrumentation manufactured by Dentsply from X1(0.17/04) through X4(0.40/0.06) were the only kind of rotary file protapers utilized for instrumenting all samples to guarantee uniform instrumentation methodology for all models.³² Protaper Next’s improved tip shape and brushing action, which aid the apical file’s advancement away from outer root concavities⁴² may be the reason for the device’s high efficiency and centric ability

while cleaning and shaping root canals.⁴³ This technique also showed resistance to cyclic fatigue because of its non-uniform design and the reduced contact areas between the file and the root canal walls.^{44,45} Clinical testing is still necessary to ensure that the results have the most significant possible impact because this study's *in vitro* evaluation does not always reflect their *in vivo* performance. Another limitation is the age group from which the samples are collected is different. We recommend doing this work again using various obturation methods, such as warm vertical compaction.

Conclusion

Within the constraints of our research, when utilized with a single-cone procedure, the kind of sealer significantly impacts how well seals may be achieved. The apical foramen could not be fully sealed to prevent a fluid leak with any sealant. In terms of their capacity to seal off the apical part of the root canal, the experimental root canal sealers (AH plus sealer, GuttaFlow 2, Guttaflow bioseal, and Totalfill Hi-flow sealer) were comparable.

Data availability

Underlying data

Zenodo: Underlying data for Apical microleakage evaluation for different endodontic sealers by spectrophotometric analysis: an *in vitro* study. <https://doi.org/10.5281/zenodo.7260728>.⁴⁶

This project contains the following underlying data:

- Article data.xlsx (Measurements of Apical microleakage)

Data are available under the terms of the [Creative Commons Zero "No rights reserved" data waiver](#) (CC0 1.0 Public domain dedication).

Reporting guidelines

Zenodo: STROBE checklist for 'In vitro Apical microleakage evaluation for different endodontic sealers by spectrophotometric analysis: an observational study'. <https://doi.org/10.5281/zenodo.7322068>⁴⁷

References

1. Ingle JL, Beveridge E: *Endodontics*. Philadelphia: Lea & Febiger;3rd. ed 1985; 178–180.
2. Tanmay H, Vivek S, Rekha S, et al.: **Triglyceride profiling of ghee containing goat tallow and admixture of soy oil and goat tallow for detecting such an adulteration**. *Indian Journal of Dairy Science*. 2018; **71**(2): 176–182.
3. Economides N, Panagiotis B, Kolokouris I, et al.: **Comparative study of the sealing ability of a polydimethylsiloxane-based root canal sealer**. *Braz. Dent. J.* 2005; **16**: 145–148. [PubMed Abstract](#) | [Publisher Full Text](#)
4. Silva EJ, Rosa TP, Herrera DR, et al.: **Evaluation of cytotoxicity and physicochemical properties of calcium silicate-based endodontic sealer MTA Fillapex**. *J. Endod.* 2013; **39**(2): 274–277. [PubMed Abstract](#) | [Publisher Full Text](#)
5. Moraes TG, Menezes AS, Grazziotin-Soares R, et al.: **Impact of Immersion Media on Physical Properties and Bioactivity of Epoxy Resin-Based and Bioceramic Endodontic Sealers**. *Polymers*. 2022; **14**(4): 729. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
6. Santos JM, Coelho CM, Sequeira DB, et al.: **Subcutaneous implantation assessment of new calcium-silicate based sealer for warm obturation**. *Biomedicine*. 2021; **9**(1): 24.
7. Zhou H-m, Shen Y, Zheng W, et al.: **Physical properties of 5 root canal sealers**. *J. Endod.* 2013; **39**(10): 1281–1286. [PubMed Abstract](#) | [Publisher Full Text](#)
8. Reynolds JZ, Augsburger RA, Svoboda KK, et al.: **Comparing dentinal tubule penetration of conventional and 'HiFlow' bioceramic sealers with resin-based sealer: An in vitro study**. *Aust. Endod. J.* 2020; **46**(3): 387–393. [PubMed Abstract](#) | [Publisher Full Text](#)
9. Reszka P, Kucharski Ł, Klimowicz A, et al.: **Alkalinizing properties of selected calcium-silicate root canal sealers. An in vitro study**. *Pomeranian Journal of Life Sciences*. 2018; **64**(4). [PubMed Abstract](#) | [Publisher Full Text](#)
10. Alghamdi F, Abduljawad F: **Comparative evaluation of the apical sealing ability among different root canal obturation techniques and endodontic sealers by using fluid filtration method: A systematic review**. *IDJSR*. 2019; **7**: 1–8. [PubMed Abstract](#) | [Publisher Full Text](#)
11. Muhammed A: **Evaluation of Sealing Ability of Three Different Root Canal Sealers (in Vitro Study)**. *Journal of Al-Rafidain University College For Sciences*. 2014; **2**: 300–310. (Print ISSN: 1681-6870, Online ISSN: 2790-2293). [PubMed Abstract](#) | [Publisher Full Text](#)
12. Mounes BB, Alhashimi R: **The push out bond strength of bioceramic seal-er (Total Fill) after warm and cold obturation tech-niques An in vitro comparative**. *Journal of Baghdad College of Dentistry*. 2022; **34**(3): 7–16. [PubMed Abstract](#) | [Publisher Full Text](#)
13. Nawaf RN: **EFFICACY OF CONTINUOUS ROTATION VERSUS RECIPROCATION KINEMATIC MOVEMENTS IN REMOVING GUTTA-PERCHA WITH CALCIUM SILICATE-BASED SEALER: SEM STUDY**. University of Baghdad; 2021.
14. Marciano M, Ordinola-Zapata R, Cunha T, et al.: **Analysis of four gutta-percha techniques used to fill mesial root canals of mandibular molars**. *Int. Endod. J.* 2011; **44**(4): 321–329. [PubMed Abstract](#) | [Publisher Full Text](#)
15. Crumpton BJ, Goodell GG, McClanahan SB: **Effects on smear layer and debris removal with varying volumes of 17% REDTA after rotary instrumentation**. *J. Endod.* 2005; **31**(7): 536–538. [PubMed Abstract](#) | [Publisher Full Text](#)
16. Jafarzadeh H, Nabavi S, Shakeri M, et al.: **The Comparison of Apical Leakage between Two Obturation Techniques (Cold Lateral Condensation and Obtura II) by Using Fluid Filtration Method**. *Razavi International Journal of Medicine*. 2018; **6**(1): 42–45.
17. Najji AN, Al-Gharrawi HA: **Comparison of The Sealing Ability of GuttaFlow Bioseal with Different Obturation Systems**

- (An in vitro study). *Journal of International Dental and Medical Research*. 2020; **13**(4): 1632-1636.
18. Oliveira PS, Ferreira MC, Paula NGN, et al.: **Postoperative Pain Following Root Canal Instrumentation Using ProTaper Next or Reciproc in Asymptomatic Molars: A Randomized Controlled Single-Blind Clinical Trial.** *J. Clin. Med.* 2022; **11**(13): 3816.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 19. Altan H, Göztaş Z, Gülsüm İ, et al.: **Comparative evaluation of apical sealing ability of different root canal sealers.** *European Oral Research*. 2018; **52**(3): 117-121.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 20. Sebastian VM, Nasreen F, Junjana P, et al.: **Comparative Evaluation of Root Reinforcement Using MTA-based, Epoxy Resin-based, and Silicene-based Endodontic Sealers in Canals Instrumented with Single-file Rotary System: An in vitro Study.** *J. Contemp. Dent. Pract.* 2022; **22**(10): 1098-1104.
 21. Kaul S, Kumar A, Badiyani BK, et al.: **Comparison of sealing ability of bioceramic sealer, AH Plus, and guttaflow in conservatively prepared curved root canals obturated with single-cone technique: an in vitro study.** *J. Pharm. Bioallied Sci.* 2021; **13**(Suppl 1): S857-S860.
[PubMed Abstract](#) | [Publisher Full Text](#)
 22. Casino-Alegre A, Aranda-Verdú S, Zarzosa-López J-I, et al.: **Intratubular penetration ability in the canal perimeter using HiFlow bioceramic sealer with warm obturation techniques and single cone.** *J. Clin Exp Dent*. 2022; **14**(8): e639-e645.
[PubMed Abstract](#) | [Publisher Full Text](#)
 23. Azadi N, Fallahdoost A, Mehrvarzfar P, et al.: **A four-week solubility assessment of AH-26 and four new root canal sealers.** *Dental research journal*. 2012; **9**(1): 31-35.
[PubMed Abstract](#) | [Publisher Full Text](#)
 24. Kataia EM, Kataia MM: **Dye extraction method evaluation of the sealing ability of three types of endodontic sealers.** *Egyptian Dent. J.* 2016; **62**(1-January (Part 1)): 251-256.
[Publisher Full Text](#)
 25. Salem AS, Saleh ARM, Elmasmari HA: **Assessment of Apical Leakage of Bioceramic Endodontic Sealer with Two Obturation Techniques.** *Open Dent. J.* 2018; **12**(1): 1101-1107.
[Publisher Full Text](#)
 26. Emmanuel S, Shantaram K, Sushil KC, et al.: **An in-vitro evaluation and comparison of apical sealing ability of three different obturation technique-lateral condensation, Obtura II, and Thermafil.** *Journal of international oral health: JIOH*. 2013; **5**(2): 35.
 27. Mostafavi AS, Rasoulzadehsheikh M, Meraji N, et al.: **Bacterial microleakage in endodontically treated teeth following two methods of postspace preparation at two-time intervals: An in vitro study.** *The Journal of Indian Prosthodontic Society*. 2022; **22**(3): 233.
[Publisher Full Text](#)
 28. Alalaf N: **Comparative Assessment of Root Canal Sealer's Apical Sealing Ability.** *Al-Rafidain Dental Journal*. 2022; **22**(1): 124-135.
[Publisher Full Text](#)
 29. Shi J, Kang W, Man D, et al.: **Apical sealing of GuttaFlow Bioseal in oval-root canals.** *Chinese Journal of Tissue Engineering Research*. 2022; **26**(21): 3389.
 30. Lee S-H, Oh S, Al-Ghamdi AS, et al.: **Sealing Ability of AH Plus and GuttaFlow Bioseal.** *Bioinorg. Chem. Appl.* 2020; **2020**: 1-8.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 31. Alegre AC, Verdú SA, López JIZ, et al.: **Intratubular penetration capacity of HiFlow bioceramic sealer used with warm obturation techniques and single cone: A confocal laser scanning microscopic study.** *Heliyon*. 2022; **8**(9): e10388.
[Publisher Full Text](#)
 32. Shaikh A, Hegde V, Khandwawala N, et al.: **A Comparative Evaluation of Sealing Ability of Three Root-end Filling Materials at Different Time Intervals Using Novel Fluid Filtration Model: An in vitro Study.** *Conservative Dentistry and Endodontic Journal*. 2022; **6**(2): 45-49.
[Publisher Full Text](#)
 33. Mendes AT, Silva PB, Sô BB, et al.: **Evaluation of physicochemical properties of new calcium silicate-based sealer.** *Braz. Dent. J.* 2018; **29**: 536-540.
[PubMed Abstract](#) | [Publisher Full Text](#)
 34. Ahmed SM, Gawali KS, Paul P, et al.: **Comparative evaluation of apical sealing efficacy of guttaflow bioseal, super-bond Rc sealer and adseal root canal sealer.** *Journal of Pharmacy And Bioallied Sciences*. 2022; **14**(5): 962.
[Publisher Full Text](#)
 35. Ebert J, Holzschuh B, Frankenberger R, et al.: **Sealing ability of different versions of GuttaFlow2 in comparison to GuttaFlow and AH Plus.** *RSBO Revista Sul-Brasileira de Odontologia*. 2014; **11**(3): 224-229.
 36. El Sayed MAAM, Taleb AAA, Balbahaith MSM: **Sealing ability of three single-cone obturation systems: An in-vitro glucose leakage study.** *J. Conserv. Dent.* 2013; **16**(6): 489-493.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 37. Jafari F, Jafari S: **Importance and methodologies of endodontic microleakage studies: A systematic review.** *J Clin Exp Dent*. 2017; **9**(6): e812-e819.
[PubMed Abstract](#) | [Publisher Full Text](#)
 38. Padmanabhan P, Das J, Kumari RV, et al.: **Comparative evaluation of apical microleakage in immediate and delayed postspace preparation using four different root canal sealers: An in vitro study.** *J. Conserv. Dent: JCD*. 2017; **20**(2): 86-90.
[PubMed Abstract](#) | [Publisher Full Text](#)
 39. Marceliano-Alves M, Alves FRF, de Melo Mendes D, et al.: **Micro-computed tomography analysis of the root canal morphology of palatal roots of maxillary first molars.** *J. Endod.* 2016; **42**(2): 280-283.
[PubMed Abstract](#) | [Publisher Full Text](#)
 40. Pitts DL, Natkin E: **Diagnosis and treatment of vertical root fractures.** *J. Endod.* 1983; **9**(8): 338-346.
[Publisher Full Text](#)
 41. de Souza RA, de Castro FPL, Pires OJ: **Research of the major methods and clinical outcomes of irrigation in endodontics: a systematic review.** *MedNEXT Journal of Medical and Health Sciences*. 2022; **3**(S3).
[Publisher Full Text](#)
 42. Dhingra A, Gupta R, Singh A: **Comparison of centric ability of Protaper Next, Wave One & Protaper using cone beam computed tomography.** *Endodontology*. 2014; **26**(2): 244.
[Publisher Full Text](#)
 43. Kuzekanani M: **Nickel-Titanium rotary instruments: Development of the single-file systems.** *J. Int. Soc. Prev Community Dent*. 2018; **8**(5): 386-390.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 44. Elnaghy A: **Response: Cyclic fatigue resistance of ProTaper Next nickel-titanium rotary instruments.** *Int. Endod. J.* 2015; **48**(11): 1101.
[Publisher Full Text](#)
 45. Topçuoğlu HS, Topçuoğlu G, Akti A, et al.: **In vitro comparison of cyclic fatigue resistance of ProTaper Next, HyFlex CM, OneShape, and ProTaper Universal instruments in a canal with a double curvature.** *J. Endod.* 2016; **42**(6): 969-971.
[PubMed Abstract](#) | [Publisher Full Text](#)
 46. Shareef LG: **Underlying data for: Apical microleakage evaluation for different endodontic sealers by spectrophotometric analysis: an in vitro study.** 2022.
 47. Shareef LG: **STROBE checklist for: in vitro Apical microleakage evaluation for different endodontic sealers by spectrophotometric analysis: an observational study.** 2022.

Open Peer Review

Current Peer Review Status: 

Version 1

Reviewer Report 19 January 2023

<https://doi.org/10.5256/f1000research.140457.r160387>

© 2023 Pawar D. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

 **Dr. Ajinkya Pawar** 

Department of Conservative Dentistry and Endodontics, Nair Hospital Dental College, Mumbai, Maharashtra, India

Why were palatal roots of maxillary molars used?

The first sentences introduction and abstract (Background) are same please revise.

Try inserting the introduction in paragraphs.

"Numerous root-filling methods and materials are available today" For example?

"The company claims that the physicochemical qualities remain constant at temperatures similar to those reached inside the root canal during heated operations around 200 C" Is there any reference where the company claims this? please add.

There is no introduction of the method that is proposed to be used in the current evaluation. Please add that and also other methods used for evaluation of apical seal and then defend the current method used in Discussion. this will increase the flow of the paper for the readers in my honest opinion.

If this was an in-vitro study why was Informed Consent taken.

If so then the patient allocation is missing. How were the patients selected or enrolled?

Pertaining this the methodology is incomplete. Please rectify the methodology to best accept the in-vitro setting of the study.

For specimen preparation representative images of the steps would be interesting for the readers of the journal. In my opinion the authors can add the same in the manuscript.

How do the authors justify having only 2 root segments to serve as control??

"kept in a sterile container for 3 days with 4 mL of 65% nitric acid (India)..." the details are missing product name; company name, city, India.

The penetration of the dye could have been recorded in pictures and should have been presented in the manuscript making it more interesting.

Lastly, the discussion should defend the methodology used.

Is the work clearly and accurately presented and does it cite the current literature?

Yes

Is the study design appropriate and is the work technically sound?

Partly

Are sufficient details of methods and analysis provided to allow replication by others?

Yes

If applicable, is the statistical analysis and its interpretation appropriate?

I cannot comment. A qualified statistician is required.

Are all the source data underlying the results available to ensure full reproducibility?

Yes

Are the conclusions drawn adequately supported by the results?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Root canal treatment, root canal obturation, root canal anatomy, root canal instruments, bio ceramics, root canal irrigation, root canal classifications, covid-19

I confirm that I have read this submission and believe that I have an appropriate level of expertise to state that I do not consider it to be of an acceptable scientific standard, for reasons outlined above.

The benefits of publishing with F1000Research:

- Your article is published within days, with no editorial bias
- You can publish traditional articles, null/negative results, case reports, data notes and more
- The peer review process is transparent and collaborative
- Your article is indexed in PubMed after passing peer review
- Dedicated customer support at every stage

For pre-submission enquiries, contact research@f1000.com

F1000Research