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A Comparative Evaluation of the Sealing Ability, Adaptability and Antibacterial Effect of Nano MTA Plus Versus Conventional MTA Fillapex: An *In-Vitro* Study

Marina S. Fahmy[1]*, Maged M. Negm[2], Noha A. Hassuna[3], Mohammed Turky[1]

[1]*Departmemt of Endodontics, Faculty of Dentistry, Minia University, Minya, Egypt;* [2]*Department of Endodontics, Faculty of Dentistry, Cairo University, Giza, Egypt;* [3]*Associate Professor of Microbiology and Immunology, Faculty of Medicine, Minia University, Minya, Egypt*

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***Correspondence:** Marina S. Fahmy, Departmemt of Endodontics, Faculty of Dentistry, Minia University, Minya, Egypt. E-mail: marinashaker55@gmail.com
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Abstract

AIM: To compare nano-mineral trioxide aggregate (MTA) Plus with MTA Fillapex sealers regarding their sealing ability, adaptability, and antibacterial effect.

METHODS: Forty freshly extracted sound single-rooted permanent maxillary central incisors were used. Following access cavity preparation and root canal preparation. All the samples (n = 40) were randomly and equally categorized into two groups (n = 20 each) according to the sealer employed: group I, in which MTA Fillapex was utilized, and group II, in which nano-MTA Plus was applied. Obturation of root canals was accomplished using the lateral compaction technique. Decoronation of all teeth was performed for standardization of the root length at 12 mm. Roots were submerged in a 2% methylene blue dye solution for 72 h. The roots were divided lengthwise. Linear apical dye penetration and gaps between gutta-percha and dentin were measured under a stereomicroscope at 30× magnification to determine the sealing ability and adaptability, respectively. While the antibacterial activity against *Enterococcus faecalis* was tested using the Agar Diffusion Test (ADT). Data were statistically estimated utilizing the Mann-Whitney test, Spearman's correlation coefficient examination, and the independent T-test.

RESULTS: Although Nano-MTA Plus showed higher sealing ability and lower antibacterial action than MTA Fillapex, we didn't record any statistically substantial variation (2.05 ± 1.43 mm and 11.00 ± 9.42 mm, respectively), (2.10 ± 1.17 mm and 14.25 ± 6.70 mm, respectively). While statistical significance was reported in adaptability, Nano-MTA Plus showed a statistically significant lower adaptability compared with MTA Fillapex (57.89 ± 23.45 μm and 26.82 ± 16.55 μm, respectively).

CONCLUSION: Nano-MTA Plus and MTA Fillapex have comparable sealing abilities, and antibacterial effects. While MTA Fillapex was superior to Nano-MTA Plus regarding adaptability.

Introduction

Endodontic therapy success is dependent on bacterial eradication and root canal obturation [1]. Because none of the existing root canal filling materials could establish a hermetic seal, they must have antibacterial properties to prevent infection. Multiple root canal-filling substances with similar qualities have been introduced, but none are optimal [2]. Apical sealing ability is the capability of a sealer to give an apical closure to avoid leakage in the root canal system [3]. The majority of endodontic failures are triggered by poor root canal cleaning and the egress of bacteria and other antigens into the periradicular tissues as a result of microleakage at the sealer-dentin and sealer-core component interfaces. As microorganisms are the most prevalent trigger of endodontic failure [4], it is critical to get rid of microbes from the root canal network [1]. Nevertheless, it is a challenge. *Enterococcus faecalis* is recognized as "the star survivor in the root canal" and is the most prevalent microbe isolated from apical periodontitis [2].

Mineral trioxide aggregate (MTA) has been regarded as an effective root canal filler substance owing to its outstanding sealing ability, biocompatibility, and capacity to induce biomineralization [5]. MTA Fillapex (Angelus, Londrina-Parana, Brazil) is a novel root canal sealer involving nanoparticulated silica, salicylate resin, bismuth trioxide, diluting resin, natural resin, MTA, and colors. According to the manufacturer, MTA Fillapex provides long-term sealing capacity and promotes the deposition of hard tissue at the root apex. A brief review of the literature revealed that there is little information available on MTA Fillapex's dentinal wall adaptability capability [6].

Recently, nanotechnology has made it possible to create nanoscale dental materials with boosted physicochemical characters. So, Nano-MTA Plus was introduced to enhance the physical traits of the substance to get the maximal biological and clinical advantages [7]. Therefore, our experiment was designed to compare Nano-MTA Plus with MTA Fillapex sealers concerning the sealing ability, adaptability, and antibacterial effect. The null hypothesis would be that there will be no variation between MTA Fillapex and Nano-MTA Plus sealers regarding sealing ability, adaptability, and antibacterial effect.

Materials and Methods

Sample size calculation

According to a previous experiment by Al-Hezaimi *et al*. [8]. A minimum overall specimen amount of 40 specimens was sufficient to estimate the influence size of 0.74, with a power ($1 - \beta = 0.90$) at a significance probability grade of $p \leq 0.05$ to compare the sealing ability, adaptability, and antibacterial effect of Nano-MTA Plus with those of MTA Fillapex. According to the sample size calculation, with 20 samples for each group, there was a 90% chance of properly refusing the null hypothesis of no substantial impact of the interaction. The specimen size was assessed based on G*Power software version 3.1.9.7, depending on Cohen's d equation to estimate the impact size.

$$d = \frac{\overline{X_1} - \overline{X_2}}{\sqrt{\frac{S_1^3 + S_2^3}{2}}}$$

Cohen's d equation.

Samples selection

The present research was accepted by the Research Ethical Approval Committee, Faculty of Dentistry, Minia University, Minya, Egypt (Meeting No. 69, Date: 24/2/2020). The study was carried out on 40 recently extracted, intact maxillary central incisors with single, straight root canals and completely developed apices. The chosen teeth were freshly extracted for periodontal causes at the outpatient clinic, Faculty of Dentistry, xxxxxx University. The teeth were washed and scaled to get rid of soft and hard deposits. Teeth were stored in a 0.1% thymol (Formula Acao, Sao Paulo, SP, Brazil) solution until use.

Samples preparation

Conventional access cavity was performed by a round diamond bur. Patency was established by using a manual stainless steel K-file ISO size # 8-10. Canals were prepared employing the crown-down method by M-pro (Alex Dent, Shanghai, China) rotary files (18/0.04, 20/0.04, and 25/0.06). Shaping and cleaning were completed up to a master apical file ISO size #40 using a manual stainless steel K-file. 1 ml of 5.25% sodium hypochlorite solution (NaOCl) was used to irrigate the canals between successive files. The last irrigation was accomplished by 5 ml of 5.25% NaOCl, 5 ml of saline solution, and 5 ml of 17% ethylene diamine tetraacetic acid solution for 3 min.

Samples classification

All samples (n = 40) were allocated into two equal groups (n = 20 each) depending on the type of sealer employed. Group I (n = 20), in which root canals were filled using gutta percha and MTA Fillapex sealer (Angelus, Londrina, BR, Brazil), while in group II (n = 20), root canals were obturated with gutta percha and Nano-MTA Plus sealer (Nano Gate, Cairo, Egypt). The cold lateral compaction technique was used, and then the samples were stored at 37°C with 100% humidity for 7 days to allow the sealer to set.

Sealing ability and adaptability testing

The teeth were measured and decoronated by means of a double-sided diamond disc to obtain a standard root length of 12 mm, except for the apical border. All root surfaces were coated with two layers of nail varnish before being submerged in a 2% methylene blue dye solution for 3 days. Roots were longitudinally split using a diamond disc and an Ochsenbein chisel (Dentsply, Maillefer, and Charlotte, USA). An apical dye penetration test was applied to assess the sealing ability, while adaptability was tested by measuring the gap distances between gutta percha and dentinal walls using a stereomicroscope at 30× magnification.

Antibacterial test

Sealers were tested for their antibacterial effect against *E. faecalis* by employing the Agar Diffusion Test (ADT). Culturing bacteria in brain heart infusion (BHI) agar in anaerobic condition at 37°C for 3 days, then 3–4 colonies were selected and subcultured in 3 mL of a BHI. Punch holes of 6 mm in diameter were made on every agar plate, and then the plate was filled with MTA Fillapex, while the other plate was filled with Nano-MTA Plus. The agar plates were incubated at 37°C for 1 day before measuring the diameters of the inhibitory growth areas with a Vernier caliper (Tolsen, Zhangjiagang, Jiangsu, China). The diameters of the inhibitory growth regions were computed for each group and statistically analyzed to assess the existence or lack of substantial variations in antibacterial efficacy.

Statistical analysis

Statistical analysis was conducted for parametric data and the Agar Diffusion Test (ADT) for antibacterial effect results by applying an independent T-test. Comparing the sealing ability and adaptability, results among the two studied divisions were done using the Mann-Whitney test (non-parametric test).

Spearman's correlation coefficient examination was applied to study the correlation among the three parameters (dye penetration, gap diameter, and inhibition zone diameter) (Figure 1).

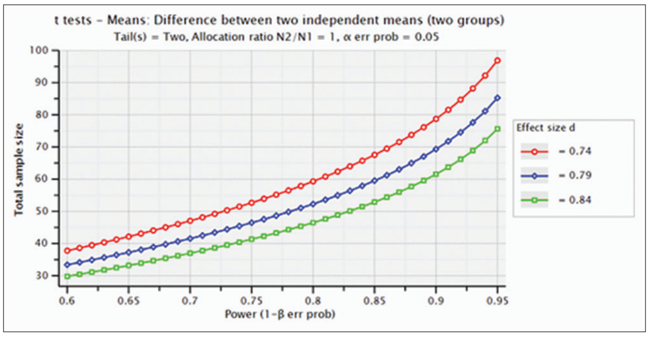

Figure 1: Independent t test total sample size calculations, number of groups (n=2)

Results

The mean of dye penetration (apical sealing ability) was 2.10 ± 1.17 mm in the MTA Fillapex group and decreased to 2.05 ± 1.43 mm in the Nano-MTA Plus group. Based on the Mann–Whitney test, the variation among the two groups was not statistically significant (p > 0.05) (Figure 2).

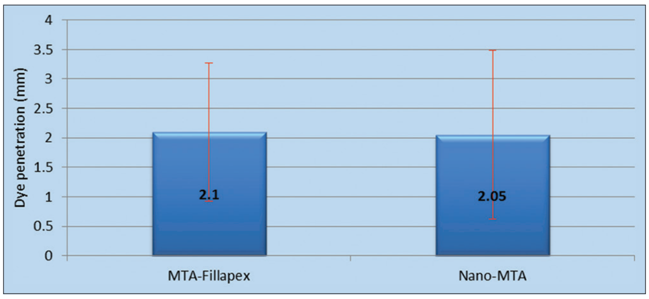

Figure 2: Bar chart depicting the mean and SD of dye penetration for the studied groups

The mean gap distance (adaptability) was (26.82 ± 16.55 μm) in the MTA Fillapex group and increased to (57.89 ± 23.45 μm) in the Nano-MTA Plus group. Based on the Mann-Whitney test, the variation among the two groups was statistically significant ($p \leq 0.001$) (Figure 3).

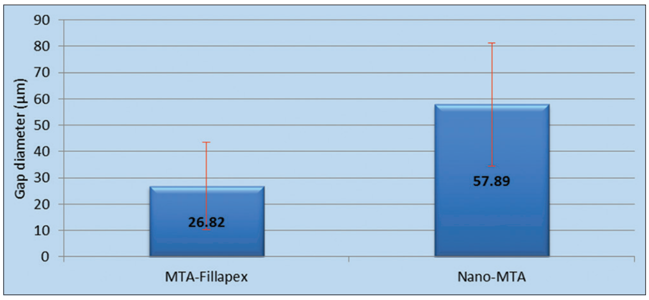

Figure 3: Bar chart depicting the mean and SD of gap diameter for the studied groups

The mean inhibition zone diameter (antibacterial effect) was 14.25±6.70 mm in the MTA Fillapex group and 11.00 ± 9.42 mm in the Nano-MTA Plus group. Based on the independent T-test, the variation among the two groups was not statistically significant (p > 0.05) (Figure 4).

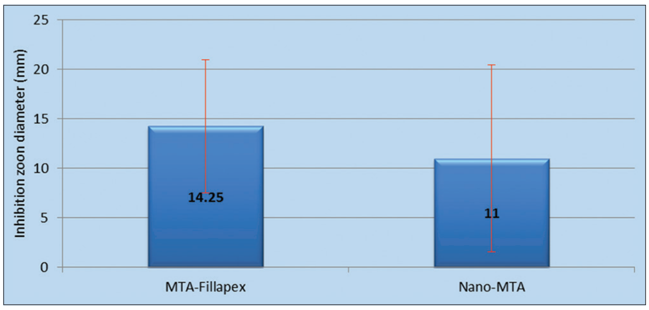

Figure 4: Bar chart depicting the mean and SD of inhibition zone diameter (mm) for the studied groups

Discussion

The most prevalent reason for endodontic failure is the spread of bacteria from root canals into periapical tissues. A very tight seal should be created to restrict the communication channel among root canal systems and periradicular tissues. The common factors associated with failure are survival of bacteria, improper obturation of the canal, overextensions of root filling materials, inadequate coronal seal, and methodological errors like improper access cavity design ledges, perforations, separated instruments, untreated accessory canals, and missed canals [9]. Among all these causes of endodontic failure, one of the principal causes is persistent microbial infection [10]. Root canal sealers are required for the formation of a bond between the obturating core component and the root canal wall, as well as the sealing of voids, auxiliary canals, apical ramifications, and numerous foramina. As a result, the interaction between the sealer and the dentin is crucial [11]. Yet, the adaptability of sealers on dentin interfaces without voids relies on sealers' physical characters [12]. Despite the sealing capability of root canal filling substance being a significant concern for endodontists [13], the flow of the sealer is a key quality for increasing its adaptability to penetrate into the root canals, minor abnormalities and ramifications, as well as the dentinal tubules [14].

The antimicrobial activity of root canal sealers could aid in removing residual bacteria untouched by chemomechanical solutions of the root canal procedure, e.g., *Enterococcus faecalis* [15]. MTA was presented as a root repair substance in 1993 [16]. More investigations discovered the reliable sealing capacity of MTA [17], [18], [19], and hence it was increasingly recommended for a variety of therapeutic purposes like coronal barrier, root-end filling, root resorption repair, apexification, and even canal-filling material [6]. The MTA-based sealers are novel endodontic components that depend on the physiochemical traits of MTA. MTA Fillapex is a recent MTA-based root canal sealer.

Nanoparticles are natural or manmade compounds that comprise particles in an unbound state or agglomerates with at least half of the compounds in the 1–100 nm size range. They have distinct features because of their tiny size, supplying them with a higher surface area and a various chemical reactivity than their bulk counterparts [20]. Trying to enhance the sealer's physical properties to obtain the maximum biological and clinical advantages, Nano MTA Plus was introduced [20]. So, our research tried to compare the sealing ability, adaptability, and antibacterial effect of MTA-Fillapex and Nano-MTA Plus as root canal sealers. Single-rooted teeth with a single root canal were utilized to standardize and eliminate variations that may arise from numerous canals and complicated curved canal shapes [21].

The cold lateral condensation technique remains one of the most favorable root canal obturation methods [22] because of its predictability, convenience of use, and conservative preparation, as well as features such as regulated positioning of gutta percha in the root canals. Mimicking clinical practice, NaOCl was used at a concentration of 5.25% in between each file and the next in a final rinse regimen, followed by EDTA solution for 1 min [23], [24]. This was supported by the experiment of Taylor *et al*. [25], who demonstrated that removing the smear layer reduces leakage independent of the obturation method utilized.

Due to its ease of employment, sensitivity, and convenience, the dye penetration test is the most commonly used method [26]. Methylene blue dye has a low molecular weight, a strong staining capacity, and a high penetrating ability [27], [28]. In the current investigation, the methylene blue dye penetration test was employed. In endodontics, a stereo-electron microscope (SEM) is utilized for the ultrastructural study of the root dentin exposed to interface filler material or the dentin wall, and this approach allows the existence of gaps to be evaluated [29]. Although the limitations of the agar diffusion test are well recognized, it is a semi-quantitative approach that cannot differentiate between the bacteriostatic and bactericidal influences of a component [30], [31].

In the present study, although both materials showed comparable sealing ability, MTA Fillapex showed better adaptability than Nano-MTA Plus sealer, which might be attributed to the fast dehydration of Nano-MTA Plus sealer, which was noticed during the mixing of powder and liquid, decreasing its flow between dentinal walls and gutta-percha. The good sealing ability and adaptability of MTA Fillapex sealer might be due to its elevated flow frequency (27 mm) and reduced film thickness, which in turn facilitate its penetration into the lateral and accessory canals, regardless of the obturation procedure. According to Camilleri *et al*. [32] and Singh *et al*., [33] MTA Fillapex is capable of expanding and enhancing its sealing efficacy over time, especially if 17% EDTA is applied as a last irrigant. MTA produced a porous structure upon hydration. Levels of ettringite and monosulfate were low. Bismuth oxide was present as an unreacted powder but was also incorporated with the calcium silicate hydrate. Since it aids in the fusion of the smear layer to the MTA Fillapex sealer mass and provides volume to the sealer that would probably penetrate the dentinal tubules.

It was, as far as we know, a pioneering assessment to compare the sealing ability and adaptability of MTA Fillapex and Nano-MTA Plus as root canal sealers.

Although numerous studies reported the excellent antibacterial properties of MTA Fillapex because of the elevated pH (9.5–10.4) for elongated time and the tendency toward maintaining the Ca^{2+} release nearly constant for up to 2 weeks [34].

The present results were also contradictory to Estrela *et al*., [35] who reported that MTA Fillapex did not exhibit any antimicrobial role against *E. faecalis* and *C. albicans*. Ramezani *et al*. [36] reported that mixing MTA with CHX 0.12% instead of distilled water increased the sealing ability and increased its antibacterial effect against *E. faecalis.* Our present study was also contradictory to Nashat *et al*., [37] who showed the presence of antibacterial properties of MTA, but this property decreased in nano-form. This is thought to be due to the decreased setting time of the nano-formed materials, which reduced the pH and released radicals during the setting reaction of the present tested nano-form materials.

The null hypothesis was partially accepted as both MTA Fillapex and Nano-MTA Plus had comparable sealing ability and antibacterial effects, while MTA Fillapex was superior to Nano-MTA Plus regarding adaptability.

Despite the low level of evidence from in vitro studies, it allows the standardization of many variables that could not be adjusted in a clinical situation.

Conclusion

Nano-MTA Plus and MTA Fillapex have comparable sealing abilities and antibacterial effects. While MTA Fillapex is superior to Nano-MTA Plus in respect of adaptability.

Recommendations

1. Future studies using a non-destructive three-dimensional (3D) approach, such as

micro-computed tomography (micro-CT), will need more accuracy and clinical relevance.

2. Further clinical studies, comparing both sealers, are required to determine their influence on the outcome of non-surgical root canal treatment.

Disclosures

Ethics committee approval

The present research was accepted by the Research Ethical Approval Committee, Faculty of Dentistry, Minia University, Minya, Egypt (Meeting No. 69, Date: 24/2/2020).

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