

Zinc oxide nano-particles as sealer in endodontics and its sealing ability

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Abstract

Aims: The aim of this study was to evaluate the sealing ability of new experimental nano-ZOE-based sealer. **Settings and Design:** Three types of nano-ZOE-based sealer (calcined at different temperatures of 500, 600 and 700°C) with two other commercially available sealers (AH26 and micro-sized zinc oxide eugenol sealer) were used. **Materials and Methods:** Zinc oxide nano-particles were synthesized by a modified sol-gel method. The structure and morphology of the prepared powders were characterized using x-ray diffraction (XRD) and transmission electron microscopy (TEM) techniques. The instrumented canals of 60 single-rooted teeth were divided into five groups ($n = 10$), with the remaining ten used as controls. The canals were filled with gutta-percha using one of the materials mentioned above as sealer. After 3, 45 and 90 days, the samples were connected to a fluid filtration system. **Statistical Analysis Used:** The data were analyzed using Student's *t*-test. **Results:** The XRD patterns and TEM images revealed that all the synthesized powders had hexagonal wurtzite structures with an average particle size of about 30-60 nm at different calcination temperatures. Microleakage in AH26 groups was significantly more than that in three groups of ZnO nano-particles at all the three evaluation intervals. Apical microleakage of ZnO micro-powders was significantly more than that of all the materials, but the sealing ability of ZnO nano-powder sealers did not differ significantly. **Conclusion:** The results of this study showed that the synthesized ZnO nano-powder sealers are suitable for use as a nano-sealer in root canal therapy to prevent leakage; however, further studies should be carried out to verify their safety.

Keywords: AH26, dental material, microleakage, nano-particle sealers, sealer, zinc oxide eugenol

Introduction

Sealers with nano-particles or commercially available sealers; which one can seal the root canal better?

Obturation of the root canal system is the final step in root canal treatment. Therefore, a permanent three-dimensional seal is developed from the apical foramen to the root canal orifice. Incomplete sealing and presence of spaces between the root canal wall and the obturating material can lead to failure in apparently good treatment.

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For over a century gutta-percha (GP) has been the most commonly used material for obturation of the root canal system. Although not the ideal, it fulfills many of the characteristics that Grossman reported.^[1] One of the disadvantages of GP, as a root canal obturation material, is its poor sealing ability; therefore, it must be used with a root canal sealer to provide an effective seal.^[2] The most commonly used sealers in root canal treatment are ZOE-based sealers, which have been modified for endodontic purposes. The powder of these sealers contains zinc oxide (ZnO), which combines with a liquid, generally eugenol.^[3] The valuable component of these sealers is ZnO, an II-VI semiconductor compound which is stably crystallized in a hexagonal wurtzite structure.^[4]

Nanotechnology is the science of evaluating and producing materials in nano-dimensions by re-location and re-arrangement of atoms to prepare materials with better properties. Presence of very small particles leads to superior properties of the material. These unique properties, which are the subject of quantum mechanics, have attracted a great deal of interest.

Nano-technology and nano-materials have become an extremely active field of research in the last decade because of their potential application in different areas like medicine, information technologies, energy storage etc., Nano-technologies have been used in the production of a wide range of dental materials: Light polymerization composite resins and bonding systems, imprint materials, ceramics, coatings for dental implants, bioceramics,^[5] mouthwashes containing fluoride^[6] and fissure sealant materials.^[7]

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Recently, the first author of this article prepared a new experimental endodontic sealer (nano-powder ZnO) in the Dental Material Research Center, Mashhad University of Medical Sciences, Iran. This sealer is similar to various ZOE-based sealers, but with different sizes of nano-particles of zinc oxide.

The morphology, size and crystallinity of the prepared nano-particles were first characterized. The sealing ability of our synthesized nano-sized zinc oxide eugenol sealer was compared with epoxy resin sealer (AH26) and micro-sized zinc oxide eugenol sealer.

Materials and Methods

Synthesis of ZnO nano-particles

In this work ZnO nano-powders were prepared by a modified sol-gel method, using gelatin.^[8] To prepare 5 g of the final product, first a solution of gelatin (type B from bovine skin, Sigma Aldrich) was prepared by dissolving 10 g of gelatin in 150 mL of deionized water at 60°C. Then, appropriate amounts of zinc nitrate ($Zn(NO_3)_2 \cdot 6H_2O$, Merck %99) were dissolved in minimum deionized water at room temperature. Then, the two solutions were mixed and stirred for 8 hours while the temperature was kept at 80°C. Finally, the pure resins were calcined at different temperatures of 500, 600 and 700°C to obtain ZnO nano-powders. Morphological and structural properties of the prepared ZnO were characterized by X-ray diffraction (XRD) and transmission electron microscopy (TEM) techniques.

Sample preparation for measuring the microleakage

In this study, 60 single-rooted anterior teeth were selected. The roots were cross-sectioned at the cemento-enamel junction with a carborundum disk (Brassler USA, Savannah, GA), except for five roots as the negative controls [Table 1]. Working length was determined by a #10 K-file visible at the apex. Instrumentation of all the teeth was performed by a step-back technique using stainless steel K-files (Dentsply Maillefer, Ballaigues, Switzerland) to ISO #35. Irrigation was performed using 1 mL of 5.25% NaOCl between each file. The smear layer was removed with 1 mL of 17% EDTA (Ariadent, Asia ChemiTeB, Tehran, Iran) for 1 min, followed by 3 mL of 5.25% NaOCl. The canals were finally flushed with 5 mL of normal saline. On completion of instrumentation, the specimens were randomly divided into five groups consisting of ten teeth in each group with the remaining five used as positive controls. The root canals were completely dried with paper points before obturation. The root canals in the first group were obturated with gutta-percha using AH26 (Dentsply, DeTrey, Konstanz, Germany) as sealer with the lateral condensation technique. The root canals in groups II to IV were obturated with the prepared ZnO nano-powders (three types: Calcined at different temperatures of 500, 600 and 700°C) and the root canals in the last group were filled with ZOE sealer (zinc oxide eugenol micro-powder). To allow the material to set, all the roots were stored at 100% humidity and 37°C for the next 72

hours in an incubator. The canals in the positive control group were not filled. After this period, the external root surfaces of the specimens in the experimental and the positive control groups were completely covered by two coats of nail varnish and Parafilm tapes (Parafilm "M", Laboratory Film, Chicago, USA) for double sealing, except for a 2-mm area around the root apex. The root surfaces of the specimens in the negative control group were completely covered [Table 1]. Then, each tooth was placed in a device for measuring its microleakage using fluid transport process, designed by Javidi *et al.*^[9] Four measurements were recorded for each tooth at 2-minute intervals over a period of 8 minutes. The amount of leakage was expressed as $\mu L/min/cm H_2O$.

Two other evaluations were performed 45 days and three months later to assess longitudinal sealing properties.

Kolmogorov-Smirnov test was used in order to verify normal distribution of parameters; thereafter, the results were analyzed by Student's *t*-test. The significance level was set at 5% for all the tests.

Results

Characterization

Figure 1 shows the XRD patterns of the ZnO nano-powders prepared at three different calcination temperatures of 500, 600 and 700°C. The obtained pattern revealed that the indexed peaks were matched with that of bulk hexagonal well-crystalline ZnO, which confirms that the synthesized nano-powders were well-crystalline ZnO.

TEM (transmission electron microscopy) images and the corresponding particle size histograms of ZnO nano-powders calcined at 500, 600 and 700°C are shown in Figure 2. The sizes of nano-particles increased with an increase in temperature. Nano-particles were spherical and hexagonal at 500 and 600°C, but at 700°C they were almost hexagonal.

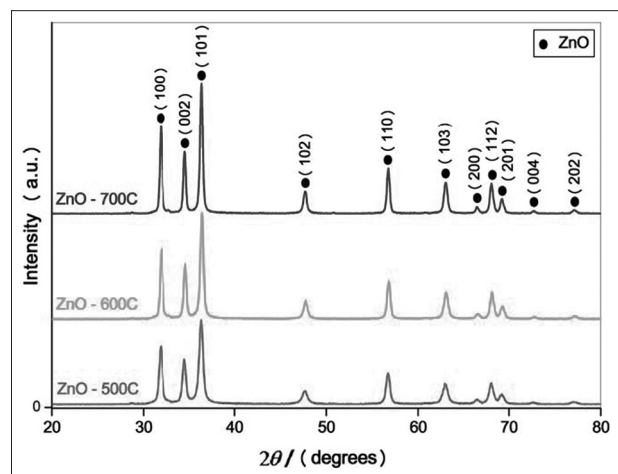


Figure 1: XRD (X-ray diffraction) patterns of ZnO nano-particles prepared at three different calcination temperatures

The average sizes of nano-particles at 500, 600 and 700°C were 29, 36 and 63 nm, respectively [Figure 3].

Microleakage

The mean microleakage in terms of $\mu\text{L}/\text{min}/\text{cm H}_2\text{O}$ is presented in Table 2.

The positive controls leaked significantly more than the experimental groups, whereas the negative controls showed no microleakage.

In all the three evaluation periods, ZnO micro-powders had the highest leakage, which was significant; also the leakage of AH26 was significantly higher than that in all the three groups of ZnO nano-particles.

In nano-ZnO groups, the nano-particles calcined at 500°C had the least amount of leakage but their differences were not significant.

Discussion

The principal aim of using nano-technologies in dental materials is to achieve higher mechanical properties, higher abrasion resistance and less shrinkage of dental composite

resins, and improved optical and esthetic properties of composite resins and ceramics.^[5] This study used ZnO nano-particles as sealer and evaluated its sealing ability in comparison with two common sealers.

XRD patterns presented in Figure 1 revealed that all the prepared samples had hexagonal wurtzite structure.

Several methods have been used to measure leakage around filling materials, including bacterial, dye and saliva penetration, radioisotopes, light microscopic methods and SEM. These methods provide qualitative, rather than quantitative information. They can show the presence or absence of leakage but not the amount.^[10] The use of fluid filtration systems for measuring leakage has been recommended to enhance reliability, reproducibility and comparability.^[10] In several studies, the change of leakage values with time have shown that longitudinal leakage studies are important in determining leakage values of materials.^[11]

ZnO is an environment-friendly material which has been used widely in medical applications such as cancer treatment^[12,13] and DNA detection.^[14] In addition, ZnO has interesting antibacterial properties.^[15-17] Because of interesting antibacterial properties of ZnO, its powder can be used for dental applications as a sealer.^[18-20] In addition, ZOE-based cements have been found to possess favorable characteristics in terms of biocompatibility. These were the reasons for selecting ZnO as the base of a nano-sealer in the present study.

To the best of our knowledge, the use of nano-structured materials as sealers in root canal therapy is limited to two or three types of nano-structured hydroxyapatite alone or in combination with epoxy resin (Nanoseal).^[7,21,22] Properties like antimicrobial activity, radiopacity, flow, film thickness and cytotoxicity have been evaluated in various studies. We could not find any published reports on sealing ability of nano-materials as sealer in root canal therapy to make comparisons.

Physical properties, biocompatibility, sealing ability, ease of handling etc., are necessary to characterize a new sealer. Sealing ability of sealers is a criterion for their selection.

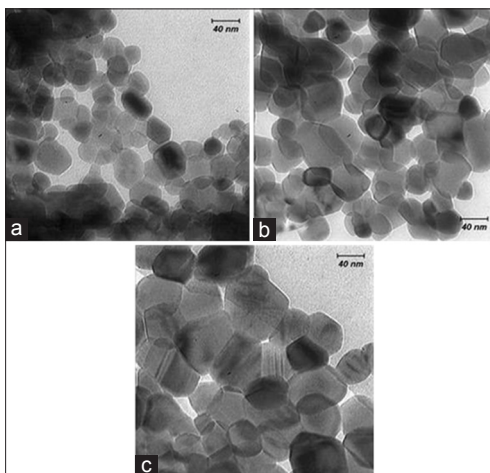


Figure 2: TEM images of ZnO nano-particles calcined at 500, 600 and 700°C (a, b and c respectively)

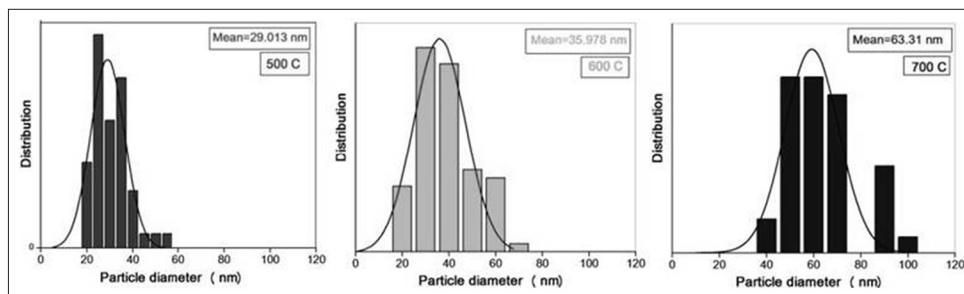


Figure 3: Histograms of ZnO nano-particles calcined at 500, 600 and 700°C. The mean sizes of ZnO nano-particles are presented in a, b and c

Table 1: Description of the groups

Groups	Method of preparation		Sealer
G1	Cross-sectioning at the CEJ	Instrumentation to ISO #35	External root coverage except for 2-mm at the apex AH26
G2	Cross-sectioning at the CEJ	Instrumentation to ISO #35	External root coverage except for 2-mm at the apex ZnO nano-powders (calcined at 500°C)
G3	Cross-sectioning at the CEJ	Instrumentation to ISO #35	External root coverage except for 2-mm at the apex ZnO nano-powders (calcined at 600°C)
G4	Cross-sectioning at the CEJ	Instrumentation to ISO #35	External root coverage except for 2-mm at the apex ZnO nano-powders (calcined at 700°C)
G5	Cross-sectioning at the CEJ	Instrumentation to ISO #35	External root coverage except for 2-mm at the apex ZnO micro-powders
C+	Cross-sectioning at the CEJ	Instrumentation to ISO #35	External root coverage except for 2-mm at the apex No obturation
C-	Intact teeth	No instrumentation	Complete coverage of the root surfaces No obturation

CEJ: Cemento-Enamel Junction

Table 2: Mean and SD ($\times 10^{-7}$) of apical microleakage of five experimental groups as $\mu\text{l. min}^{-1} \cdot \text{cm H}_2\text{O}^{-1}$

Groups	3 days after obturation	45 days after obturation	90 days after obturation
G1	7.75±6.17	7.65±6.00	7.52±6.03
G2	0.72±3.82	0.72±3.82	0.31±3.50
G3	1.17±0.99	1.42±1.36	1.69±1.68
G4	2.52±4.25	2.40±4.05	2.39±4.05
G5	80.29±108.64	119.68±142.88	162.44±207.64

Laboratory evaluation of the sealing ability of new endodontic sealer products is necessary prior to their clinical use.

The microleakage measurements of the synthesized nano-powders and also the conventional ZOE and AH26 sealers were performed at 3-, 45-, and 90-day intervals to check the stability of their sealing properties. The results shown in Table 1 indicate that the ZnO nano-powders prepared at different calcination temperatures exhibited less microleakage compared to ZOE and AH26. The minimum microleakage in ZnO samples corresponded to that in nano-powders calcined at 500°C. Therefore, microleakage of ZnO nano-powders increased with an increase in calcination temperature because of an increase in nano-particle size [Figure 2; TEM images], leading to a decrease in effective surface. As a result, one can conclude that the root canal can be sealed better by using smaller nano-powder particle sizes. In addition, all the groups exhibited significant differences in leakage in comparison with commonly used ZOE sealer.

Conclusion

The results of this study showed that synthesized ZnO nano-powders exhibited less microleakage in comparison with AH26 and ZOE, making them suitable for use as a nano-sealer in root canal treatment. Nevertheless, further studies should be carried out and limitations and the potential

unknown risks involved in the use of ZnO nano-powders as a medical material should be considered to verify their safety.

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References

- Grossman L. Root canal therapy. Philadelphia: Lea and Febiger; 1940. p. 189.
- Younis O, Hembree JH. Leakage of different root canal sealants. Oral Surg Oral Med Oral Pathol 1976;41:777-84.
- Metzger Z, Basrani B, Goodid HE: Instruments, Materials, and Devices. In: Hargreaves KM, Cohen S: Pathways of the pulp. St, Louis: CV Mosby; 2011. p. 263-4.
- Nomura K, Ohta H, Ueda K, Kamiya T, Hirano M, Hosono H. Thin-film transistor fabricated in single-crystalline transparent oxide semiconductor. Science 2003;300:1269-72.
- Dikova T, Abadjiev M, Balcheva M. Clinical application of the contemporary nano-materials (part 1 – laboratory composites). Journal of IMAB 2009;9:67-70.
- Sun L, Chow LC. Preparation and properties of nano-sized calcium fluoride for dental applications. Dent Mater 2008;24:111-6.
- Li P, Xiao LY, Li W, Tan H, Liao YM, Yuan ML. Evaluation on cytotoxicity of a new nano-hydroxyapatite as root canal filling sealer. Hua Xi Kou Qiang Yi Xue Za Zhi 2005;23:427-30.
- ZakAK, MajidWH, Darroudi M, Yousefi R. Synthesis and characterization of ZnO nanoparticles prepared in gelatin media. Mater Lett 2011;65:70-3.
- Javidi M, Naghavi N, Roohani E. Assembling of fluid filtration system for quantitative evaluation of microleakage in dental materials. Iran Endod J 2008;3:68-72.
- Ozturk B, Ozer F, Belli S. An *in vitro* comparison of adhesive systems to seal pulp chamber walls. Int Endod J 2004; 37:297-306.
- Moradi S, Naghavi N, Rohani E, Javidi M. Evaluation of microleakage following application of a dentin bonding agent as root canal sealer in the presence or absence of smear layer. J Oral Sci 2009;51:207-13.
- Hanley C, Layne J, Punnoose A, Reddy KM, Coombs I, Coombs A, *et al.* Preferential killing of cancer cells and activated human T cells using ZnO nanoparticles. Nanotechnology 2008;19:295103.

13. Nair S, Sasidharan A, Divya Rani VV, Menon D, Nair S, Manzoor K, *et al.* Role of size scale of ZnO nanoparticles and microparticles on toxicity toward bacteria and osteoblast cancer cells. *J MaterSci Mater Med* 2009; 20(Suppl 1):S235-41.
14. Kumar N, Dorfman A, Hahm J. Ultrasensitive DNA sequence detection using nanoscale ZnO sensor arrays. *Nanotechnology* 2006; 17:2875.
15. Applerot G, Lipovsky A, Dror R, Perkas N, Nitzan Y, Lubart R, *et al.* Enhanced antibacterial activity of nanocrystalline ZnO due to increased ROS-Mediated cell injury. *Adv Funct Mater* 2009;19:842-52.
16. Li Q, Chen SL, Jiang WC. Durability of nanoZnO antibacterial cotton fabric to sweat. *J Appl Polym Sci* 2007;103:412-6.
17. Zhang L, Ding Y, Povey M, York D. ZnO nano fluids-A potential antibacterial agent. *Prog Nat Sci* 2008;18:939-44.
18. Camps J, Pommel L, Bukiet F, About I. Influence of the powder/ liquid ratio on the properties of zinc oxide-eugenol-based root canal sealers. *Dent Mater* 2004;20:915-23.
19. Takatsuka T, Tanaka K, Iijima Y. Inhibition of dentine demineralization by zinc oxide: *In vitro* and *in situ* studies. *Dent Mater* 2005; 21:1170-7.
20. Wong RH, Palamara JE, Wilson PR, Reynolds EC, Burrow MF. Effect of CPP-ACP addition on physical properties of zinc oxide non-eugenol temporary cements. *Dent Mater* 2011;27:329-38.
21. Aal-Saraj AB, Ariffin Z, Masudi SM. An agar diffusion study comparing the antimicrobial activity of Nanoseal with some other endodontic sealers. *Aust Endod J* 2012;38:60-3.
22. Collares FM, Leitune VC, Rostirolla FV, Trommer RM, Bergmann CP, Samuel SM. Nanostructured hydroxyapatite as filler for methacrylate-based root canal sealers. *Int Endod J* 2012; 45:63-7.

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