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Sealing Ability and Adaptability of Nano Mineral Trioxide Aggregate as a Root-End Filling Material

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Abstract

AIM: The aim of the study was to study the comparison between nano-MTA and MTA as a root-end filling materials regarding adaptability and sealing ability.

MATERIALS AND METHODS: Forty extracted human maxillary incisors with straight roots were used. After root canals preparation and obturation, the apical 3 mm of each root was resected perpendicular to the long axis of the tooth. Root end cavities were prepared to a depth of 3 mm parallel to the long axis of the tooth. The teeth were randomly divided into two main equal groups of 20 samples each according to the root-end filling material used either MTA or nano-MTA. Ten samples from each group were sectioned longitudinally into two equal halves to measure the sealing ability and another ten samples from each group were sectioned transversally to obtain 1 mm thick section to measure the adaptability of both materials. All samples were photographed under the SEM at three different magnifications (×1000). The gap thickness between the root end filling material and the retro cavity dentine walls were measured at seven selected points at the material-dentine interface in micrometers (μm).

RESULTS: Nano-MTA and MTA showed no statistically significant difference in the gap thickness between dentin-material interface in both longitudinal and transverse sections. Regarding the sealing ability, the mean value in MTA was (3.27 ± 0.77), while the mean in nano-MTA was (3.15 ± 0.71). Regarding the adaptability, the mean value in MTA was (2.46 ± 0.60), while the mean in nano-MTA was (2.05 ± 0.712). Both materials showed good sealing ability and good adaptation to the dentinal wall.

CONCLUSION: Nano-MTA revealed good sealing ability and adaptability comparable to MTA when used as a retrograde filling material.

Introduction

Surgical endodontic treatment is an option for teeth with apical periodontitis and may be indicated for teeth with periapical pathology when non-surgical retreatment is impractical or unlikely to improve the previous results or when a biopsy is needed [1], [2]. The placing of a root-end filling after root-end resection is mandatory to support bone regeneration. Various materials have been advocated for use as retrograde filling in which the main aim is to be able to prevent the infiltration of bacteria into the root canals [3], so root-end filling materials should have appropriate sealing ability and adaptability as the amount of microorganisms in the infected area is a key factor determining the healing or persistence of the apical lesion [4].

Mineral trioxide aggregate (MTA) has been introduced in endodontics in 1993 by Mahmoud Torabinrjad for its advantageous properties of high biocompatibility; especially its ability to enhance cementogenesis, promoting healing, superior sealing ability, and ability to set in the presence of moisture. The literature has addressed the need for osteopromotion for materials to become more osteogenic, osteoconductive, and osteoinductive [5]. The development and modification of dental cements have taken place for many years to bring about an optimal interaction between hard tissues and dental cements. Nanotechnology has enabled the production of nanoscale dental materials with improved physicochemical properties. Moreover, the hard tissue response and osteopromotion of MTA cements may be improved by adding nano-additives to their composition [5]. Thus, the aim of the present study was to compare between nano-MTA and MTA as a root-end filling materials regarding adaptability and sealing ability.

Materials and Methods

Based on a previous study [6], the sample size was calculated and found to be 10 extracted teeth, this number is increased to a total of 20 (20 teeth per group) to compensate for losses during evaluation by scanning electron microscope. The mean and standard deviation of gap of MTA as a root end filling material was found to be 4.48 ± 0.12, assuming the nano-MTA will reduce the mean of gap to 4.32, using t-test with the assumption of a type I error of 0.05, and the power of 0.8. The minimal sample size accepted is 10 per one group (total 40 for both groups regarding adaptability and sealing ability) using P.S version 3.1.6.

Total of forty maxillary anterior single-rooted teeth were freshly extracted for orthodontic or periodontal purposes from the oral surgery department, Faculty of Dentistry, Ain-Shams University and Faculty of Oral and Dental Medicine, Future University in Egypt. Teeth were selected according to the eligibility criteria as listed in Table 1.

Table 1: Eligibility criteria for teeth selection

Inclusion criteria	Exclusion criteria
Teeth with intact broad root and mature apecies	Teeth with evidence of root fracture defects and cracks (using preoperative periapical radiograph mesiodistally and buccolingually).
Teeth with single root and single root canal (Type I) according to Vertucci classification.	Teeth with any abnormalities as internal or external resorption or root canal calcification.
Root canals showing normal anatomy without (Dilacerations, severe curves, calcifications, any type of root canal other than type I).	Teeth with open apex.
Teeth with adequate root length.	Teeth with previous root canal treatment.

All teeth were scaled using ultrasonic scaler to remove any calculus deposits, stains, and any organic debris from root surface. Samples were stored in clean glass bottle containing normal saline solution at room temperature before experiment until usage.

Biomechanical preparation of teeth

The anatomical crowns of all the teeth were amputated horizontally at the CEJ using a diamond disc mounted on a micromotor handpiece under cooling water spray. Canal patency was checked with K- file size # 10. Shaping of the canals was performed in a crown-down technique using ProTaper Universal rotary instruments in an endodontic motor according to the manufacturer's instructions. During instrumentation, irrigation was done with 2.5% sodium hypochlorite solution and a K-file size # 15 was used to remove debris between each file to maintain patency of the canal. After instrumentation, the entire smear layer was removed using 3 ml of sodium hypochlorite solution for 3 min to remove the organic part followed by 17% ethylene-diaminetetraacetic acid for 60 s using saline in between to avoid any chemical reaction between the two solutions. Final flush was performed by 2.5% sodium hypochlorite solution in all teeth.

Obturation of the root canals

Teeth were obturated using the conventional lateral compaction technique with standard 2% master apical gutta-percha point which was selected according to the size of the prepared canal (size 50) and AH plus sealer was used and mixed according to the manufacture instructions. Master cone was tested visually and confirmed by tactile test to feel the frictional fit at the apical third (Tug back action), then confirmed radiographically to match the laboratory situation. Obturation was considered completed when the spreader could no longer penetrate more than 1–2 mm beyond the root canal orifice. All excess sealer and gutta-percha cones were removed with a heated instrument then a small sized amalgam condenser was used to condense gutta-percha in a vertical direction at the orifice. Following obturation, all orifices were sealed with temporary filling material.

Root end resection and cavity preparation

Following obturation, the teeth were sectioned perpendicular to the long axis of the root at 3 mm from the apex using diamond stone mounted on a high-speed handpiece under copious amount of water coolant. Root-end cavity preparation was then done to prepare a Class I cavity design with a 3 mm depth and 1 mm in diameter (confirmed by periodontal probe) at the apical end of all roots using Satalec ultrasonic tip AS3D driven by a piezoelectric ultrasonic unit at a low power setting. Cutting was done in a back-and-forth motion with cutting tip enveloped in water spray. Post-operative radiograph was taken after retro-preparation of all roots to confirm that the cavity was confined inside the root canal (Figure 1).

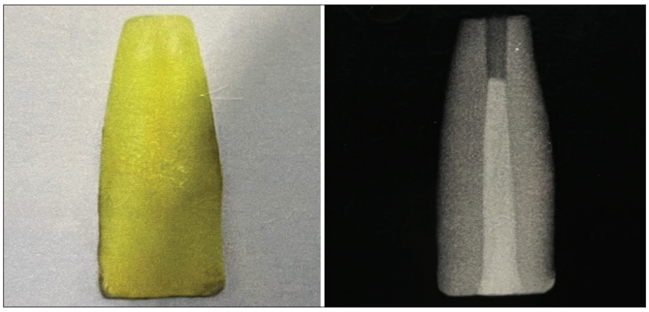

Figure 1: A photograph showing a tooth with root-end resection; and a radiograph showing the tooth after ultrasonic root-end preparation

Samples grouping

After root-end cavity preparation, the 40 specimens were randomly divided into two equal experimental groups according to the material to be used: Group I: Nano MTA (n = 20 teeth) and Group II: MTA (n= 20 teeth). And then each group was subdivided into two equal groups according to method of evaluation: Subgroup A: Sealing ability (Figure 2a and b): Ten samples from each experimental group were selected for examination under Scanning Electron Microscope (longitudinal section) after being stored for 48 h in saline. Subgroup B: Adaptability (Figure 2c and d): Ten samples from each experimental group were selected for examination under scanning electron microscope (Transverse section) after being stored for 48 h in saline.

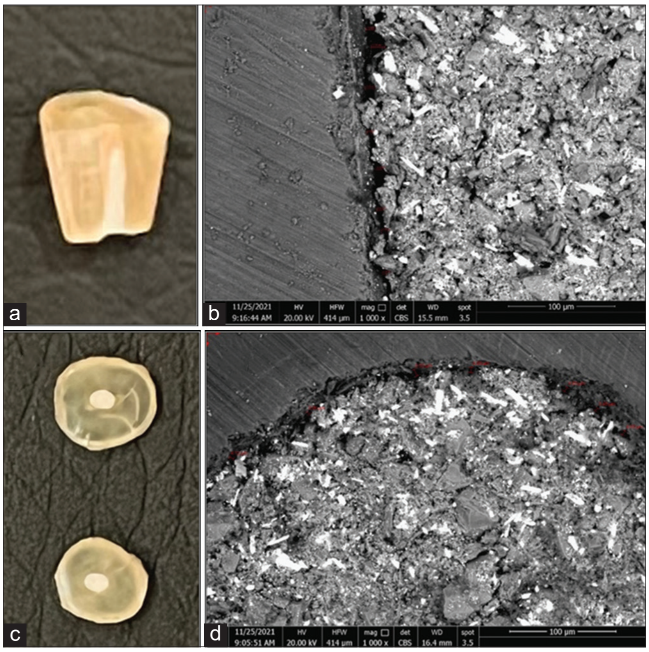

Figure 2: (a) A longitudinal section in the apical 3 mm into two halves using a slow-speed diamond saw; (b) scanning electron microscopic evaluation of sealing ability with 1000x magnification in a longitudinal section; (c) a transverse section to obtain 1 mm thick section using a slow-speed diamond saw; (d) scanning electron microscopic evaluation of adaptability with 1000× magnification in a transverse section

Retrograde filling

After drying the retrograde cavities by paper points, each cavity was filled with one of the test retrograde filling materials according to its group. Both materials mixture was done by mixing powder on a clean and dry glass slap gradually with sterile distilled water to reach thick creamy consistency using a powder to liquid ratio of 3:1 for MTA [7] and 1:4 for nano-MTA [8]. The mix was carried into the retro-cavities in one increment using a clean sterilized MTA carrier (MAP system, Himat company MTA applicator) and condensed by a suitable sized endodontic plugger. Finally, the excess material was removed using large sized disposable bond brush.

Storage of the samples

Root-end fillings were allowed to set completely in moist gauze for 4 h [9] before final immersion in saline for storage. Setting of the filling materials was verified by exerting light pressure on the surface of filling with periodontal probe. All the samples were then stored suspended in sterile closely fitted labeled glass tubes containing normal physiologic saline solution and incubated at 37°C degrees temperature to simulate the clinical situation until time of study.

Methods of evaluation

Sealing ability

Using Scanning Electron Microscope (Quanta FEG 250 Scanning Electron Microscope "FEI Company, Hillsboro, Oregon-USA"), samples were mounted onto SEM stubs. Applied SEM conditions were a 10.1 mm working distance, with an in-lens detector with an excitation voltage of 20kV. The samples were viewed under magnification (×1000) and the gap thickness between the root end filling material and the retro cavity dentine walls was measured at seven selected points at the material-dentine interface. The photomicrographs at ×1000 magnification were analyzed on the software Image Tool 3.0 and the extent of gap was measured linearly, in micrometers (μm). The average between the seven points was calculated, and the mean and SD of sample gaps were calculated.

Adaptability

The measurements were done with same technique as the longitudinal section by measuring the distance between the root end filling material and the retro cavity dentine walls at seven selected points at the material-dentine interface in the transverse section.

Statistical analysis

The mean and standard deviation values were calculated for each group in each test. Data were explored for normality using Kolmogorov–Smirnov and Shapiro–Wilk tests, data showed parametric (normal) distribution. Paired sample t-test was used to compare between two groups in related samples. Independent sample t-test was used to compare between two groups in non-related samples. Two-way ANOVA was used to test the interaction between variables. The significance level was set at $p \leq 0.05$. Statistical analysis was performed with IBM® SPSS® Statistics Version 20 for Windows.

Results

Sealing ability

Scanning electron microscopic observations of sectioned samples retro-sealed with nano-MTA root-end filling showed homogenous filling material. Nano-MTA had an overall good marginal adaptation with the retro-cavity margins. At higher magnifications, the surface of nano-MTA appeared homogenous. Almost no voids or cracks could be detected within the material bulk (Figure 3a and b).

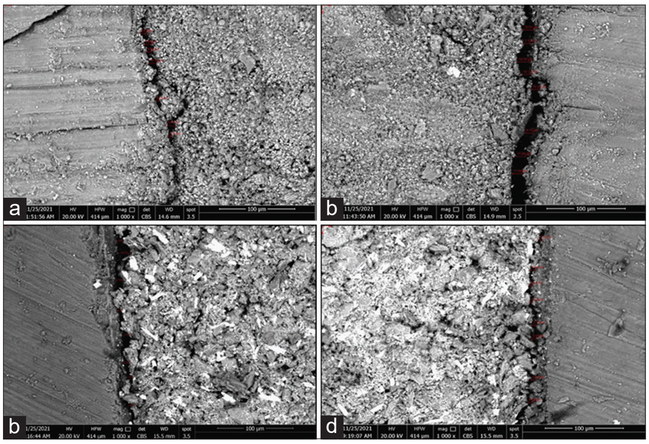

Figure 3: (a) SEM micrograph for longitudinal section of retro-cavity sealed with nano-MTA showing good adaptation. Homogenous filling mass is clearly detectable (×1000). (b) SEM micrograph for longitudinal section of retro-cavity sealed with nano-MTA showing homogenous material with almost no cracks or voids (×1000). (c) SEM micrograph for longitudinal section of retro-cavity sealed with MTA. The interface between MTA and retro-cavity margins appeared to be relatively good but with thin areas of gaps (×1000). (d) SEM micrograph for longitudinal section of retro-cavity sealed with MTA showing few cracks and voids within the bulk of the material (×1000)

Nano-MTA had good homogenous interfacial adaptation to the dentinal wall. Close adaptation of the material to the dentinal wall was evident with minimal gap thickness. No evidence of penetration of the nano-MTA root-end filling material into the dentinal tubules. The recorded mean gap thickness between dentin-material interfaces was 3.154 ± 0.710 μm with minimum value of 2.16 μm and maximum value of 3.86 μm.

While scanning electron microscopic observations of sectioned samples retro-sealed with mineral trioxide aggregate root-end filling showed homogenous filling material with an overall acceptable degree of marginal adaptation at the interface. At higher magnifications, the surface of the tested material seemed to be irregular in structure. The presence of few voids and cracks was clearly at certain areas within the bulk of the material (Figure 3c and d).

MTA material particles were closely adapted to the dentinal walls with intimate contact at the interface in most areas. Minimal gap thickness was observed in some areas at the interface. No evidence of penetration of the MTA root-end filling material into the dentinal tubules was noted. The recorded mean gap thickness between dentin-material interfaces was (3.274 ± 0.770 μm) with minimum value of (2.40 μm) and maximum value of (4.89 μm). However, there was no statistically significant difference between (nano-MTA) and (MTA) where (p = 0.722) as presented in Table 2.

Table 2: The recorded means, standard deviation (SD) values of the gap thickness between dentin-material interface measurements in (μm) in the longitudinal section of nano-MTA and MTA groups

Variables	Sealing			
	Mean	SD	Min	Max
Nano-MTA	3.154	0.710	2.16	3.86
MTA	3.274	0.770	2.40	4.89
p-value	0.722ns			

SD: Standard deviation, p: Probability level, ns: non-significant (p > 0.05).

Adaptability

The recorded mean gap thickness between dentin-material interfaces in nano-MTA group was 2.205 ± 0.712 μm with minimum value of 1.21 μm and maximum value of 2.98 μm (Figure 4a) while the MTA group showed a mean gap thickness between dentin-material interfaces was 2.460 ± 0.599 μm with minimum value of 1.08 μm and maximum value of 3.24 μm (Figure 4b). However, there was no statistically significant difference between nano-MTA and MTA where p = 0.397 as presented in Table 3.

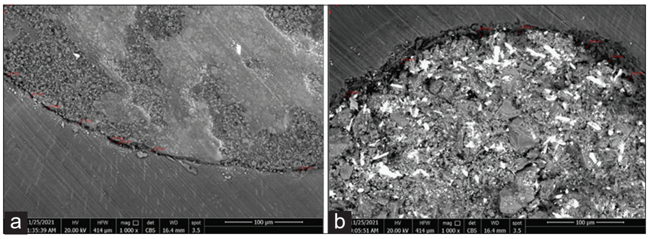

Figure 4: (a) SEM micrograph for transverse section of retro-cavity sealed with nano-MTA showing good adaptation. Homogenous filling mass with no voids or cracks is clearly detectable (×1000). (b) SEM micrograph for transverse section of retro-cavity sealed with MTA. The interface between MTA and retro-cavity margins appeared to be relatively good but with thin areas of gaps. Figure showing few cracks and voids within the bulk of the material (×1000)

Table 3: The recorded means, standard deviation (SD) values of the gap thickness between dentin-material interface measurements (μm) in the transverse section of nano-MTA and MTA groups

Variables	Adaptability			
	Mean	SD	Min	Max
Nano-MTA	2.205	0.712	1.21	2.98
MTA	2.460	0.599	1.08	3.24
p-value	0.397ns			

SD: Standard deviation; p: Probability level. ns: non-significant (p > 0.05).

Discussion

Mineral trioxide aggregate (MTA) represents the gold standard material to which other repair materials are compared. It has excellent properties that support its clinical use such as its biocompatibility, antimicrobial activity, and its ability to promote regeneration of the original tissues when placed in contact with dental pulp or peri-radicular tissues. Several studies have demonstrated the fact that MTA leaks significantly less than other root-end filling materials. Fischer *et al*. [10] proved that Mineral Trioxide Aggregate was the most effective root-end filling material against penetration of S. marcescens compared to other materials including zinc-free amalgam, Intermediate Restorative Material (IRM), Super-EBA. In their study, MTA required longer time to leak compared to all the other material. In addition, four of the MTA samples had not exhibited any leakage.

Although MTA has superior biocompatibility and sealing ability when compared to the traditional materials used in root-end filling and root repair, it has a long setting time and poor handling properties. Recently, experimental nano Mineral Trioxide Aggregate (nano-MTA) was prepared by Nano Gate Company in Egypt. Nano-MTA is a newly introduced root-end filling cement. The manufacturers claim that it has similar composition to MTA, but with the reduction of its particle size to obtain a high specific powder surface area that may lead to a better and faster hydration process. It is understood that in mixing and working phase, more particles are involved in the reaction in the hydration phase resulting in the formation of a less porous set material. Komabayashi and Spangberg [11] showed that MTA's particle size has a great impact on the extent of particles penetrating the dentinal tubules. This study was carried out to compare the newly introduced nano material nano-MTA with the gold standard material MTA. This comparison was carried out by evaluating the sealing ability and adaptability of the two materials.

Freshly extracted human teeth were used to improve the reliability of the investigation by simulating the clinical situation. Single rooted teeth with broad straight roots and single root canal were used for standardization and elimination of variables from multiple canals and complex curved canal morphology.

During instrumentation, irrigation was performed using 2.5% NaOCl which allowed the irrigating solution to perform its effect as an antibacterial agent and organic tissue solvent while limiting its cytotoxic effect due to the relation between cytotoxicity and concentration [12], [13]. The application time was 1-min as no significant statistical difference was found between 1, 3, and 5 min application of NaOCl [14]. Smear layer was removed using 3 ml of 2.5% NaOCl for 3 min followed by 3 ml of 17 % EDTA solution for 1 min [15], [16]. This was approved through the work of Taylor *et al*. [17] who showed that removal of the smear layer decreases leakage, regardless of the obturation technique used. Gettleman *et al*. [18] and Shahravan *et al*. [19] suggested its removal as it contains bacteria and necrotic tissues, it may limit the optimum penetration of disinfecting agents and medicaments into the dentinal tubules, and it obstructs the extension of sealer tags into the dentinal tubules and thereby decreases adhesion by micromechanical forces. Irrigation with distilled water was used as a final flush to ensure removal of all NaOCl and EDTA remnants in accordance with Zmener *et al*. [20]. It is known that NaOCl breaks down to sodium chloride and oxygen. Oxygen causes strong inhibition of the interfacial polymerization of resin bonding materials. The generation of oxygen bubbles at the resin dentin interface may also interfere with resin infiltration into the dentinal tubules dentin causing detrimental effects on the dentin bonding performance of dentin adhesive systems [21], [22].

Lateral condensation technique with gutta-percha in conjuction with an insoluble root canal sealer was used for obturation of the canals due to its predictability, relative ease of use, conservative preparation, and controlled placement of the obturating materials [23], [24]. The apical root-end resection was done perpendicular to the long axis of the root with no bevel that allows more conservation of root structure, improved the crown root ratio, removes the majority of apical ramification, and reduced the number of exposed open cut dentinal tubules. On the other hand, long bevels would require the removal of an excessive amount of root structure. Another consideration for the 0° bevel is that the cavo-surface marginal dimensions of the preparation will be considerably decreased; therefore, allowing an easier and more predictable chance for marginal seal and reduce the risk of lingual perforation [25]. Kim and Kratchman [26] suggested that at least 3 mm of the root-end must be removed in root-end resection because 98% of the apical ramifications and 93% of the lateral canals exist in 3 mm of the root-end.

Retro-cavities were performed by a specially designed diamond coated ultrasonic retro-tip mounted on an ultrasonic unit. Using coolant was important to avoid overheating and to decrease number of cracks formed in dentine. The use of contra-angle retro-tip facilitated retro preparation into the canal, aid in access, and provided superior control. Diamond coated retro-tip was used as it is faster than other types of ultrasonic tips (stainless steel and zirconium coated retro-tips) to prepare root-end cavities [27]. The ultrasonic unit used was adjusted at low intensity since this produced less cracks [28]. Gorman *et al*. [29] showed that root-end cavities prepared with ultrasonic showed the presence of significantly less smear layer and debris compared with those prepared by conventional technique.

All specimens, in whom sealing ability and adaptability were evaluated, were stored with the external surfaces submerged in phosphate buffered saline to provide a buffer system to maintain the physiological pH (7–7.6) that simulates the clinical situation. Parirokh *et al*. [30]suggested that *in vitro* studies evaluating the sealing ability of MTA should use phosphate buffered saline as a media to simulate the environment in the human body, thus producing more clinically relevant results.

In the present study, scanning electron microscope (SEM), a method of histological evaluation, has been used to assess the retro-cavity seal quality. SEM has greater resolution, improved interface magnification, superior field depth, and therefore better gap visualization [31], but it has several shortcomings. It is a surface phenomenon and may not represent the seal and the adaptation of two surfaces in three dimensions. That is why in this study we decided to evaluate both sealing ability and adaptability by examination of a longitudinal and a transverse section of the specimens respectively under the SEM to obtain a full three-dimensional representation for the material-dentine interface [32]. This was done by measuring more than one point (7–8 different points) between the material-dentine interface for more clear observation of the gap to permit more accurate and reliable results, and to avoid the limited evaluation in other studies [33].

The results of this study showed that nano-MTA has better sealing ability than MTA but with no statistically significant difference between them, and this was in accordance with Dewi *et al*. [34] who stated that there was less microleakage score detected in modified MTA (MTA Flow) when compared to conventional MTA with no statistically significant difference. The better sealing ability may be largely due to the particle size of the modified MTA, which was homogenous and smaller than 10 μm while the insignificant difference can be justified by the similarity in the chemical composition between the two materials. Furthermore, Abd Elhamid and Abdel-Aziz [32] found insignificant statistical difference between the nano-MTA and nanohydroxyapatite materials in sealing ability, which can be attributed to the small particles size of both materials.

Studies proved that smaller particle sizes can minimize spacing between particles, increase surface area, and better interlock powder particles to improve integrity, making the material more resistant to liquid penetration [35]. Good marginal sealing ability of MTA may be due to its hydrophilic properties, and formation of an interfacial layer between the material and dentin [36]. It was found that the further hydration of MTA powder by moisture can result in an increase in the compressive strength and decrease leakage [37].

However, Arroyave *et al*. [38] found that MTA had significantly more leakage than IRM and super ethoxy benzoic acid (EBA). The cause of this conflict may be due to the difference in methodology since that study used the dye penetration technique for evaluation. Our study was also not in concurrence with Hirschberg *et al*. who showed that samples in the EndoSequence bioceramic root repair material group leaked significantly more than samples in the MTA group [39].

According to the present study, there was no statistically significant difference in marginal adaptation between nano-MTA and MTA although nano-MTA group showed better adaptation. The good adaptation of both materials to cavity margins might be linked to the nature of these materials. The powder consists of fine hydrophilic particles that absorb water during hydration. Therefore, the material expands during solidification, which must have played a role in its superior adaptation to dentinal walls. Moreover, studies illustrated that smaller particle sizes and a more fluid consistency can better fill the irregularities of a dentinal wall to minimize gaps and that this can be done even faster with hydroxyapatite formation [7].

The marginal adaptation results were comparable with the sealing ability results, as the better the sealing ability the better the adaptation. However, Xavier *et al*. [40] evaluated the sealing ability and adaptation of retro-filling materials and found no correlation between marginal gaps and degree of leakage, but this may be due to using dye penetration technique as an evaluation method for the sealing ability which proved then to be an unreliable method.

This study was done to evaluate the sealing ability and adaptability of the newly introduced nano-MTA and the gold standard MTA. However, it is still open for further clinical trials to identify its clinical use and determine its healing ability.

Conclusion

Our results revealed that nano-MTA has similar sealing ability and adaptability to MTA as a root-end filling material when it was evaluated under scanning electron microscope.

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