

# In vitro Comparative Evaluation of the Fracture Resistance of Simulated Immature Teeth reinforced with Different Apical Barriers and Obturation Combination

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## ABSTRACT

**Aim:** This study aimed to assess and compare the fracture resistance of simulated immature teeth reinforced with Biodentine (BD) and mineral trioxide aggregate (MTA) as apical barriers and two root canal backfilling combination (gutta-percha/AH26, MetaSeal).

**Materials and methods:** A total of 70 extracted human maxillary incisors were randomly divided into seven groups (n=10). The positive control group was not instrumented. For the other groups, coronal access was made and root canals were instrumented using the ProTaper, up to F5 followed by six Peeso reamers which were allowed to pass 1 mm beyond the apex to size 6 (1.7 mm) to simulate immature teeth. The apical 4 mm of their root canals was filled with either MTA or BD apical barrier, then backfilled with gutta-percha/AH26 or MetaSeal obturation combination. The negative control group was left unfilled. Composite resin was used to restore the coronal access cavities. The maximum load for fracture of each tooth was recorded utilizing a universal testing machine. Data were analyzed using two-way analysis of variance.

**Results:** The noninstrumented group I had the highest fracture resistance and differed significantly ( $p < 0.05$ ) from the negative control groups. On the contrary, no significant difference was found between BD and MTA groups, regardless of the backfilling combination ( $p > 0.05$ ).

**Conclusion:** There was no difference between MTA and BD apical barriers and the backfilling combination regarding their resistance to root fracture.

**Keywords:** Apical barrier, Biodentine, Fracture resistance, Immature teeth, MTA.

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## INTRODUCTION

Children between 8 and 12 years old are more prone to traumatic dental injuries which often can lead to pulp necrosis. Consequently, the development of the root stops and the root canal remains large, with thin fragile walls and open root apex. These changes make the root canal instrumentation troublesome and prevent the formation of a hermetic apical seal.<sup>1,2</sup>

Therefore, in order to permit easy condensation of the root canal filling materials and encourage an apical seal, it is essential to create an artificial apical barrier or induce apical closure with calcified tissue (apexification). Subsequently, the goal of the treatment of immature teeth is to produce a barrier to place the root canal filling material against it, thereby preventing the materials' extrusion into the surrounding periapical tissue and providing a restoration that reinforces the thin fragile root walls.<sup>3</sup>

Traditionally, calcium hydroxide paste was used to promote the formation of effective hard tissue apical barrier (apexification)<sup>4</sup> that permits filling of the root canal space with the traditional methods. However, this material has many drawbacks, such as delayed treatment that might require from 5 to 19 months with subsequent multiple visits and the possibility of increased root fracture due to adverse effects on the properties of the dentinal collagen network.<sup>5,6</sup>

Mineral trioxide aggregate (MTA) was used as an alternate treatment modality to calcium hydroxide, with the advantages of antimicrobial action, biocompatibility, good sealing ability, low cytotoxicity, and able to set in the presence of blood and moisture contamination. However, the potential obstacles of MTA are long setting time, poor mechanical properties, and difficult handling characteristics.<sup>7-9</sup>

Recently, various new calcium silicate-based materials have been developed in an attempt to produce alternatives to improve the potential drawbacks of MTA.<sup>10-12</sup> Biodentine (BD) is one such material that hardens within 9 to 12 minutes and has good handling properties. Laurent et al<sup>13</sup> suggested that BD can be used as a restorative material in addition to other endodontic indications. Its composition is tricalcium silicate, dicalcium silicate, calcium carbonate, zirconium oxide, calcium oxide, and

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iron oxide in powder form. The liquid is calcium chloride, hydrosoluble polymer, and water.

During apexification procedures, an emerging inquiry should be answered, which material to select for filling the remaining canal space after applying an apical plug? There were literatures that assess the efficacy of various filling methodologies, including fiber post, gutta-percha, and composite resin on the reinforcement capacity of immature teeth after an apical MTA plug has been placed to induce apexification.<sup>14</sup> However, a consensus on this matter has not been reached.

To increase the fracture resistance of immature roots, diverse materials have been investigated in a variety of research. The combination of apical barrier materials and root canal fillings has a role in this reinforcement. Regarding this issue, there were few data on the ability of BD to reinforce the root by apexification with combination of root canal filling materials; therefore, the purpose of this *in vitro* study was to evaluate and compare the fracture resistance of teeth with immature apices treated with apical BD and MTA placement along with two root canal obturation combination.

## MATERIALS AND METHODS

A total of 70 freshly extracted human maxillary central incisors that were extracted due to periodontal reasons were used in the current study. The selection of teeth was based on confirmation of the preoperative radiographs of the absence of previous root canal treatment, cracks, resorptions, or calcifications. Moreover, dimensions of each tooth at the cemento-enamel junction (CEJ) were measured using digital calipers (Mitutoyo Co., Tokyo, Japan):  $5.63 \pm 0.5$  mm faciolingually and  $6.37 \pm 0.4$  mm mesiodistally. For standardization, each tooth specimen's length was adjusted to be 12 mm measured from the apex to the CEJ facially using a diamond disk (Isomet 1000, Beuhler Ltd., Lake Bluff, IL, USA).<sup>15,16</sup> Approval to use human teeth was granted by the research ethics committee at the Faculty of Dentistry, Tanta University, Egypt.

Ten teeth were not instrumented and served as the positive control group (group I). The 60 remaining teeth were prepared as follows: Coronal access was made using a size 3 round bur (Dentsply Maillefer, Tulsa, OK, USA) and an Endo Z bur (Dentsply Maillefer). The root canals were prepared using ProTaper rotary instruments (Dentsply Maillefer, Ballaigues, Switzerland) up to F5 (#50/0.05). The canals were instrumented with Peeso reamers (size 1–6) (Mani Inc., Tochigi, Japan) until size 6 (1.7 mm) could be passed 1 mm beyond the apex to simulate immature teeth.<sup>15</sup> The root canals were irrigated using 3 mL 2.5% sodium hypochlorite (NaOCl) after each instrument, and a final flush with 5 mL 17% ethylenediaminetetraacetic acid (EDTA) was carried out

to remove the smear layer. Finally, the root canals were rinsed with distilled water and dried using paper points (Dentsply Maillefer).

To simulate clinical situations, calcium hydroxide paste (UltraCal XS; Ultradent, South Jordan, USA) was placed in the canals using a 29-gauge NaviTip (Ultradent). The root canal accesses were then sealed using a cotton pellet and a temporary filling material (Cavit™-G; 3M ESPE, Seefeld, Germany), and the samples were kept for 1 week at 37°C under 100% relative humidity. Then the cotton pellet and the temporary filling materials were removed from the access cavities and calcium hydroxide was removed with size 50 K stainless steel hand file (Mani Inc., Tochigi, Japan) that was introduced 1 mm shorter than the working length and gently manipulated to remove the paste. This removal procedure was accompanied with irrigation by 10 mL of 2.5% NaOCl and a final flush with 5 mL 17% EDTA. Finally, the root canals were rinsed with distilled water and dried using paper points.

Sixty teeth were then randomly divided into six experimental groups (n=10) according to the intraradicular treatment performed as follows:

*Group II:* MTA apical barrier + gutta-percha/AH26.

*Group III:* MTA apical barrier + gutta-percha/MetaSeal.

*Group IV:* BD apical barrier + gutta-percha/AH26.

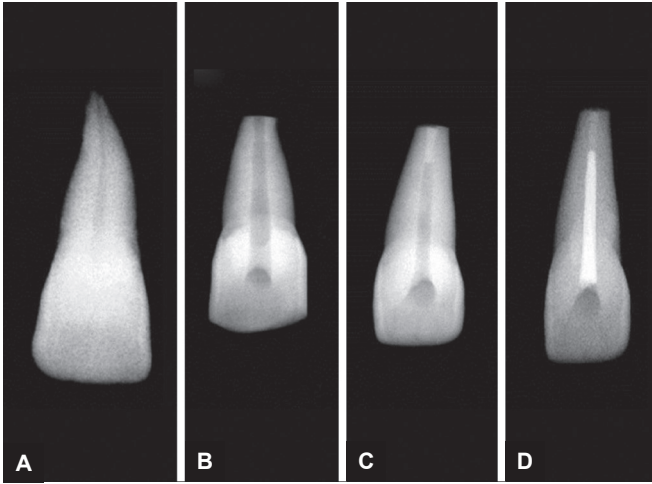
*Group V:* BD apical barrier + gutta-percha/MetaSeal.

*Group VI:* MTA apical barrier without backfilling (MTA negative control).

*Group VII:* BD apical barrier without backfilling (BD negative control).

Mineral trioxide aggregate plus powder (Prevest-Den pro, Jammu City, India; Avalon Biomed Inc., Bradenton, FL, USA) was mixed with distilled water in a proportion of 3:1 according to the manufacturer's instructions. Then MTA mix was placed into the canals using lentulo-spiral (Dentsply Maillefer), introduced 3 mm short of the working length, and condensed apically by gentle packing with hand pluggers (Dentsply Maillefer) to obtain a 4 mm apical plug while the canal at its apical end was closed with finger pressure to prevent material extrusion during barrier placement. A moistened paper point was left in the canal to facilitate the proper setting of the material and access cavities were sealed with cotton pellet and Cavit. After 24 hours, Cavit, cotton pellet, and paper point were removed and a finger plugger was introduced to test proper setting of MTA.<sup>17</sup>

Biodentine (Septodont, Saint-Maur-des-Fosses, France) liquid from a single-dose container was emptied into a powder containing capsule and mixed for 30 seconds in amalgamator (Softly8; de Götzen, Italy) according to the manufacturer's instructions. Biodentine was then placed with a carrier and adapted to the canal walls using a hand plugger to obtain a 4 mm apical plug.



**Figs 1A to D:** Radiographs of specimens groups: (A) Positive control; (B) simulated immature tooth; (C) negative group; and (D) after obturation

The teeth were stored at 37°C and 100% humidity for 1 week and then radiographs were taken to assess the quality of the apical plug.

After apical barrier placement, 20 teeth (10 MTA barrier, 10 BD barrier) were backfilled using gutta-percha/AH26 sealer (Dentsply, De Trey, Konstanz, Germany) according to the manufacturer's instructions using the lateral condensation technique (groups II–IV), while in groups III and V, 20 teeth were backfilled using gutta-percha/MetaSeal sealer (Parkell, Inc., Edgewood, NY, USA) according to the manufacturer's instruction, using cold lateral condensation technique. In groups VI and VII, the teeth were left with either MTA or BD apical barrier without backfilling (negative control groups) (Figs 1A to D).

The coronal access cavities of all teeth were sealed with resin composite (Filtek Z250 XT; 3M ESPE, Seefeld, Germany), and all specimens were stored in 100% humidity at 37°C for 2 weeks until fracture resistance testing.

## FRACTURE RESISTANCE TEST

To achieve clinical imitation of tooth surrounded by the periodontal ligaments, the teeth were embedded in molten wax to 2.0 mm below the CEJ until uniform layer thickness of 0.2 to 0.3 mm wax was coating the roots. Afterward, the wax-covered roots were mounted perpendicularly in plastic holder (20 mm diameter and height), then filled with self-cure acrylic resin (Acrostone, Cairo, Egypt). This will leave a gap of 2 mm between the top of the acrylic and the CEJ facially and lingually to imitate the physiological relationship between the bone crest and the tooth.<sup>14</sup> After the polymerization of the acrylic resin, warm water was used to facilitate removal of the wax from the root surface after its removal from acrylic resin. Thereafter, the resin sockets were filled with light body polyether impression material (Impregum™; 3M

ESPE, Seefeld, Germany) and the teeth were reimmersed into the sockets.

A specially designed jig was constructed to align the specimens at an angle of 45° to the horizontal plane and attached securely to the lower member of a universal testing machine (Model 3345; Instron Industrial Products, Norwood, MA, USA). The load was applied to a specially designed metal rod with round tip (3.8 mm diameter). This rod was attached to the loading cell of the upper member of the testing machine. The teeth were subjected to a gradual and slowly increasing force at a cross-head speed of 1 mm/minute until a fracture occurred.<sup>18</sup> The load at failure was manifested by an audible crack and confirmed by a sharp drop on a load deflection curve recorded using computer software (Instron® Bluehill Lite Software). The maximum force required to fracture each specimen was recorded in Newton (N).

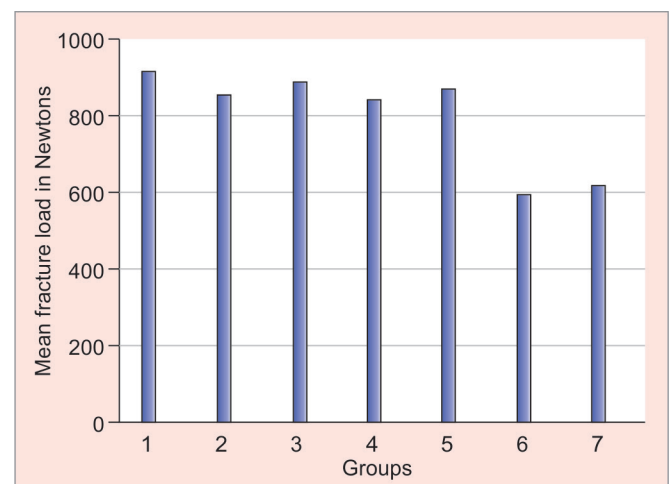
## STATISTICAL ANALYSIS

Statistical analysis was performed using Statistical Package for the Social Sciences (SPSS) 11.0 software for Windows (SPSS Inc., Chicago, IL, USA). Fracture resistance data [Newton (N)] were submitted to two-way analysis of variance (two-way ANOVA). Multiple comparisons were made using Tukey's *post hoc* test; *p* values less than 0.05 were considered to be statistically significant.

## RESULTS

Mean values and their particular standard deviations of the force required to fracture the teeth are summarized in Graph 1 and Table 1. The results of the two-way ANOVA test revealed that a significant difference existed between the groups ( $p < 0.05$ ).

The negative control groups (groups VI–VII) showed the lowest fracture resistance compared with the other



**Graph 1:** Graphical representation of mean fracture load of immature root of all tested groups

**Table 1:** Mean fracture values in Newton (N) ± standard deviations for all groups

Groups	Number of specimens	Mean forces (N) ± standard deviation
I (positive control)	10	913.26 ± 164.40 <sup>a</sup>
II	10	854.00 ± 174.80 <sup>a</sup>
III	10	886.09 ± 150.98 <sup>a</sup>
IV	10	840.72 ± 148.16 <sup>a</sup>
V	10	868.96 ± 161.91 <sup>a</sup>
VI (MTA negative control)	10	590.28 ± 112.26 <sup>b</sup>
VII (BD negative control)	10	615.47 ± 117.96 <sup>b</sup>

Within the same column, groups with the same letters are not statistically significant ( $p < 0.05$ )

groups. The positive control group (group I) had the highest fracture resistance and differed significantly ( $p < 0.05$ ) from the negative control groups. For the experimental groups, the mean load to fracture ranged from 840.72 to 886.09 N with no significant difference between either of groups II to V ( $p > 0.05$ ), suggesting a comparable reinforcing of the backfilling materials along with either apical barrier used. No significant difference was found between MTA and BD apical barriers ( $p > 0.05$ ) regardless of backfilling material.

## DISCUSSION

Apexification and reinforcement of root canal-treated immature teeth is a highly critical task. Therefore, the selection of a material with a reinforcing effect on the weakened root is mandatory. In preceding studies, the fracture resistance of immature teeth restored with various methodologies, such as MTA, composite, fiber post, and gutta-percha, was tested,<sup>2,14,19</sup> while in the current study, we tested the root-reinforcing effect of BD and MTA as an apical barrier with different obturation combination in comparison with the fracture resistance of intact immature rat.

The site of maxillary central incisor teeth explains its high susceptibility to trauma and external impacts; thus, in the current study, they were selected for testing. In testing the fracture resistance in the current study, a jig that allowed placement of the samples in the universal testing device at an angle of 45° was used to simulate the average angle of contact between maxillary and mandibular incisors in class I occlusion.<sup>20,21</sup>

Mineral trioxide aggregate was assessed in the present study as an artificial apical barrier due to its adaptation, low microleakage, pH 12.5 when set, adequate radiopacity, amenability to hard tissue deposition over its surface, and favorable clinical outcomes.<sup>22</sup> In addition, MTA may indirectly affect the inhibition of dentin metalloproteinase, possibly preventing degradation of the dentin collagen matrix,<sup>23</sup> and it may bond with root dentin that

is initially mechanical and later becomes chemical.<sup>24,25</sup> The selection of the use of 4 mm MTA apical barrier in the current study was due to the best sealing ability compared with thinner applications in agreement with Valois and Costa.<sup>26</sup> However, MTA has some obstacles.<sup>7-9</sup> Therefore, BD was selected to assess in the present study as an alternative apical barrier.

Biodentine cement is of the same class as MTA. It is biocompatible, capable of inducing the apposition of reactionary dentin by stimulating odontoblast activity and reparative dentin by induction of cell differentiation. Its consistency is better suited to clinical use than that of MTA, its presentation ensures a better handling and safety than that of MTA, and faster setting and lower risk of bacterial contamination than with MTA.<sup>27,28</sup>

In the present study, the selection of gutta-percha with two different sealer materials combining with either MTA or BD apical barrier was based on the following: Metaseal (hybrid root seal) is a recent generation of methacrylate materials. It is a self-adhesive, hydrophilic dual-cure sealer eliminating the necessity of a separate priming step. It seals the root canal with a 4-methacryloxyethyl trimellitic anhydride (4-META) hybrid layer that bonds to dentin and a hydrophobic radical that bonds to the solid filling material. It uses the beneficial properties of methacrylate to reduce leakage and provide a reinforcement to the tooth.<sup>29-31</sup>

AH26 is an epoxy resin-based sealer with high polymerization time and creep capacity that may enable better penetration into the dentinal tubules which, in turn, facilitate the interlocking between sealer and dentin and promote more adhesion and higher resistance to sealer dislodgement from the dentin surface. In addition, the formation of covalent bonds by an open epoxide ring of that sealer to any exposed amino groups in the collagen of dentin and its high-quality properties including very low shrinkage while setting and long-term dimensional stability may enhance the root fracture resistance.<sup>32-34</sup>

The results of the current study indicated that the negative control group with no backfilling exhibited a significantly lower fracture resistance value compared with the experimental groups. All the materials tested had a reinforcing effect to the weakened structure to some degree. In the current study, the immature unfilled group (negative control) was weaker than the mature intact group (positive control), and this confirmed that any substantial loss or any modification of natural root canal geometry would affect the tooth strength, in agreement with Lang et al.<sup>35</sup> The importance of placement of filling material to substitute the lost part of the tooth was thus reassured. In all experimental groups, there was no statistically significant difference between them or with the positive control intact teeth. This might be a reflection that the filling compensated the reduced strength from

the lost dentin substance. This was in agreement with El-Ashry et al.<sup>36</sup>

According to the results of the current study, BD showed comparable results with MTA when used with the two selected obturation combination so it is considered an alternative to the use of MTA as an apical barrier in the treatment of immature teeth. In previous studies, it was reported that the use of MTA has a reinforcing effect to immature root fracture.<sup>23</sup> The observed statistical increase in the fracture resistance of BD and MTA in comparison to the negative control group could be due to the hydroxyapatite-like layer that formed between the dentin and the tricalcium silicate materials,<sup>37</sup> which suggests the formation of good bonding in agreement with El-Ma'aita et al.<sup>38</sup>

The present *in vitro* model had some limitations; it cannot reliably simulate clinical conditions, especially in regard to tooth fracture as a result of a traumatic injury and the tissue composition or physical characteristics of the natural immature teeth and the simulated immature teeth.<sup>39</sup> Further clinical research should be conducted to validate the results.

## CONCLUSION

Within the limitations of this study, it can be concluded that all the tested obturating combinations exerted some degree of reinforcement to simulated immature teeth with either apical MTA or BD barrier. This methodology may be recommended for immature teeth that require a higher degree of reinforcement owing to their excessively thin and weakened dentinal walls.

## REFERENCES

1. Cvek M. Prognosis of luxated non-vital maxillary incisors treated with calcium hydroxide and filled with gutta-percha. A retrospective clinical study. *Endod Dent Traumatol* 1992 Apr;8(2):45-55.
2. Wilkinson K, Beeson T, Kirkpatrick T. Fracture resistance of simulated immature teeth filled with Resilon, gutta percha, or composite. *J Endod* 2007 Apr;33(4):480-483.
3. Al Ansary M, Day P, Duggal M, Brunton P. Interventions for treating traumatized necrotic immature permanent anterior teeth: inducing a calcific barrier and root strengthening. *Dent Traumatol* 2009 Aug;25(4):367-379.
4. Felipe W, Felipe M, Rocha M. The effect of mineral trioxide aggregate on the apexification and periapical healing of teeth with incomplete root formation. *Int Endod J* 2006 Jan;39(1):2-9.
5. Ghose L, Baghdady V, Hikmat B. Apexification of immature apices of pulpless permanent anterior teeth with calcium hydroxide. *J Endod* 1987 Jun;13(6):285-290.
6. Andreasen J, Farik B, Munksgaard E. Long-term calcium hydroxide as a root canal dressing may increase risk of root fracture. *Dent Traumatol* 2002 Jun;18(3):134-137.
7. Torabinejad M, Smith P, Kettering J, Pitt Ford T. Comparative investigation of marginal adaptation of mineral trioxide aggregate and other commonly used root-end filling materials. *J Endod* 1995 Jun;21(6):295-299.
8. Torabinejad M, Chivian N. Clinical applications of mineral trioxide aggregate. *J Endod* 1999 Mar;25(3):197-205.
9. Boutsoukis C, Noula G, Lambrianidis T. Ex vivo study of the efficiency of two techniques for the removal of mineral trioxide aggregate used as a root canal filling material. *J Endod* 2008 Oct;34(10):1239-1242.
10. Gomes-Filho J, Rodrigues G, Watanabe S, Estrada Bernabé PF, Lodi CS, Gomes AC, Faria MD, Domingos Dos Santos A, Silos Moraes JC. Evaluation of the tissue reaction to fast endodontic cement (CER) and Angelus MTA. *J Endod* 2009 Oct;35(10):1377-1380.
11. Camilleri J. Characterization and hydration kinetics of tricalcium silicate cement for use as a dental biomaterial. *Dent Mater* 2011 Aug;27(8):836-844.
12. Grecha L, Mallia B, Camilleri J. Investigation of the physical properties of tricalcium silicate cement-based root-end filling materials. *Dent Mater* 2013 Feb;29(2):e20-e28.
13. Laurent P, Camps J, About I. Biodentine™ induces TGF-β1 release from human pulp cells and early dental pulp mineralization. *Int Endod J* 2012 May;45(5):439-448.
14. Schmoldt S, Kirkpatrick T, Rutledge R, Yaccino J. Reinforcement of simulated immature roots restored with composite resin, mineral trioxide aggregate, gutta-percha, or a fiber post after thermocycling. *J Endod* 2011 Oct;37(10):1390-1393.
15. Hemalatha H, Sandeep M, Kulkarni S, Yakub SS. Evaluation of fracture resistance in simulated immature teeth using Resilon and Ribbond as root reinforcements an *in vitro* study. *Dent Traumatol* 2009 Aug;25(4):433-438.
16. Seto B, Chung K, Johnson J, Paranjpe A. Fracture resistance of simulated immature maxillary anterior teeth restored with fiber posts and composite to varying depths. *Dent Traumatol* 2013 Oct;29(5):394-398.
17. Stefopoulos S, Tsatsas D, Kerezoudis N, Eliades G. Comparative *in vitro* study of the sealing efficiency of white vs grey ProRoot mineral trioxide aggregate formulas as apical barriers. *Dent Traumatol* 2008 Apr;24(2):207-213.
18. Melo M, Valle A, Pereira J, Bonachela W, Pegoraro L, Bonfante G. Evaluation of fracture resistance of endodontically treated teeth restored with prefabricated posts and composites with varying quantities of remaining coronal tooth structure. *J Appl Oral Sci* 2005 Jun;13(2):141-146.
19. Tuna E, Dincol M, Gencay K, Aktoren O. Fracture resistance of immature teeth filled with BioAggregate, mineral trioxide aggregate and calcium hydroxide. *Dent Traumatol* 2011 Jun;27(3):174-178.
20. Pene J, Nicholls J, Harrington G. Evaluation of fiber-composite laminate in the restoration of immature, non-vital maxillary central incisors. *J Endod* 2001 Jan;27(1):18-22.
21. Stuart C, Schwartz S, Beeson T. Reinforcement of immature roots with a new resin filling material. *J Endod* 2006 Apr;32(4):350-353.
22. Alobaid A, Cortes L, Lo J, Nguyen T, Albert J, Abu Melha AS, Lin LM, Gibbs JL. Radiographic and clinical outcomes of the treatment of immature permanent teeth by revascularization or apexification: a pilot retrospective cohort study. *J Endod* 2014 Aug;40(8):1063-1070.
23. KHatibovic-ofman S, Raimundo L, Zheng L, Chong L, Friedman M, Andreasen J. Fracture resistance and histological findings of immature teeth treated with mineral trioxide aggregate. *Dent Traumatol* 2008 Jun;24(3):272-276.

24. Sarkar NK, Caicedo R, Ritwik P, Moiseyeva R, Kawashima I. Physicochemical basis of the biologic properties of mineral trioxide aggregate. *J Endod* 2005 Feb;31(2):97-100.
25. Cauwels R, Pieters I, Martens L, Verbeeck R. Fracture resistance and reinforcement of immature roots with gutta percha, mineral trioxide aggregate and calcium phosphate bone cement: a standardized *in vitro* model. *Dent Traumatol* 2010 Apr;26(2):137-142.
26. Valois C, Costa ED Jr. Influence of the thickness of mineral trioxide aggregate on sealing ability of root-end fillings *in vitro*. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2004 Jan;97(1):108-111.
27. Wang X, Sun H, Chang J. Characterization of  $\text{Ca}_3\text{SiO}_5/\text{CaCl}_2$  composite cement for dental application. *Dent Mater* 2008 Jan;24(1):74-82.
28. Colon P, Bronnec F, Grosgeat B, Pradelle-Plasse N. Interactions between a calcium silicate cement (Biodentine) and its environment. *J Dent Res* 2010;89:Abstract no. 401.
29. Chang J, Hurst T, Hart D, Estey A. 4-META use in dentistry: a literature review. *J Prosthet Dent* 2002 Feb;87(2):216-224.
30. Pinna L, Brackett M, Lockwood PE, Huffman BP, Mai S, Cotti E, Dettori C, Pashley DH, Tay FR. *In vitro* cytotoxicity evaluation of a self-adhesive, methacrylate resin-based root canal sealer. *J Endod* 2008 Sep;34(9):1085-1088.
31. Yamanaka Y, Shigetani Y, Yoshida K, Yoshida N, Okiji T. Immunohistochemical analysis of subcutaneous tissue reactions to methacrylate resin-based root canal sealers. *Int Endod J* 2011 Jul;44(7):669-675.
32. Teixeira F, Teixeira E, Thompson J, Trope M. Fracture resistance of roots endodontically treated with a new resin filling material. *J Am Dent Assoc* 2004 May;135(5):646-652.
33. Cobankara F, Adanir N, Belli S. Evaluation of the influence of smear layer on the apical and coronal sealing ability of two sealers. *J Endod* 2004 Jun;30(6):406-409.
34. Versiani M, Carvalho-Junior J, Padilha M, Lacey S, Pascon E, Sousa-Neto M. A comparative study of physicochemical properties of AH Plus and Epiphany root canal sealants. *Int Endod J* 2006 Jun;39(6):464-471.
35. Lang H, Korkmaz Y, Schneider K, Raab W. Impact of endodontic treatments on the rigidity of the root. *J Dent Res* 2006 Apr;85(4):364-368.
36. El-Ashry B, M. Eid G, Ibraheem D. Fracture resistance of immature roots obturated with three different filling materials. *J Am Sci* 2012;8(5)617-624.
37. Han L, Okiji T. Bioactivity evaluation of three calcium silicate-based endodontic materials. *Int Endod J* 2013 Sep;46(9):808-814.
38. El-Ma'aita A, Qualtrough A, Watts D. Resistance to vertical fracture of MTA-filled roots. *Dent Traumatol* 2014 Feb;30(1):36-42.
39. Tanalp J, Dikbas I, Malkondu O, Ersev H, Gungor T, Bayirli G. Comparison of the fracture resistance of simulated immature permanent teeth using various canal filling materials and fiber posts. *Dent Traumatol* 2012 Dec;28(6):457-464.