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Sealer adaptation in the dentinal tubules: a scanning electron microscopic study

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A B S T R A C T

Aim: The purpose of this study was to evaluate the adaptability of 3 different endodontic sealers to the root canal wall using a scanning electron microscope (SEM).

Methods: Thirty extracted human single-rooted teeth were randomly classified into three equal main groups according to the type of sealer used: Tech Biosealer Endo, Flavonoid-based experimental sealer and MTA-Fillapex sealer. All root canals in this study were obturated with gutta-percha using lateral condensation technique after preparing mix from each tested sealer. The samples were examined under SEM to determine two main aspects: Gap and Interface.

Results: The result indicated that Tech Biosealer Endo had shown the best adaptation to canal walls at all root levels, followed by the MTA-Fillapex, and the most diminutive adaptation was seen in the Flavonoid-based experimental sealer. SEM showed the best adaptation for all tested sealers to root dentin was at the middle root level, followed by the apical root level, while the coronal root level showed the worst adaptation ($p<.05$).

Conclusions: SEM analysis indicated that among the tested sealers, Tech Biosealer Endo achieved the best overall adaptability to root dentin, particularly at the middle root level. This suggests that sealer composition significantly influences the quality of the interface between the sealer and root canal walls, impacting the potential for successful endodontic treatment outcomes.

1 Introduction

Successful root canal treatment requires a complete obturation of the root canal system, and the use of Gutta-percha with a sealer for root canal filling is generally accepted in endodontics (1). However, it was emphasised that Gutta-percha does not adhere to the dentinal walls (2) and could not prevent leakage by itself (3). Thus, the role of the sealer is critical for the sealing ability of obturation material, and many root canal sealers have been developed to fill residual gaps between the Gutta-percha and the canal wall(2). Mineral trioxide aggregate (MTA) was introduced as a root repair material 1993(4). Further studies revealed a successful sealing ability of MTA (5, 6, 7), and it was gradually advised for various clinical applications such as root-end filling, apexification, repair of root resorption, coronal barrier, and even as a canal-filling material (8). Tech Biosealer Endo is a recently introduced MTA-based sealer. Its powder is a mixture of white Portland cement, calcium chloride, anhydrous calcium sulfate, phyllosilicate, sodium fluoride and bismuth oxide. Its liquid is composed of Dulbecco's phosphate-buffered saline. It is essential to evaluate this sealer's characteristics, as any new dental product must be tested before being cleared for clinical use. MTA Fillapex is another MTA-based sealer mainly composed of MTA, resin (salicylate, diluting, natural), bismuth oxide, nano-particulate silica and pigments. According to the manufacturer, MTA Fillapex provides long-term sealing capacity and promotes complex tissue deposition at the root apex by including MTA. A short survey in the literature demonstrated that limited data about the dentinal wall adaptation property of MTA Fillapex are available. Propolis is a resinous material collected by honey bees from various plant species. It has attracted increased interest due to its bioactivities, such as anti-inflammatory, anti-tumour and antimicrobial activity against different pathogenic microorganisms. The precise composition of raw Propolis varies with the source. Flavonoid has been considered the main primary biologically active component in Propolis.

This study aimed to evaluate the adaptability of Propolis as an endodontic sealer (a Flavonoid-based experimental sealer) for the dentinal walls in the apical, middle, and coronal third and compare it to the newly introduced Tech Biosealer Endo and MTA-Fillapex sealer using a scanning electron microscope (9).

2 Materials and Methods

Specimen Preparation

Thirty single-rooted upper central incisors with a fully-formed apex were selected for this study. The teeth were cleaned using ultrasound to remove debris and soft-tissue remnants and were stored in distilled water until use. The crowns of the teeth were sectioned 16 mm from the apex perpendicular to the tooth's long axis using a diamond disc (Dfs, Germany) with water coolant. A size 10 K-file was used to negotiate each root canal, and the working length was established by subtracting 1mm from this length (15 mm). The canals were instrumented using a ProTaper rotary system (Dentsply Maillefer, Ballaigues, Switzerland) up to F5 and irrigated with 2ml of 2.5% Sodium Hypochlorite (Prime Dental Products Pvt Ltd), 2m of 17% ethylenediaminetetraacetic acid (EDTA-Meta Biomed Co. Ltd), and finally rinsed with 2 ml of saline and dried with paper points # 50 (Dentsply Maillefer). The thirty teeth were divided into three equal groups (n= ten each) according to the type of sealer used. Group I used Tech Biosealer Endo (Isasan SRL, Rovello Porro, CO, Italy). Group II used a Flavonoid-based experimental sealer (Imtenan Health Shop, Cairo, Egypt). While in group III, MTA-Fillapex (Angelus, Londrina PR, Brazil) was used. All the root canals in this study were obturated with a gutta-percha master cone size 50 using the lateral condensation technique after preparing a mix from each tested sealer. All sealers were mixed and manipulated according to the manufacturer's instructions except for the Flavonoid-based experimental sealer, for which 6g of Propolis powder was placed in a container, and 20 ml of a 70% ethanol solution was poured into the container and shaken briefly. The mixture in the well-sealed container was stored in a warm and dark place for one week. The container was shaken once or twice per day during the soaking period. After two weeks, the ethyl alcohol was partially evaporated by gently heating the mixture with an open container in a water bath for approximately 30 minutes at low heat (not more than 60 ºC). The evaporation continued until a homogeneous product was obtained with a paste-like consistency (10). As soon as the paste reached a creamy consistency, it was used as an experimental sealer. The coronal part of the root canal was filled with intermediate restorative material, and the teeth were stored at 37°C and 100% humidity for two weeks, allowing the complete setting of the sealers. Two longitudinal grooves were prepared on each root's buccal and lingual surface using a diamond disc without penetrating the canal. The roots were then split into two halves using a chisel. For each root, the half containing the most visible part of the apex was conserved and coded.

Specimens Treatment for Scanning

The root sections were treated with 32% phosphoric acid for 10 seconds to remove the dentin within the root before examination. The specimens were soaked in a solution of 2.5% glutaraldehyde and 2% paraformaldehyde in 0.1M sodium cacodylate buffer at a pH of 7.4 for 12 hours at a temperature of 4°C. Then, they were utterly processed using hexamethyldisilazane drying. The discs were embedded in epoxy resin and cured for 18 hours. The surface of each epoxy cast was polished using wet SiC papers of decreasing abrasiveness up to 1200 grit and soft tissue with increasingly refined diamond suspension to a size of 0.05m. The specimens were then sonicated in 100% ethanol for 5 minutes, demineralised with 6N hydrochloric acid for 30 seconds, and deproteinised with 1% sodium hypochlorite for 10 minutes. The specimens were mounted on aluminium stubs and coated with gold/palladium using a K550X sputter coater at 40mA for 90 seconds. The interfaces were observed under the SEM Model Quanta 250 FEG (Field Emission Gun) at two different magnifications, 500x and 1000x. A qualitative evaluation of the dentin sealer interface was carried out. The penetration of the sealer in the dentinal wall irregularities, presence of gaps between the sealer and the dentinal walls, presence of voids or air bubbles, adaptation of the sealer and its proximity to the walls, the homogeneity of the sealer through its thickness, and the presence of cracks in the sealers' bulk were studied. The photographs were taken at the root's coronal, middle, and apical levels, and the findings were recorded.

Measurement of Gaps:

The samples were examined under SEM to determine gaps. Images of each root canal third (coronal, middle, and apical) were examined under magnification of 1000X and were then subjected to image analysis using Image J 1.41a software for Windows. The surface area

Table (1): Mean percentage surface area of gaps for three sealers at all root levels.

Sealers / Root Third	Tech Biosealer Endo	Flavonoid-based	MTA-Fillapex
Coronal (%)	0.1893± 0.017 Za	0.3742± 0.098 Xa	0.2924± 0.039Ya
Middle (%)	0.1185± 0.021 Zb	0.2731± 0.051 Xb	0.2206± 0.0.47 Yb
Apical (%)	0.1027± 0.032 Zb	0.2624± 0.048 Xb	0.2173± 0.019 Yb

The same small letters within the same column indicate the non-significant difference
The same capital letters within the same row indicate the non-significant difference
P-value was set at $P \leq 0.05$.

of gaps was then calculated as a percentage of the total surface area examined of the sealer.

Measurement of Interface:

The study examined the sealer-dentin interface using SEM at different magnifications (500X and 1000X) to assess two factors: the interfacial layer formation and intratubular sealer penetration. Each sample was assigned a score between 0 and 2 based on the following criteria (12, 13):

- 0: no interfacial layer at the sealer-dentin interface

- 1: the presence of an interfacial layer at the sealer-dentin interface

- 2: intratubular sealer penetration

Unaware of the study, two examiners made the observations and repeated them twice to ensure consistency.

3 Results

Gaps

Tech Biosealer Endo showed the highest percentage of gaps, typically 0.1893 ± 0.017 %, followed by the middle root third 0.1185 ± 0.021 %. The apical root third showed the lowest percentage of gaps, 0.1027 ± 0.032 %. Flavonoid-based experimental sealer showed the highest percentage of gaps coronally at 0.3742 ± 0.098 %, followed by the middle root third at 0.2731± 0.051 %. At the same time, the apical root third showed the lowest percentage of gaps, 0.2624 ± 0.048 %. Similarly, MTA-Fillapex showed the highest percentage of gaps, coronally 0.2924 ± 0.039%, followed by the middle root third 0.2206 ± 0.0.47 %. At the same time, the apical root third showed the lowest percentage of gaps, 0.2173 ± 0.019 %. Flavonoid-based experimental sealers showed statistically significant gaps at all root levels, followed by MTA-Fillapex. At the same time, the Tech Biosealer Endo showed statistically significantly the lowest percentage of gaps at all root levels. The coronal root level for three sealers showed the highest percentage of gaps than the other two levels, with the difference statistically significant (Table 1, Fig. 1).

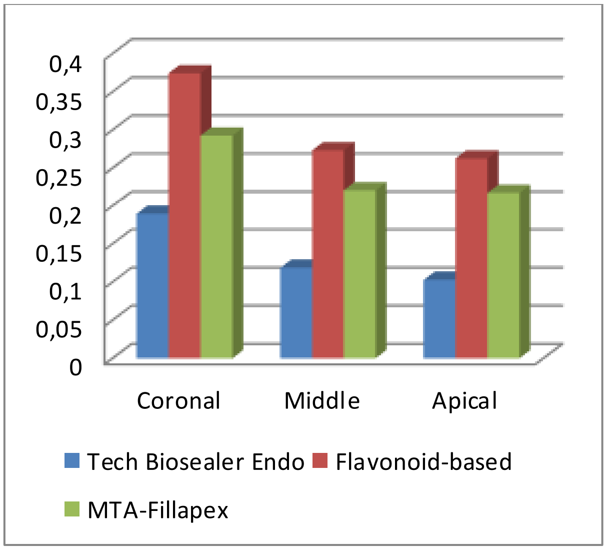

Figure (1): Mean percentage of gap surface area for three sealers at all root levels

Interface

The adaptability scores for Tech Biosealer Endo were reported as follows: 1.8 ± 0.4216 for the coronal third and 1.9 ± 0.316 for both the middle and apical thirds. This sealer exhibited superior adaptation in the middle and apical sections compared to the coronal section, which showed lesser adaptation (refer to Table 2 and Figures 2, 3, 4). The Flavonoid-based experimental sealer scored 1.1 ± 0.316 at the coronal third, 1.3 ± 0.483 at the middle third, and 1.2 ± 0.421 at the apical third. This sealer achieved its best adaptation at the middle third, followed by the apical and then the coronal third, with the most diminutive adaptation (see Table 2 and Figure 5). MTA-Fillapex recorded scores of 1.3 ± 0.483 at the coronal third, 1.6 ± 0.516 at the middle third, and 1.4 ± 0.516 at the apical third, indicating progressively better adaptation from the coronal to the apical third (detailed in Table 2 and Figure 6). Among the sealers, Tech Biosealer Endo showed the highest overall adaptability to canal walls across all root levels, followed by MTA-Fillapex, with the Flavonoid-based experimental sealer showing the least. Scanning electron microscopy confirmed that the middle root level exhibited the best sealer adaptation for all materials, followed by the apical level, while the coronal level had the poorest adaptation. Significant differences were observed in the adaptability across all root levels between the sealers (as outlined in Table 2 and Figure 2).

Table (2): Mean and median of adaptability scores recorded at different root thirds for three sealers.

Root Third	Sealers	Tech Biosealer Endo	Flavonoided -based	MTA-Fillapex
Coronal	Mean	1.8 ± 0.4216 ^{Xa}	1.1 ± 0.316 ^{Za}	1.3 ± 0.483 ^{Ya}
	Median	2	1	1
Middle	Mean	1.9 ± 0.316 ^{Xa}	1.3 ± 0.483 ^{Ya}	1.6 ± 0.516 ^{Xa}
	Median	2	1	2
Apical	Mean	1.9 ± 0316 ^{Xa}	1.2 ± 0.421 ^{Za}	1.4 ± 0.516 ^{Ya}
	Median	2	1	1

The same small letters within the same column indicate the non-significant difference.
The same capital letters within the same row indicate the non-significant difference.
P-value was set at $P \leq 0.05$.

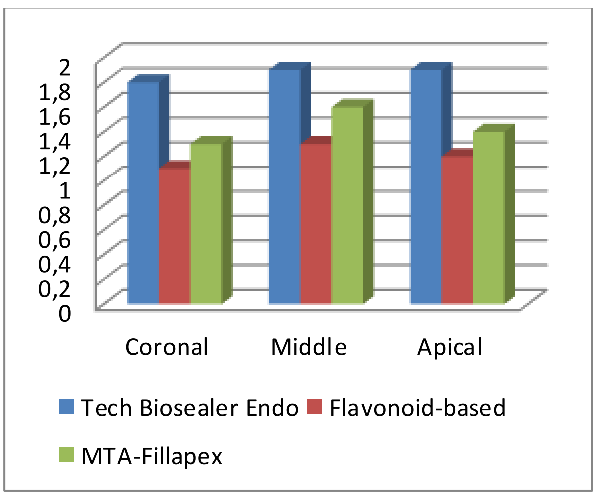

Figure (2): Mean adaptability score for three sealers at all root levels

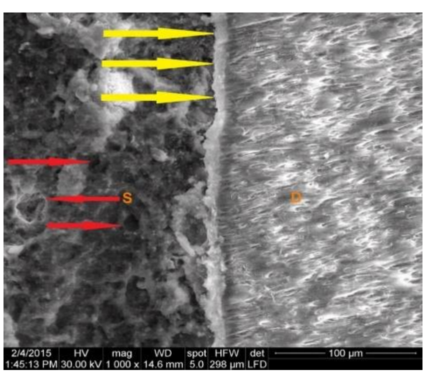

Figure (3): Photomicrograph of the Tech Biosealer Endo/dentin interface at the middle third (score 2) showing uniform interfacial layer with resin tag formations (yellow arrow) and a few gaps (red arrows). D, dentin. S, sealer
D, dentin. S, sealer

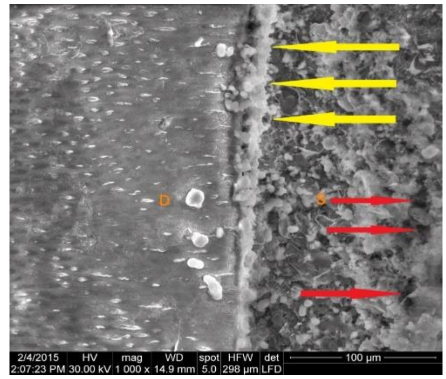

Figure (4): Photomicrograph of the Tech Biosealer Endo/dentin interface at the apical third (score 2) showing uniform interfacial layer (yellow arrow) and gaps (red arrows). D, dentin. S, sealer

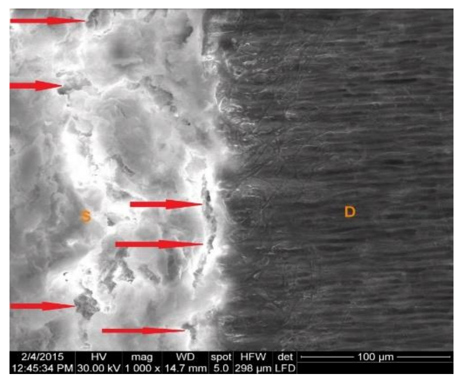

Figure (5): Photomicrograph of the Flavonoid-based experimental sealer /dentin interface at the coronal third showing gaps at the interface (score 1). Red arrows indicate gaps. D, dentin. S, sealer.

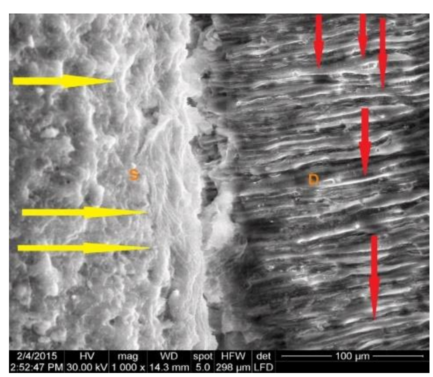

Figure (6): Photomicrograph of the MTA-Fillapex /dentin interface at the middle third (score 2) showing uniform interfacial layer (yellow arrows) with intratubular penetration (red arrows). D, dentin. S, sealer

4 Discussion

The success of endodontic treatment heavily relies on effective root canal obturation. The widely accepted method employs gutta-percha cones in conjunction with sealer cement. These sealers serve as adhesives, conforming the gutta-percha to the irregularities of the canal walls and filling any voids. However, leakage between the sealer and canal walls is a known issue, underscoring the importance of a sealer's ability to bond effectively to tooth structures. Grossman (14) outlines that an ideal sealer should offer an excellent seal, dimensional stability, extended working time, resistance to bodily fluids, strong adhesion, and biocompatibility. The therapeutic and biological virtues of propolis, a natural product, have gained attention in recent research, reinforcing its traditional use in folk medicine and endodontics for applications like pulp regeneration (15) and rigid tissue bridge formation (16). Known for its robust antimicrobial and anti-inflammatory properties, propolis is a rich source of flavonoids, which has influenced its designation as a Flavonoid-based experimental sealer. Similarly, MTA, a calcium silicate cement noted for its ability to set in biological fluids (17), has seen widespread use. Newer formulations like Tech Biosealer Endo and MTA-Fillapex have been developed to leverage these properties, aiming for improved biocompatibility and sealing characteristics.

Adherence to the root canal wall is crucial for sealers (18). Tay et al. (19) emphasise the significance of the sealer/dentin interface as the critical zone of sealing. Our study employed a lateral compaction technique to simulate real-life application conditions and assessed sealer adaptation across three segments of the root canal—coronal, middle, and apical—using a scanning electron microscope (SEM). This method provides a high-resolution, magnified interface view, allowing for detailed analysis. We removed the smear layer with alternating rinses of 17% EDTA and 2.5% NaOCl to ensure optimal sealer penetration.

Our findings indicated that the Flavonoid-based experimental sealer exhibited the highest gap percentages, followed by MTA-Fillapex, with Tech Biosealer Endo showing the least. This could be linked to their solubility characteristics. The lateral compaction method, which applies less force at the coronal level, likely contributed to the higher gap percentages observed. Notably, Tech Biosealer Endo displayed superior interfacial adaptability and intratubular penetration across all root levels, corroborated by its phyllosilicate content, known for its swelling properties upon hydration (21, 22), which enhances sealer expansion and adaptation. Sodium fluoride in Tech Biosealer Endo might also enhance its expansion properties, ranging from 0.10% to 6.72% (24, 25, 26), thus improving sealing effectiveness.

In contrast, the low MTA content in MTA-Fillapex may account for its less favourable results. The adaptation and penetration qualities were generally better at the middle root level than the apical third, possibly due to easier removal of the smear layer and the structural characteristics of the dentine at different root levels. These findings align with previous research by Al-Haddad et al. (28). Prati et al. (29) and provide a contrast to studies that employed different methodologies, such as those by Ozasir et al. (32) and Kuci et al. (33). In conclusion, Tech Biosealer Endo demonstrated the most effective tubular penetration and adaptation to the canal walls among the sealers tested, particularly at the middle root level. This was followed by the apical level, with the coronal level showing the most diminutive adaptation, reinforcing the importance of material composition and application technique in achieving optimal endodontic sealing.

5 Conclusions

Based on the conditions and findings of this study, the following conclusions can be drawn:

1. Among the sealers evaluated, Tech Biosealer Endo demonstrated superior tubular penetration and adaptability to the root canal walls, indicating its potential effectiveness in enhancing the seal and preventing microleakage in endodontic treatments.

2. For all sealers tested, adaptability to root dentin was most favorable at the middle root level, with the apical level also showing significant adaptation. Contrastingly, the coronal root level exhibited the least adaptability. This gradient in adaptation effectiveness underscores the importance of sealer selection and application technique based on the specific root canal region to optimize treatment outcomes.

Conflict of interest:

There are no financial, personal, or professional conflicts of interest to declare.

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